

Multiple-Host Sharing, Long-Term Persistence, and Virulence of *Escherichia coli* Clones from Human and Animal Household Members[∇]

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During a 3-year surveillance, six household members (five humans and the family dog) yielded 14 *Escherichia coli* clones. Virulence genes, group B2, and having caused cystitis (in the mother or dog) corresponded to colonization endpoints (number of samples, colonies, hosts, and dates). The dog’s cystitis clone was the most extensively recovered clone.

Extraintestinal pathogenic *Escherichia coli* (ExPEC) causes most episodes of urinary tract infection (UTI) in humans, dogs, and cats (9, 25, 26, 32). In *E. coli* UTI, the immediate source of the causative strain is usually the host’s own fecal flora (4, 9, 23, 30). Household members, including pets, commonly share *E. coli* clones, suggesting within-household transmission (2, 3, 5, 6, 13, 24). Although pets sometimes carry ExPEC clones that cause UTI in human household members (6, 13, 24), the reverse scenario has not been reported.

We investigated this phenomenon in relation to an acute

UTI episode involving the pet dog of a household that 2 years previously had been studied around the mother’s acute cystitis episode (6). We analyzed fecal *E. coli* collected from household members at the time of the dog’s UTI episode and 1 and 11 months later and then compared newly and previously recovered clones.

Subjects and methods. The subjects studied included six cohabiting individuals: a mother (age 53 years), a father (age 52 years), two daughters (both age 15 years), a son (age 13 years), and a dog (6). Fecal samples were submitted concurrent

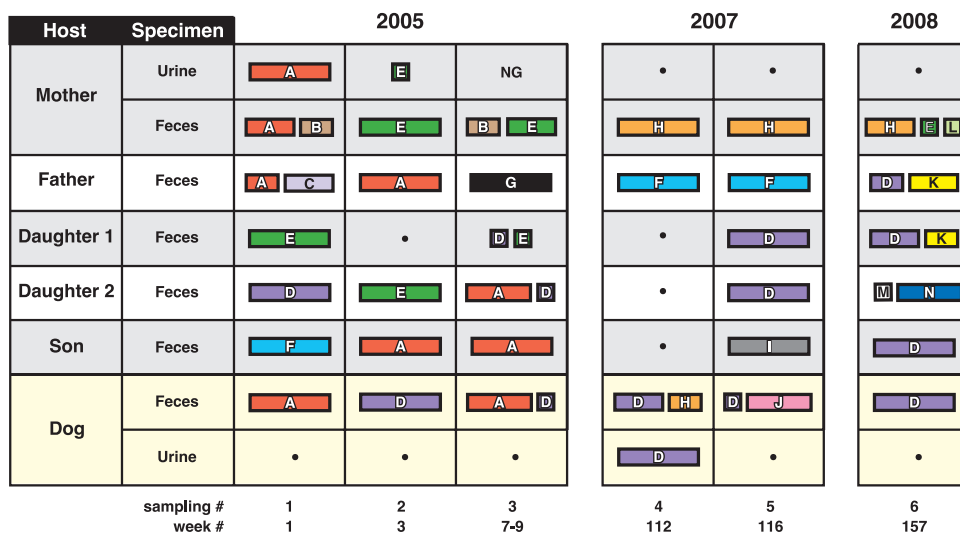


FIG. 1. Distribution of *E. coli* clones by host over time among five human household members and their pet dog. Each color represents a different unique clone (a total of 14 clones, arbitrarily designated A to L). The width of each rectangle is proportional to the number of colonies accounted for by the particular clone among the total number of colonies analyzed from the particular sample (usually, five colonies per sample). Sequential sampling number (#) and week number, counting from the mother’s initial UTI episode, are shown below the chart. Rectangles with heavy black borders indicate symptomatic UTI episodes (mother, sampling 1; dog, sampling 4). Results for samplings 1 to 3 were reported previously (6). The daughters were redesignated here according to age (1 = older, 2 = younger, the reverse of the previously used designations [6]). Gray and white background = humans; buff background = dog.

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TABLE 1. Distribution of 14 unique *E. coli* clones among six household members over six sampling points (3 years)

Host	Sampling point(s) ^a at which clone was encountered in indicated host													
	<u>A</u> ^b	B	C	<u>D</u> ^b	<u>E</u> ^b	<u>F</u> ^b	G	<u>H</u> ^b	I	J	<u>K</u> ^b	L	M	N
Mother	1	1, 3			2, 3, 6			4, 5, 6					6	
Father	1, 2		1	6		4, 5	3				6			
Daughter 1 ^c				3, 5, 6	1, 3						6			
Daughter 2 ^c	3			1, 3, 5	2								6	6
Son	2, 3			6		1			5					
Dog	1, 3			2, 3, 4, 5, 6				4		5				

^a Sampling points are numbered 1 (initial, week 1), 2 (week 3), 3 (week 7 or 9), 4 (week 112), 5 (week 115), and 6 (week 157). Results for sampling points 1, 2, and 3 were reported previously (6). Daughter 1 was not sampled at point 2, whereas at point 3 she was sampled at week 9 (versus week 7 for all other subjects). None of the three children was sampled at point 4.

^b Clones encountered in multiple hosts (shared clones) are underlined. Clones A and D caused acute UTI in the mother (sampling point 1) and dog (sampling point 4), respectively.

^c The daughters were redesignated here according to age (1 = older, 2 = younger, the reverse of the previously used designations [6]).

with the dog’s UTI episode (dog, mother, and father) and again 1 and 11 months later (six subjects). The dog’s acute urine sample was collected by the family’s veterinary clinic in a sterile container during spontaneous voiding. Procedures for recovering five *E. coli* colonies (as available) per sample were as described previously (6), except that MacConkey agar was used instead of eosin-methylene blue agar. Unique clones were resolved by random amplified polymorphic DNA analysis and XbaI pulsed-field gel electrophoresis (6). Sequence types (STs) were newly defined by seven-gene multilocus sequence typing

(<http://web.mpiib-berlin.mpg.de/mlst/dbs/Ecoli/>). The *E. coli* phylogenetic group, 12 UTI-associated O types, and 55 ExPEC virulence factor genes were determined by PCR (1, 7, 16, 18). The virulence score was the number of virulence genes detected.

Shared clones were considered those recovered concurrently or sequentially from multiple hosts. Colonization was considered the presence of a clone in feces. Long-term persistence was considered the presence of a clone in samples separated by ≥1 year. Colonization behaviors included a clone’s cumulative

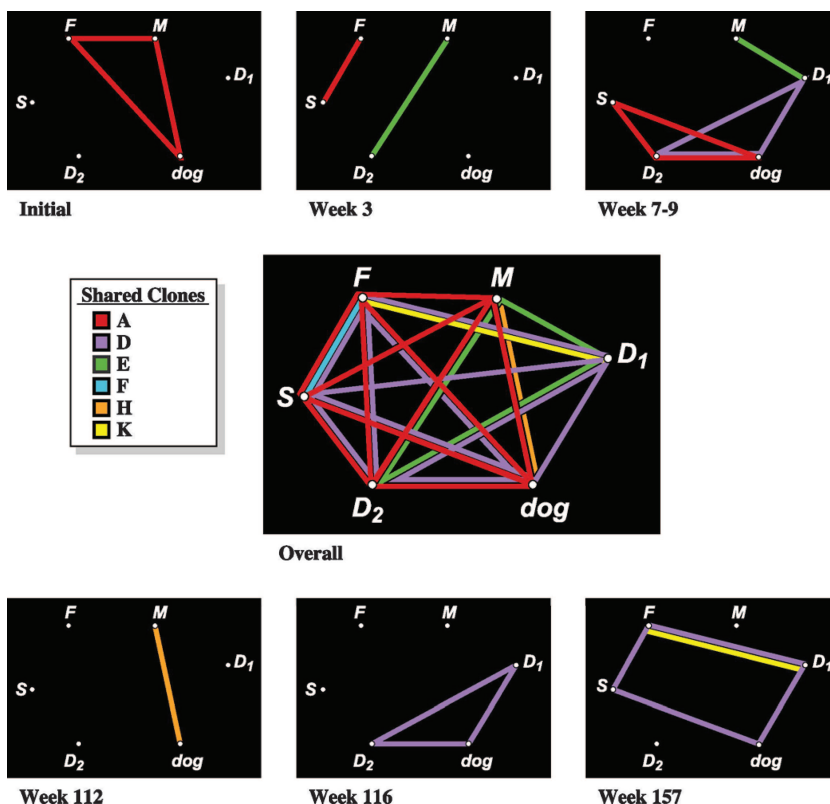


FIG. 2. Clone-sharing relationships among six household members at each of six sampling points and overall. A colored line indicates detection of the corresponding clone (see the color code key) in both individuals connected by the line, either concurrently at the same sampling point (top and bottom small boxes for sampling points 1 to 6) or at any time during the study (center large box). F, father; M, mother; S, son; D₁, daughter 1; D₂, daughter 2. Results for samplings 1 to 3 were reported previously (6). Daughters were redesignated here according to age (1 = older, 2 = younger, the reverse of the previously used designations [6]).

TABLE 3. Colonization behaviors in relation to UTI status and other bacterial traits among 14 *E. coli* clones recovered from five human members and a canine member of a household over 3 years^a

Trait ^b (no. of clones with trait)	Median no. of samples/ clone (range)		<i>P</i> value	Median no. of colonies/ clone (range)		<i>P</i> value	Median no. of hosts/ clone (range)		<i>P</i> value	Median no. of time points/clone (range)		<i>P</i> value
	Trait present	Trait absent		Trait present	Trait absent		Trait present	Trait absent		Trait present	Trait absent	
UTI clone (2)	11.5 (9–14)	1 (1–7)	0.01	42.5 (38–47)	4.5 (1–21)	0.02	5 (5–5)	1 (1–3)	0.01	4.5 (3–6)	1 (1–4)	0.04
Group B2 (4)	6 (1–14)	1 (1–7)	0.09	26.5 (5–47)	4 (1–21)	0.03	3.5 (1–5)	1 (1–3)		3 (1–6)	1 (1–4)	
<i>pic</i> (2)	11.5 (9–14)	1 (1–7)	0.01	42.5 (38–47)	4.5 (1–21)	0.02	5 (5–5)	1 (1–3)	0.01	4.5 (3–6)	1 (1–4)	0.04
<i>usp</i> (4)	6 (1–14)	1 (1–7)	0.09	26.5 (5–47)	4 (1–21)	0.03	3.5 (1–5)	1 (1–3)		3 (1–6)	1 (1–4)	
<i>clbB</i> (3)	9 (3–14)	1 (1–7)	0.01	38 (15–47)	4 (1–21)	0.01	5 (2–5)	1 (1–3)	0.01	3 (3–6)	1 (1–4)	0.02
<i>clbN</i> (2)	8.5 (3–14)	1 (1–9)	0.066	31 (15–47)	4.5 (1–38)	0.088	3.5 (2–5)	1 (1–5)	0.099	4.5 (3–6)	1 (1–4)	0.04
<i>malX</i> (6)	2.5 (1–14)	1 (1–7)		10 (5–47)	4 (1–21)	0.03	2 (1–5)	1 (1–3)		2 (1–6)	1 (1–4)	

^a Samples, total of number of samples that yielded the clone at any time during the study; colonies, total number of colonies accounted for by the clone during the study; hosts, total number of hosts that yielded the clone at any time during the study; time points, number of sampling points at which the clone was encountered in one or more hosts.

^b The traits shown are those that yielded $P < 0.05$ by the Mann-Whitney U test (exact, two tailed) for an association with one or more of the colonization behavior endpoints. P values of < 0.10 are shown. UTI clone, urine isolate from mother's or dog's acute UTI episode. Group B2, phylogenetic group B2; *pic*, protein involved in colonization (serine protease); *usp*, uropathogenicity-specific protein; *clbB* and *clbN*, colibactin synthesis; *malX*, pathogenicity island marker. Additional traits that yielded only borderline associations ($0.10 > P > 0.05$) with one or more of the colonization behaviors included *sfa/foc* (S and F1C fimbriae; $P = 0.07$ for dates, samples, and colonies) and the *H7 flhC* allele (flagellin; $P = 0.099$ for hosts, $P = 0.088$ for samples). No assessed trait yielded a borderline or significant negative association with any of the colonization behavior endpoints.

members, with most host pairs sharing multiple clones. Third, the two UTI clones were encountered much more frequently and extensively as asymptomatic intestinal colonizers than as uropathogens, calling into question what triggered the UTI episodes. Fourth, four clones exhibited prolonged persistence (1 to 3 years) within the household, being isolated either continuously or intermittently from the same or different hosts over the interval. Finally, group B2, virulence gene content, and UTI clone status exhibited borderline or significant associations with multiple colonization endpoints, suggesting that colonization and transmission fitness may contribute to UTI pathogenesis, whereas virulence-associated traits may promote colonization and transmission.

Documentation of UTI in a companion animal caused by an *E. coli* strain concurrently present in a human household contact is, to our knowledge, novel. This represents an interesting counterpoint to previously documented UTI episodes caused in humans by *E. coli* strains that colonized the family's dog or cat (6, 13, 24). These findings suggest that UTI may sometimes be a zoonosis in either direction (human to pet or pet to human). In this regard, certain virulent *E. coli* serotypes or STs are commonly encountered among both humans and companion animals (8, 12, 14, 15, 17, 21, 31, 32). The two putatively zoonotic clones identified here represent two such STs, i.e., ST73 (clone D) and ST95 (clone A).

The extremely prolonged (1- to 3-year) within-household persistence demonstrated for clones D, E, F, and H suggests that identification of external sources for colonizing ExPEC clones may prove challenging, given the possibly great separation in time between acquisition and detection. Notably, the apparent persistence of clone D in certain hosts (the dog and both daughters) over 2 to 3 years is among the longest reported colonizations of any host(s) with the same *E. coli* clone (27–29).

Study limitations include the small numbers, five-colony typing approach (which detects mainly high-prevalence clones), long intervals between certain sampling points, absence of information regarding host exposures and interactions, and lack of environmental sampling. Strengths include the participation

of all household members, the protracted study period, the capture of two acute UTI episodes, extensive molecular typing, and a sample size sufficient for statistical analysis.

In summary, we observed long-term intestinal colonization of multiple human household members and the family dog with an *E. coli* clone that caused acute UTI in the dog. We also documented other dog-human strain sharing, long-term persistence and possible (nonsexual) host-to-host transmission of additional clones, and statistical associations among virulence traits, UTI occurrence, and colonization endpoints. These findings provide added evidence that the household functions as an integrated microbiological unit, demonstrate long-term persistence potential for certain ExPEC clones within individuals and households, support a possible link between virulence and colonization or transmission, and imply that, in some instances, UTI may represent a zoonosis in either direction.

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