GENOME ANNOUNCEMENTS

Draft Genome Sequences of Six *Escherichia coli* Isolates from the Stepwise Model of Emergence of *Escherichia coli* O157:H7[▽]

L. V. Rump, ^{1,4} E. A. Strain, ² G. Cao, ³ M. W. Allard, ¹ M. Fischer, ⁴ E. W. Brown, ¹ and N. Gonzalez-Escalona ^{1*}

Division of Microbiology¹ and Biostatistics Branch,² Center for Food Safety and Applied Nutrition, Food and Drug Administration, 5100 Paint Branch Parkway, College Park, Maryland 20740; Joint Institute for Food Safety and Applied Nutrition (JIFSAN), University of Maryland, College Park, Maryland 20742³; and Institute of Food Chemistry, University of Hamburg, Hamburg, Germany⁴

Received 25 January 2011/Accepted 31 January 2011

Enterohemorrhagic *Escherichia coli* (EHEC) of serotype O157:H7 has been implicated in food-borne illnesses worldwide. An evolutionary model was proposed in which the highly pathogenic EHEC O157:H7 serotype arose from its ancestor, enteropathogenic *E. coli* (EPEC) O55:H7 (sorbitol fermenting [SOR $^+$] and β-glucuronidase positive [GUD $^+$]), through sequential gain of virulence, phenotypic traits, and serotype change. Here we report six draft genomes of strains belonging to this evolutionary model: two EPEC O55:H7 (SOR $^+$ GUD $^+$) strains, two nonmotile EHEC O157:H $^-$ strains (SOR $^+$ GUD $^+$) containing plasmid pSFO157, one EHEC O157:H7 (SOR $^-$ GUD $^+$) strain, and one O157:H7 strain containing plasmid pSFO157 (SOR $^+$ GUD $^+$).

Enterohemorrhagic Escherichia coli (EHEC) O157:H7 has become a significant worldwide cause of food-borne illness since its discovery about 20 years ago. It frequently causes large outbreaks of severe enteric infections, including bloody diarrhea, hemorrhagic colitis (HC), and hemolytic-uremic syndrome (HUS) (1, 4). This serotype expresses the somatic (O) 157 and flagellar (H) 7 antigens, so these traits are extensively used in clinical analysis to identify this highly pathogenic serotype (1). A stepwise evolutionary model has been proposed in which the highly pathogenic enterohemorrhagic E. coli (EHEC) serotype O157:H7 arose from its ancestor, enteropathogenic E. coli (EPEC) O55:H7, belonging to clonal complex (CC) A1/A2 (sorbitol fermenting and β-glucuronidase positive [SOR+ GUD+]), through sequential acquisition of virulence and phenotypic traits and serotype change (2, 3, 9). After the somatic antigen change from O55 to O157 gave rise to a probably extinct intermediary (CC A3), two separate O157 CCs evolved, splitting into two diverging clonal groups. One was composed of sorbitol-fermenting (SF) nonmotile O157:H⁻ strains containing plasmid pSFO157 (CC A4) (SOR+ GUD+). The other was composed of non-sorbitolfermenting (NSF) O157:H7 strains containing plasmid pO157 (CC A5) (SOR⁻ GUD⁺). The latter by a mutational inactivation of the uidA gene lost its β -glucuronidase activity, which is the most typical O157:H7 phenotype at present (CC A6) (3). These CC A6 strains expanded and spread geographically and account for most of the diseases caused by EHEC (10).

So far, only four complete genome sequences for pathogenic *E. coli* O157:H7 belonging to the CC A6 have been reported and deposited in GenBank: Sakai (NC_002695) (5), EDL933 (AE005174) (8), TW14359 (CP001368) (7), and EC4115 (NC_011353). Recently, the genome of an ancestral O55:H7 strain, CB9615 (NC_013941), was made available (11). In the present publication, we announce the availability of six draft genome sequences for other enteropathogenic *E. coli* strains belonging to the stepwise model of emergence of *E. coli* O157:H7 (2). The strains sequenced are EPEC O55:H7 3256-97 (CC A2) and USDA 5905 (CC A2), O157:H7 LSU-61 (CC A unknown), EHEC O157:H⁻ 493/89 (CC A4) and H2687 (CC A4), and EHEC O157:H7 G5101 (CC A5) (3).

Genomic DNA from each strain was isolated from overnight cultures with a DNeasy blood and tissue kit (Qiagen). The genomes were sequenced by 454 Titanium pyrosequencing (Roche), according to the manufacturer's instructions at 20× coverage. Genomic sequence contigs for strains 3256-97 and USDA 5905 were assembled with the 454 Life Sciences Newbler software package version 2.3 (Roche) using the complete *E. coli* O55:H7 strain CB9615 genome (see above for accession number) as a reference. Genomic sequence contigs for strains LSU-61, 493/89, H2687, and G5101, were assembled with the complete *E. coli* O157:H7 strain Sakai genome as a reference. Sequences were annotated with the NCBI Prokaryotic Genomes Automatic Annotation Pipeline (http://www.ncbi.nlm.nih.gov/genomes/static/Pipeline.html) (6).

A detailed report of a full comparative analysis between the genomes of these six isolates will be included in a future publication.

Nucleotide sequence accession numbers. The draft genome sequences for these six *E. coli* strains are available in GenBank under accession no. AEUB00000000, AEUA000000000,

^{*} Corresponding author. Mailing address: FDA, CFSAN, 5100 Paint Branch Parkway HFS-712, College Park, MD 20740. Phone: (301) 436-1937. Fax: (301) 436-2644. E-mail: narjol.gonzalez-escalona@fda.hhs.gov.

[∇] Published ahead of print on 11 February 2011.

AETY000000000. AETZ00000000, AEUC0000000, and AETX000000000.

This project was supported by an appointment to the Research Fellowship Program for the Center for Food Safety and Applied Nutrition administered by the Oak Ridge Associated Universities through a contract with the FDA.

REFERENCES

- 1. Feng, P. 1995. Escherichia coli serotype O157:H7: novel vehicles of infection
- and emergence of phenotypic variants. Emerg. Infect. Dis. 1:47–52.

 2. Feng, P., K. A. Lampel, H. Karch, and T. S. Whittam. 1998. Genotypic and phenotypic changes in the emergence of *Escherichia coli* O157:H7. J. Infect. Dis. 177:1750–1753.
- 3. Feng, P. C., et al. 2007. Genetic diversity among clonal lineages within Escherichia coli O157:H7 stepwise evolutionary model. Emerg. Infect. Dis. 13:1701-1706.
- 4. Griffin, P. M., and R. V. Tauxe. 1991. The epidemiology of infections caused

- by Escherichia coli O157:H7, other enterohemorrhagic E. coli, and the associated hemolytic uremic syndrome. Epidemiol. Rev. 13:60-98.
- 5. Hayashi, T., et al. 2001. Complete genome sequence of enterohemorrhagic Escherichia coli O157:H7 and genomic comparison with a laboratory strain K-12. DNA Res. 8:11-22.
- 6. Klimke, W., et al. 2009. The National Center for Biotechnology Information's Protein Clusters Database. Nucleic Acids Res. 37:D216-D223.
- 7. Kulasekara, B. R., et al. 2009. Analysis of the genome of the Escherichia coli O157:H7 2006 spinach-associated outbreak isolate indicates candidate genes that may enhance virulence. Infect. Immun. 77:3713-3721.
- 8. Perna, N. T., et al. 2001. Genome sequence of enterohaemorrhagic Escherichia coli O157:H7. Nature 409:529-533.
- 9. Whittam, T. S., et al. 1993. Clonal relationships among Escherichia coli strains that cause hemorrhagic colitis and infantile diarrhea. Infect. Immun. **61:**1619-1629.
- 10. Wick, L. M., W. Qi, D. W. Lacher, and T. S. Whittam. 2005. Evolution of genomic content in the stepwise emergence of Escherichia coli O157:H7. J. Bacteriol. 187:1783-1791.
- 11. Zhou, Z., et al. 2010. Derivation of Escherichia coli O157:H7 from its O55:H7 precursor. PLoS One 5:e8700.