Supplemental Figure S1. Newly identified Prophages.

The gene organization of the three newly identified prophages is shown. For comparison, the three prophages are shown with similar known phages or prophages. Protein-coding sequences (CDSs) are colored according to their assigned functional categories, as shown in the box. For each phage pair, homologous CDSs have the same number. The amino acid sequence identities between homologous CDSs are shown below the CDSs in the reference phages. The integration sites of the three prophages are also shown. A) Prophage Sp7 of O157 Sakai and a prophage (PPO157_s3) identified in segment 167/168 of strain #3. B) The CPZ-55 prophage of K-12 (GenBank accession number U00096) and a prophage or prophage-like integrative element (PPO157_s2) identified in segment 256/257 of strain #2. C) Phage P4 (GenBank accession number X51522) and a P4-like prophage (PPO157_s4) identified in segment 448/448.1 of strain #4.

Supplemental Figure S2. Curlin production in the nine O157 strains.

Each O157 strain was aerobically grown in Luria-Bertani (LB) medium overnight at 37˚C, and 20 µl of each overnight culture was inoculated into 1 ml of fresh LB medium. After incubation for 3 hrs at 37˚C, the cultures were diluted with fresh LB medium, spread onto “Congo red indicator plates” containing 10 g/liter Casamino acids (NIHON PHARMACEUTICAL CO., LTD., Tokyo, Japan), 1 g/liter yeast extract (Difco Laboratories, Sparks, MD, USA), 15 g/liter agar (KYOKUTO PHARMACEUTICAL INDUSTRIAL CO., LTD, Tokyo, Japan), 20 mg/liter Congo red

and 10 mg/liter Coomassie brilliant blue (Sigma Aldorich Co., St..Louis, MO, USA), and incubated at 28˚C for 48 hrs to detect curlin production as described by Hammar, M., et al. (Proc. Natl. Acad. Sci. USA, 93: 6562-6566, 1996). Because single base changes in the *csgD* promoter region induce variable expression of the curlin biosynthesis genes and the frequency of the base change is not high in O157 strains (Uhlich G. A., et al., Appl. Environ. Microbiol., **67:** 2367-2370, 2001), we analyzed 1,000-15,000 colonies on the Congo red indicator plates for each of the nine O157 strains used in this study. From O157 strains Sakai, #3, #6, #7, #8, and #9, we isolated Congo red binding-positive colonies that produce curlin, although the frequencies of positive colonies varied among these O157 strains. However, in strains #2, #4, and #5, all of which contain IS insertions in either of the two curlin biosynthesis operons, we observed no Congo red binding-positive colonies although we repeated this screening many times. Note that the plates of strains Sakai, #6, and #8 contain both Congo red binding-positive (indicated by black arrow heads) and negative colonies.