

Q&A ATCC® Excellence in Research Webinar “Food Safety: Improving Detection of non-O157 Shiga Toxin-Producing *Escherichia coli* (STEC)”

General Questions

1. Will we be able to download the presentation?

This presentation will be available to watch on demand [here](#).

2. Why can't ATCC ship “Big Six” purified strains overseas?

Although ATCC confirms that each DNA preparation does not contain any viable starting organism, the Commerce Control List includes and restricts any genetic elements or nucleic acid that contains the sequences coding for the toxins. DNA from the non-toxigenic strains is available for international distribution.

3. How does ATCC specifically determine the serotype of a strain of *E. coli*?

We analyze both the O and H antigens using antisera from various commercial sources. O typing can be performed directly on the regular growth (after boiling). To test for H type, the isolates were passed through motility media a few times to enrich for cells with flagella.

4. Did ATCC test these non-O157 STEC strains on any of the commercial molecular tests?

We confirmed the identity of the strains and the presence of Shiga toxin and intimin genes. We have not tested their performance in any commercial molecular tests.

5. Is it possible to determine the serogroups using chromogenic agar, or is this more of a confirmatory assay?

Rainbow agar is a selective medium and its technical instructions for use indicate that each serogroup has a unique color or shade. This is the result of the combination of two different enzymes on colorimetric substrates. Since many non-O157 toxigenic strains overproduce β -galactosidase relative to β -glucuronidase on this medium, isolates appear purple, violet, or blue. However, you should consider alternative, approved methods for verification of serogroup.

6. Did ATCC determine the relative levels of Shiga toxin produced by these isolates?

We only confirmed the presence of the toxin in culture supernatant using an enzyme immunoassay.

7. Did ATCC determine the *eae* variant subtypes?

No. We only performed a confirmatory PCR test for *eae*. We have not sequenced the *eae* gene to determine (*eae* (gamma), *eae* (beta), *eae* (theta), *eae* (epsilon) variants. Eventually, the genome sequence of these strains will be available for such an analysis.

8. Does ATCC offer non-O157 STEC testing to external entities?

ATCC does not perform product testing. There are many reference labs available to perform such testing if you are unable to perform the testing internally.

9. Who makes the rainbow agar?

Biolog makes rainbow agar. You can find more information about this product on their website, <http://www.biolog.com/agar/>.

10. Are the non-toxicogenic “Big Six” strains available for world-wide distribution?

Yes, the non-toxicogenic “Big Six” strains are available to all countries except those restricted by the U.S. government.

11. Can the non-toxicogenic “Big Six” strains be used as positive controls in the routine screening of STEC strains in meat products, regardless of the detection system used?

Non-toxicogenic strains are suitable for assays using antibody-based detection of the O-antigen. They are not suitable for molecular tests or antibody tests that target the Shiga toxin.

12. What are the ATCC numbers of the non-toxicogenic STEC “Big Six” strains?

Currently, ATCC has nine available non-toxicogenic STEC “Big Six” strains. These can be found under the following catalog numbers: ATCC® BAA-2212™, 12795™, 23982™, BAA-2214™, 29552™, 33780™, 43887™, BAA-2216™, and BAA-2190™.