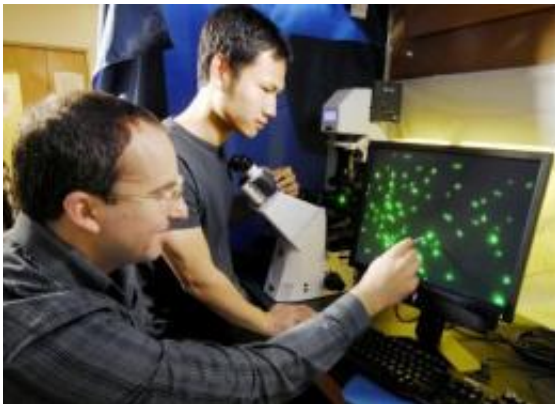


Study provides new way to classify *E. coli* bacteria and test for fecal contamination

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Georgia Tech civil and environmental engineering assistant professor Kostas Konstantinidis (left) and biology graduate student Chengwei Luo examine environmental and clinical *E. coli* cells, which cannot be differentiated with microscopy techniques. Credit: Georgia Tech/Gary Meek

The meaning of the standard fecal coliform test used to monitor water quality has been called into question by a new study that identified sources of *Escherichia coli* bacteria that might not indicate an environmental hazard.

Fecal pollution of surface waters is measured by the concentration of *E. coli* bacteria in the water because *E. coli* is believed to live only in the intestines and waste of humans and other warm-blooded animals, and quickly die outside its host. The presence of *E. coli* in water also serves

as a marker for other potentially more harmful organisms that may accompany it. Positive *E. coli* tests may lead to the summertime closing of beaches and other recreational bodies of water.

In this new study, researchers report identifying and sequencing the genomes of nine strains of *E. coli* that have adapted to living in the environment independent of warm-blooded hosts. These strains are indistinguishable from typical *E. coli* based on traditional tests and yield a positive fecal coliform result though researchers say they may not represent a true environmental hazard.

"The basis for *E. coli*'s widespread use as a fecal pollution indicator is the traditional thinking that *E. coli* cannot survive for extended periods outside a host or waste, but this study indicates that's not true," said Kostas Konstantinidis, an assistant professor in the Georgia Tech School of Civil and Environmental Engineering. "These results suggest the need to develop a new culture-independent, genome-based coliform test so that the non-hazardous environmental types of *E. coli* are not counted as fecal contamination."

A paper describing the research was published April 11 in the early edition of the journal [Proceedings of the National Academy of Sciences](#). The work was sponsored by the National Science Foundation and the National Institutes of Health.

Konstantinidis and Georgia Tech School of Biology graduate student Chengwei Luo compared the genomes of 25 different strains of *E. coli* and close relatives, which were sequenced by the Center for Microbial Ecology at Michigan State University, the Broad Institute in Massachusetts, or were publicly available in the National Center for Biotechnology Information database. Nine strains that were recovered primarily from environmental sources encoded all genes required for classification as *E. coli*.

"The orders-of-magnitude higher abundances of the group of organisms represented by these nine strains in environmental samples relative to those in human feces and the clinic indicate that they represent truly environmentally adapted organisms that are not associated primarily with mammal hosts," explained Konstantinidis, who also holds a joint appointment in the Georgia Tech School of Biology.

By comparing the full genomes of the samples, the Georgia Tech researchers identified 84 genes specific to or highly enriched in the genomes of the environmental *E. coli* and 120 genes specific to the strains commonly found in the gastrointestinal tract of healthy humans, which are called commensal *E. coli*. They also detected recent genetic exchange of core genes within the environmental *E. coli* and within the commensal strains, but not from commensal genomes to their environmental counterparts.

The environment-specific bacteria included genes important for resource acquisition and survival in the environment, such as the genes required to utilize energy sources and to break down dead cellular material. In contrast, the gastrointestinal *E. coli* included several genes involved in the transport and use of nutrients thought to be abundant in the gut.

"The genomic data suggest that the environmental *E. coli* are better at surviving in the external environment, but are less effective competitors in the gastrointestinal tract than commensal *E. coli*, which tells us that the environmental bacteria are highly unlikely to represent a risk to public health," explained Konstantinidis.

Collectively, this data also indicates that the environmental *E. coli* strains represent a distinct species from their commensal *E. coli* counterparts even though they are identified as *E. coli* based on the standard taxonomic methods. This work is consistent with a more stringent and ecologic definition for bacterial species than the current definition and

suggests ways to start replacing traditional, culture-based approaches for defining diagnostic phenotypes of new species with genomic-based procedures.

The scientific, medical, regulatory and legal communities expect species to reasonably reflect the traits and habitat of an organism -- especially an organism like *E. coli* that has ramifications for diagnostic microbiology and for assessing fecal pollution of natural ecosystems. Efforts toward a more refined definition of this bacterial species are needed, according to Konstantinidis.

This study's findings provide a way to start redefining *E. coli* species and testing for [fecal contamination](#) with procedures based on genomics and ecology.

"We are now working to develop a molecular assay that uses the gastrointestinal-specific genes as robust biomarkers to count commensal *E. coli* cells in environmental samples more accurately than current methods," added Konstantinidis.

Provided by Georgia Institute of Technology

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