

Study reveals intestinal bacteria succession during recovery from cholera in Bangladesh

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A new study delineates a sequential pattern of changes in the intestinal microbial population of patients recovering from cholera in Bangladesh, findings that may point to ways of speeding recovery from the dangerous diarrheal disease. The report also finds what appear to be consistent differences between the gut microbial population - also called the microbiota - of individuals in developed countries like the U.S. and those the developing world and provides some of the most complete evidence that the gut microbiota usually return to normal after cholera infection. Their paper appears in the online journal *mBio*.

"Healthy gut bacterial communities are known to benefit immune regulation, metabolism and potentially even the nervous system, so if cholera or other diarrheal diseases permanently impact the microbiota, there could be long-term effects on human health," explains Regina LaRocque, MD, MPH, of the Massachusetts General Hospital (MGH) Division of Infectious Diseases, co-senior author of the paper. "Also by studying how the gut microbiota usually recover after cholera, we gain new ideas about how to manage severely disturbed microbial communities, ideas that can be used to protect against diseases that often follow such disturbance, such as infections that commonly follow antibiotic use."

The investigators analyzed stool samples from two groups of cholera patients in Dhaka, Bangladesh. One group of 13 patients had received antibiotic treatment the day before the first sample was taken; in the other group of 10 patients, samples were taken right before they



received treatment. Control samples were taken from 27 uninfected household members of the first patient group. Follow-up samples from both patients and the controls were taken 7 and 30 days after initial sampling. For comparison, similar samples were taken from a group of 18 patients hospitalized for diarrhea caused by a pathogenic *E. coli* infection.

To get a complete picture of all bacterial species in samples, the researchers used metagenomic gene sequencing, an approach for sequencing organisms taken from nature rather than laboratory cultures. The results revealed four distinct bacterial groups, depending on the stage of infection and recovery. At the pretreatment/infection stage, the population was dominated by the pathogenic Vibrio cholerae bacteria, as would be expected. The early stage - 1 to 7 days after treatment - saw the growth of opportunistic species not usually found in a healthy gut and often tolerant of an oxygen-rich environment that would be deadly to other species. The mid-stage population that began appearing after day 7 featured oxygen-intolerant bacterial species often seen in American individuals but much less frequently in developing nations. By day 30 the late-stage population was similar to what was seen in the uninfected controls as well as in healthy populations throughout the developing world. Result for the patients infected with E. coli showed similar patterns of succession; but in addition to being dominated by E. coli at the infection stage, late-stage species began appearing as early as day 7 in some patients.

"The results for the mid-stage cholera group, which included bacteria usually less abundant in the developing world, were probably the most interesting," says LaRocque, an assistant professor of Medicine at Harvard Medical School. "We don't completely understand why those species occur during cholera infection, but that phenomenon may offer clues as to why we observe different species of gut bacteria among humans in different parts of the world. While one theory holds that the



differences reflect species range - that maybe some species simply don't exist in some countries - our data finding microbes typical of the <u>developing world</u> at the mid-stage in Bangladeshi patients argue against that hypothesis."

The researchers also investigated different mechanisms that could be behind the changing microbial populations and found evidence of the action of four ecological processes: ingestion of bacteria on food, the presence of abnormal oxygen levels in the gut, the availability of carbohydrates and other nutrients, and the presence of bacteria-infecting phage viruses.

"The data suggesting that the gut may become more oxygenated than normal at the early stage of recovery implies that treatment designed to reduce gut oxygen levels might help normal, anaerobic bacteria return to the gut sooner," says lead author, Lawrence David, PhD. "Among other factors we will be investigating is whether an individual's microbiota affects the risk of contracting cholera, information that could help us better understand risk factors for the disease." Formerly with the Center for Systems Biology at Harvard University, David is now an assistant professor of Molecular Genetics and Microbiology at Duke University.

Provided by Massachusetts General Hospital

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