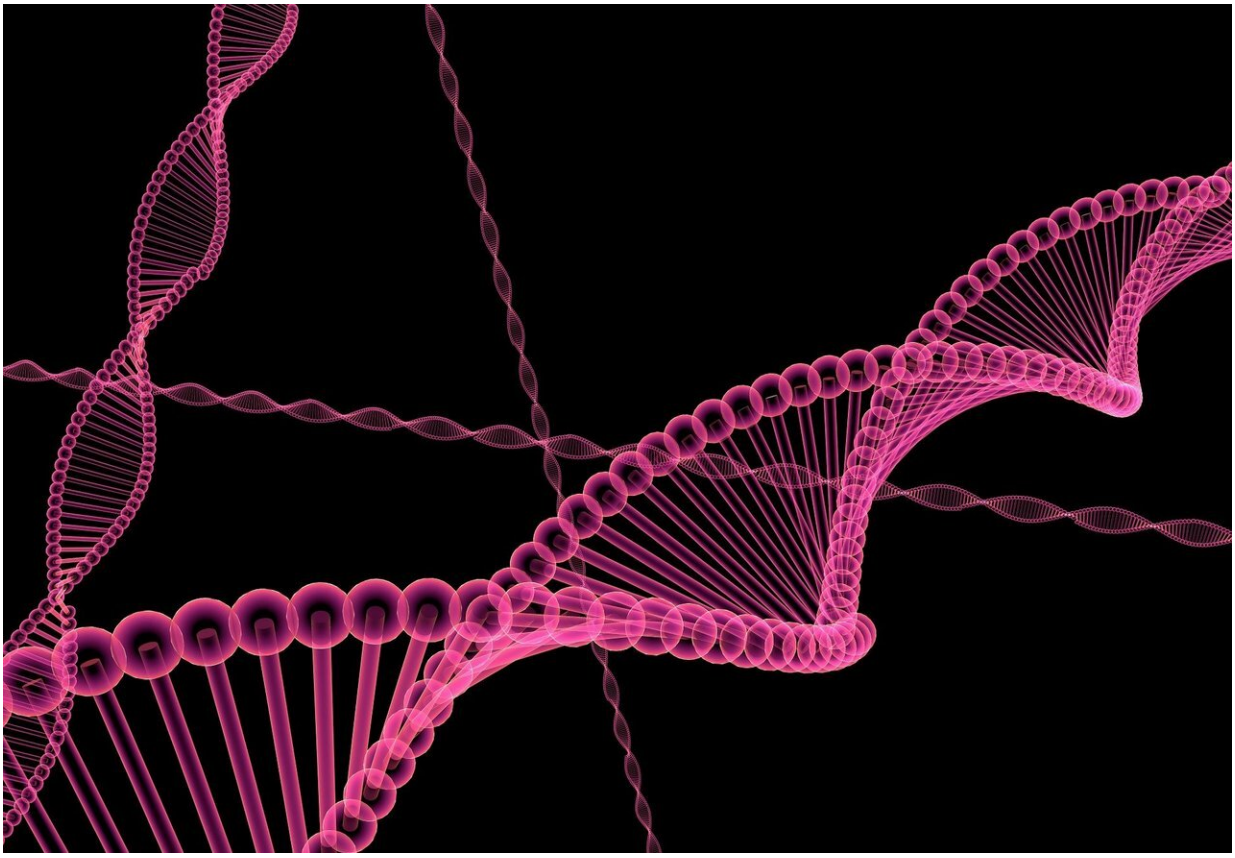
DNA metabarcoding useful for analyzing human diet

October 8 2019



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A new study demonstrates that DNA metabarcoding provides a promising new method for tracking human plant intake, suggesting that similar approaches could be used to characterize the animal and fungal

components of human diets. The study, published in the journal *mSystems*, demonstrated that dietary plant DNA can be amplified and sequenced from human stool using methods commonly applied to wildlife studies.

"DNA sequencing has given us a large amount of new data on things like microbiology in the gut and personal genetics. This study suggests that the same powerful technology could also start telling us about what we eat, which is often a hard thing to measure," said senior study author Lawrence David, Ph.D., assistant professor, Center for Genomic and Computational Biology, Duke Molecular Genetics and Microbiology.

Many preexisting methods for dietary assessment exist, but most rely on a person's ability to report what they ate. This means that they are subject to errors in memory, bias people have in reporting, and the cognitive ability of a person answering a survey. DNA metabarcoding is an alternative way to obtain dietary information that uses food DNA in stool as a biomarker. Researchers can amplify food DNA from a fecal sample, sequence it, and map those sequences back to foods using a reference database. "I think of DNA metabarcoding very much like a barcode at a supermarket. We can think of a particular DNA sequence as a unique identifier for a particular food species," said second study author Brianna Petrone, a graduate student at Duke University School of Medicine.

Dr. David and co-first author Aspen Reese, Ph.D., now a junior fellow at Harvard University, launched the study after they met the ecologists Rob Pringle, Ph.D., at Princeton University and Tyler Kartzinel, Ph.D., now at Brown University, who originally used DNA metabarcoding to study complex food webs of herbivores in the African savanna. "We wondered whether their method would work in people," said Dr. David. "There is a growing body of work in the microbiome field indicating that specific foods are likely to be altering or shaping levels of specific bacteria in the

gut, but we often don't have accompanying diet data for the microbiome studies."

To conduct their study, the researchers pulled DNA out of cold storage that had been extracted from stool samples from a previous study. "We happened to do a study a couple years ago where we were preparing foods for participants in a microbiome diet intervention, and we knew exactly what they were eating in a given week when their stool was being collected," said Dr. David.

The researchers sequenced a barcode region from chloroplast DNA in stool samples from 11 individuals consuming both controlled and freely selected diets. They successfully amplified plant DNA in roughly 50% of samples, which increased to 70% in samples from individuals eating a controlled plant-rich diet. The majority of sequenced plant DNA matched common human food plants, including grains, vegetables, fruits and herbs. "Overall, there was good broad agreement between the foods that were listed in the diaries kept by the study participants and the ones that we sequenced from stool," said Dr. David. "If a food was written in the diet record, about 80% of the time, we also found it by this metabarcoding approach."

The relatively high PCR failure rate and inability to distinguish some dietary plants at the sequence level suggest the potential for future refinements to improve the method. For example, cabbage, broccoli, Brussel sprouts and kohlrabi are all cultivars of the same species, and researchers were unable to tell them apart by their sequence in the chloroplast barcode region. Coffee was the only food recorded in the diet that was never detected with DNA metabarcoding, perhaps because its DNA was deteriorated or diluted by roasting and brewing.

Dr. David forsees DNA metabarcoding being used in future studies, as well as renewed the possibility of diet analysis in older studies. "Similar

to this study, I could imagine this getting used on archived DNA to see whether or not there are underlying dietary differences that might explain some of the microbiome patterns that may have been observed in a study," said Dr. David. "Going forward, we can also imagine this being used in new microbiome studies to identify relationships between specific foods and gut bacteria, as well as in broader studies of nutrition as a complement to traditional diet assessment techniques."

Provided by American Society for Microbiology