

EXPLOITING ANAEROBIC CONSORTIA AS NEW TOOLS FOR BIOMASS BREAKDOWN AND SUSTAINABLE CHEMISTRY

Michelle A. O'Malley, Department of Chemical Engineering, UC-Santa Barbara
momalley@ucsb.edu

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Anaerobic microbes work together in complex communities that decompose and recycle carbon biomass throughout the Earth. Compared to microbes that thrive in the presence of oxygen, anaerobic consortia remain understudied and recalcitrant to culture. However, they are a vast, untapped resource for novel enzymes and strains that degrade woody biomass into sugars for value-added chemical production. Here, we performed several enrichment experiments to isolate biomass-degrading consortia from goat feces, and identify microbes that drive the activity and stability of these cultures. Fecal samples were challenged by four types of biomass (alfalfa, bagasse, xylan, and reed canary grass) and two types of antibiotic treatments (chloramphenicol, penicillin-streptomycin) during cultivation to identify important cross-domain partnerships; 10 billion metagenomic reads spread across 402 enrichment samples tracked biological diversity as the cultures converged to a minimal set of ~20 microorganisms that were stable after more than ten culture generations. Nearly 200,000 carbohydrate active enzymes (CAZyme) domains were identified from the fecal samples alone, constituting nearly 25% of the known CAZymes in existence. 724 genomes were assembled for previously uncultured novel microbes within the herbivore rumen. Surprisingly, consortia dominated by anaerobic fungi generated more than twice the amount of methane compared to prokaryotic consortia, suggesting that fungi play a key role in methane release in ruminant herbivores. The most active microbial consortia comprise cross-domain partnerships between anaerobic fungi from the genus *Neocallimastix* and *Piromyces*, methanogenic archaea from the genus *Methanobrevibacter*, and bacteria from the phylum *Firmicutes*, some of which were enriched nearly 20-fold from the fecal microbiome, produce high yields of methane off-gas, and are capable of cryopreservation and revival. New routes for metabolic cooperation between enriched consortia were also identified, suggesting that an array of bacteria support biomass-degrading microbes by providing essential amino acids while consuming deleterious byproducts. Overall, our analysis points to natural compartmentalization between anaerobes as a means to degrade crude biomass, which can be exploited to harness nature's microbes for sustainable chemical production.