Cometabolic stimulation of anaerobic coal bioconversion using a phenanthrene-degrading enrichment

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The biological conversion of coal to methane typically involves syntrophic consortia that decompose coal macromolecules to monomers and subsequently ferment the monomers to methanogenic substrates. However, due to structural heterogeneity and recalcitrance of coal matrices, coal biodegradation is very slow, especially under anoxic conditions.

We investigated anaerobic cometabolic degradation of coal by a marine phenanthrene (PNT)-degrading enrichment. This enrichment couples PNT oxidation to sulfate reduction and activates PNT by carboxylation [1]. Incubations were performed anoxically (80% N₂: 20% CO₂) at 35^oC in the dark and used ground Pust lignite (Montana, USA, <250-µm particle size). Incubations with lignite + PNT showed $\sim 2x$ faster sulfate reduction and ~1.5x higher microbial diversity, compared to those with PNT only. However, without PNT added, sulfate reduction was minimal. Additionally, lignite + PNT treatments had distinct microbial communities. Bacteria were dominantly polymer, amino acid, fatty acid, and sugar utilizing heterotrophs (e.g. Desulfobacterium, Melioribacter, Syntrophus). Archaea were dominantly from methylotrophic and acetoclastic methanogen clades (e.g. Methanosaeta, Methanosphaera, Methanolobus). During incubation, lignite particles clumped together and adhered to PNT particles, consistent with biofilm-like material around PNT particles observed in SEM images. Potential coal-related metabolites (e.g. dimethylbenzenethiol and mercaptosuccinic acid) only detected in lignite + PNT treatments indicated that some fraction of lignite was degraded or solubilized.

Polycyclic aromatic hydrocarbon (PAH) degraders typically cometabolize a wide range of PAHs [2,3,4]. Hence, it is possible that enzymes released by our PNT-degrading inoculum fortuitously degraded the aromatic fraction of lignite. Our results suggest that a variety of enrichments that degrade coal model compounds (e.g., PAHs, alkanes) could enhance cometabolic degradation of coal by providing compatible enzymatic systems.

[1] Davidova *et al.* (2007) *ISME J.* **1**, 436-42. [2] Chen & Aitken (1999), *ES&T* **33**, 435–439. [3] Annweiler et al. (2001) *Appl Environ Microbiol.* **67(11)**, 5077–5083. [4] Gai et al. (2007) *Appl.Environ.Microbiol.*, **73(9)**, 2832-8.

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