

## Transport of Crude Oil Aggregates and Associated Microbial Populations: Impact on Biodegradation Potential

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**Background/Objectives.** Storm-driven washover events play a critical role in remobilization of MC252 crude oil in coastal headland beaches contaminated by the Macondo oil spill. Oil forms such as surface residue balls (SRBs) and submerged oil mats (SOMs) were mobilized across the beach profile on Fourchon Beach, Louisiana. SOMs and SRBs are sand and oil aggregates that can be remobilized from the intertidal and subtidal environment and moved across the beach profile by tropical storms and strong cold fronts. The objectives of this study are to understand the extent of alkylated dibenzothiophenes and phenanthrenes weathering in different storm-mobilized oil forms and the associated microbial populations within the aggregates. The overall goal is to understand the potential for biodegradation of crude oil components in these dynamic beach systems.

**Approach/Activities.** SOMs and SRBs were collected after multiple washover events on Fourchon Beach, Louisiana between 2011 and 2013. Concentration of alkylated polycyclic aromatic hydrocarbons (PAHs) in SOMs and SRBs were quantified by GC-MS. Alkylated PAH concentrations were used to construct weathering ratio of 3-ring alkylated phenanthrenes and dibenzothiophenes to 4-ring chrysenes. Bacterial populations in the SOMs and SRBs samples were identified by 16S rRNA sequencing using the Illumina Miseq platform. In addition, laboratory microcosms studies exploring PAH biodegradation in SRBs were conducted under aerobic and anaerobic conditions. For each redox condition, treatments included addition of nutrients (N and P), and examining undisturbed and crushed SRB aggregates over a time frame of 150 days.

**Results/Lessons Learned.** SRBs were observed to be more susceptible to PAH weathering than SOMs, presumably due to differences in oxygen availability in the two oil forms. Weathering ratios in SOMs sampled from offshore and onshore were similar to the oil sampled near the wellhead immediately after the spill began, demonstrating poor weathering of 3-ring PAHs. Alkylated dibenzothiophenes were preferentially weathered to alkylated phenanthrenes. Metagenomic analysis of SRBs and SOMs demonstrated that *Gammaproteobacteria* was the dominant population followed by *Alphaproteobacteria*. In the most weathered SRBs, *Acetobacteraceae* belonging to *Alphaproteobacteria* was the most abundant population. In SRBs and SOMs a diverse set of PAH-biodegrading marine organisms were present at high abundance including *Hyphomonas*, *Parvibaculum*, *Porphyrobacter*, *Alcanivorax*, *Altermonas*, *Idiomarina*, *Halothiobacillus*, *Halomonas*, *Marinobacter*, *Oceanospirillaceae*, *Porticoccus*, *Pseudoalteromonas*, *Singularimonas*, *Thiomicrospira*, and *Vibrio*. In the SRB microcosms, biodegradation was observed only in the aerobic microcosm treatment with crushed SRBs and the decreases observed were statistically significant compared to all other treatments ( $P < 0.05$ ). No apparent decrease in PAHs were observed in anaerobic and aerobic microcosms with intact SRBs both in the presence and absence of added nutrients. Mobilization of these aggregates during storms serves to move the oil components and the associated microbial populations to different locations across the beach profile. These processes provide important controls on the rate and extent of biodegradation of crude oil components after spills.