FINAL STUDY REPORT

HUNTSMAN STUDY NUMBER:	HMSC-169
SPONSOR:	ITOPF
PROJECT NAME:	**P**hotomodification **O**f **L**ow-sulfur-fuel-oils **I**nvestigations of **T**oxic **E**ffects (POLITE).
TEST MATERIAL:	U/VLSFO
LOCATION:	Huntsman Marine Science Centre St. Andrews, NB Canada
Principal Investigator:	Benjamin de Jourdan, PhD. Huntsman Marine Science Centre St. Andrews, NB Canada
Co-Principal Investigator:	Christoph Aeppli, PhD Bigelow Laboratory for Ocean Sciences East Boothbay, ME USA
EXPERIMENTAL INITIATION DATE:	May 1st, 2022
REPORTING PERIOD DATE:	May 1st 2022 – May 1st, 2024
PROJECT COMPLETION DATE:	May 1st, 2024

Executive Summary

The following report outlines the activities from the entire project that began May 1st 2022 and ended May 1st, 2024. During this project we completed work that addressed aspects of all three of our goals;

1. How toxic are U/VLS fuel oils compared to conventional fuel oils?
2. How does photomodification change toxicity?
3. Can we effectively predict the toxicity photomodified and non-photomodified U/VLS fuel oils?

In this study we conducted 31 distinct toxicity bioassays (Figure 1) with American lobster (*Homarus americanus*), Atlantic cod (*Gadus morhua*), and green sea urchin (*Strongylocentrotus droebachiensis*) using a conventional heavy crude oil (CONV), an offshore Newfoundland crude oil (ESRF), an ultra-low sulfur fuel oil (ULSFO), and 14 very low sulfur fuel oils (VLSFO) provided by the Australian Maritime Safety Authority (AMSA). The bioassays were conducted with exposure media generated from paired water accommodated fractions (WAFs) of the oil and seawater, with one WAF prepared in the dark (e.g., standard preparation) and one prepared under a UV light (e.g., irradiated) to induce photomodification.

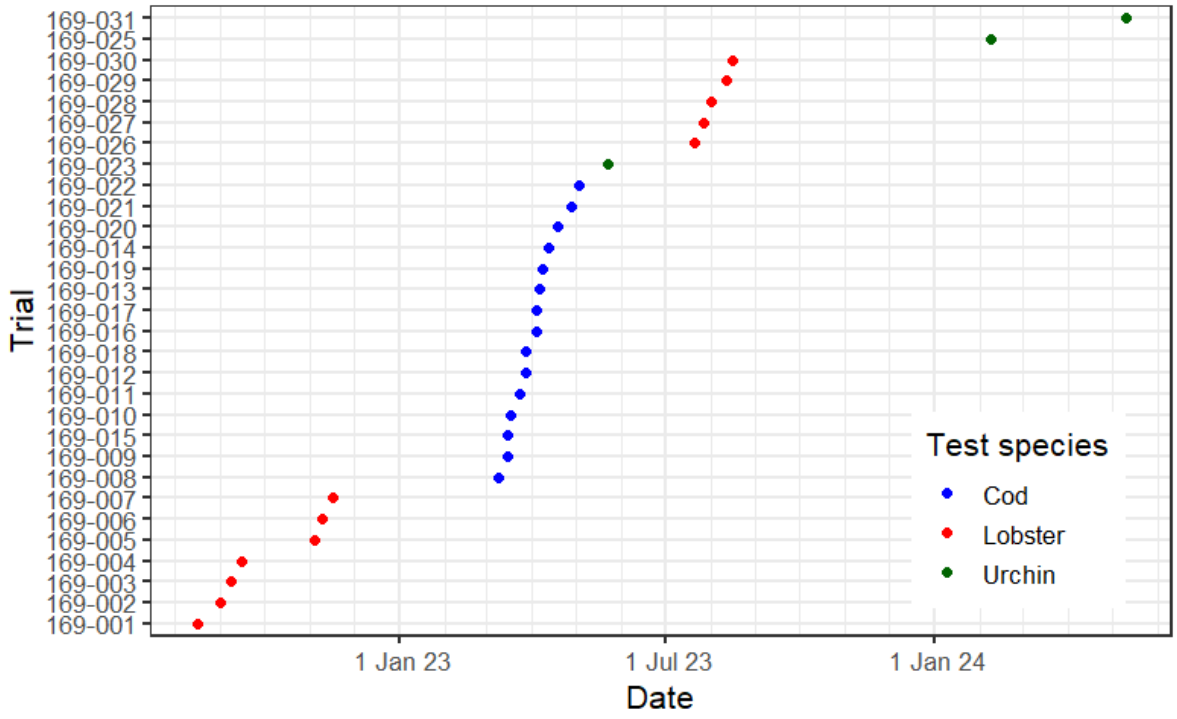

Figure 1: Timeline of the bioassays conducted over the course of this study.

Following irradiation, the exposure metrics (e.g., fluorometry units (RFU), total organic carbon (TOC), biomimetic extraction solid phase microextraction (BE-SPME), and polycyclic aromatic compounds (PACs)) for many of the products tested significantly increased, suggesting that there were photoproducts being formed. The amount of photoproducts formed varied between oil samples, with some of the tested VLSFOs not showing any detectable changes following irradiation. In nearly all cases the observed toxicity in the UV treated WAF was equal or greater than the WAF prepared in the dark. Larval lobster immobilization was the most sensitive endpoint across the tested species, and it showed a full range of responses with the tested products varying from 10 - 100% immobilization (Figure 2).

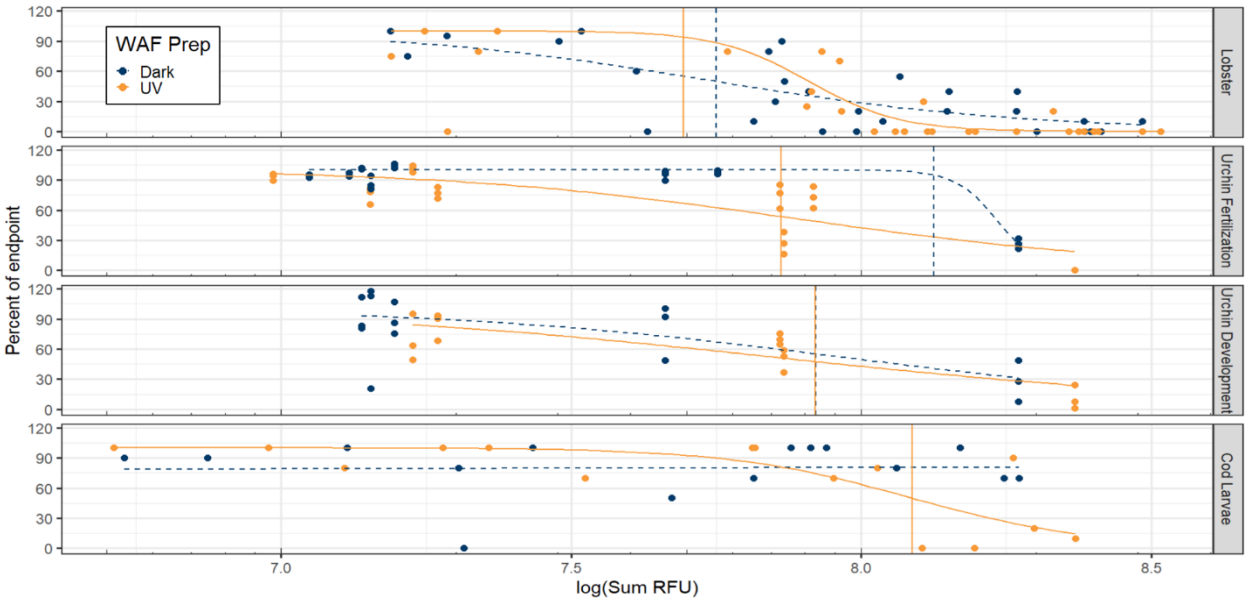

Figure 2: Normalized responses showing lobster immobilization (top), urchin fertilization and development (middle) and cod mortality (bottom) following exposure to irradiated (solid orange circles) and non-irradiated (solid blue circles) WAFs. The species are ordered by sensitivity top to bottom, with the vertical lines showing the EC50 on the basis of fluorometry units

The immobilization response largely followed a concentration gradient with TOC, BE-SPME, and PAC based toxic units. However, using only waterborne PAC concentrations to calculate toxic units, the predicted toxicity was consistently lower than what was observed for many of the VLSFOs products, both with and without UV irradiation. These results suggest that there are other aspects that aren't measured by traditional GC-MS (e.g., oxidized products) which are contributing to the toxicity of the VLSFOs.

The results highlight a wide range of responses across fuel types, with significant differences in sensitivity across species. The impact of UV light on the observed toxicity underscores the importance of addressing and incorporating modifying factors when

determining the toxicity of complex mixtures. The SARA fraction (Saturates, Aromatics, Resins, and Asphaltenes), specifically the aromatics and asphaltenes, have significant correlations with the formation of photoproducts and their associated toxicity.

The data generated within this study will be used to develop and validate models, to predict and assess the toxicity of these new generation fuel oils.