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## Short-Term Immune Response of Largemouth Bass (Micropterus salmoides) to Crude Oil Exposure

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In North America, crude oil is transported through pipelines and by rail in ever increasing amounts. Often, the railways and pipelines follow or cross major rivers. Furthermore, in 2019, approximately 1,000 tons of oil were spilled into water systems throughout the world, the largest collective tonnage occurring in North America. But the impacts of oil spills in freshwater systems, particularly sub-lethal effects on fish and other aquatic organisms, are significantly understudied. Therefore, to fundamentally improve our understanding of how oil spills affect freshwater fish, we designed and conducted an experiment to develop an immune response timeline for largemouth bass exposed to crude oil. The goal was to evaluate largemouth bass response at three specific time points: 2, 7, and 14 days postexposure. Largemouth bass were exposed through intraperitoneal injections of a 10% crude oil solution or dimethyl sulfoxide alone as a control. Fish were held in flow-through tanks and euthanized at sampling. Immune responses were evaluated by differential white blood cell composition in peripheral blood and macrophage aggregates in the spleen histology. The observed changes in immune status included a shift in white blood cell composition from greater numbers of lymphocytes and fewer numbers of granulocytes and monocytes (normal, healthy status) to a state of decreasing lymphocyte numbers and increasing numbers of granulocytes and monocytes. In addition, we observed increases in the numbers of macrophage aggregates present in the spleens of exposed bass over the observation period after exposure. Changes in both endpoints occurred at 2 days post exposure and continued until 14 days post exposure. These changes in peripheral blood cell composition and macrophage aggregates indicate a change in immune status in response to crude oil exposure. Moreover, these results clarify the temporal changes that occur in largemouth bass immune responses after an oil spill.