Development of a Complete Life Cycle Sediment Toxicity Test for the Sheepshead Minnow (*Cyprinodon variegatus*)

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Existing sediment toxicity test methods are limited to acute and chronic exposure of invertebrates and acute exposure of vertebrates, with limited guidance on the chronic exposure of vertebrates, specifically fishes. A series of life stage-specific studies were conducted to determine optimal test conditions for exposing the sheepshead minnow (Cyprinodon variegatus) to contaminated sediment throughout its complete life cycle. Specifically, these methods will be used to expose fish to oil-contaminated sediment collected from Barataria Bay, LA. Reference sediment used in these studies was collected from a marsh habitat in Choctawhatchee Bay, FL and had similar grain size characteristics as Barataria Bay. The hatching success of the egg stage was tested in well plates of various sizes (6, 12, 24 wells) with various amounts of sediment under stagnant and agitated conditions. Test chambers for larvae, juvenile and adult stages were developed to accommodate dissolved oxygen and fish density demands, minimize ammonia accumulation, and represent realistic estuarine exposure conditions. Larvae were exposed in static conditions, while studies were conducted to compare static and flow through exposure for juveniles and adults. Comparisons among flow through and static conditions included flow rates and types (pulse, continuous). Studies were also conducted to develop methods to collect spawned eggs from test chambers to determine hatch success of next generation embryos. These test development methods can be used as guidance for future life cycle sediment exposure studies involving fishes.