Executive Summary for Framework for Assessment of Causal Relationships between Early Life Stage Development Anomalies of Clupea pallasi and Cosco Busan Oil (October 2010)

A number of research and damage assessment studies were initiated following the *Cosco Busan* Oil spill (CBOS) in San Francisco Bay on 7 November 2007 culminating in a 2010 laboratory assessment of the effects of non-contaminant stressors. The integrated results from analytical analyses demonstrated a chemical signature of *Cosco Busan* oil (CBO) and also demonstrated that the signature was present in two of the water samples collected adjacent to areas that showed shoreline impact. The water samples were collected in November 2007; subsequently the beaches were cleaned of oil prior to the Pacific herring spawn in January – March 2008. Results from assessments of Pacific herring embryo development at sites potentially impacted by CBO showed that significant developmental abnormalities occurred in eggs spawned in the intertidal zone in 2008, but in the absence of a CBO chemical signature in the eggs. Following those initial assessments, follow-on work was performed to experimentally demonstrate the presence or absence of a CBO signature in developing herring eggs, specifically exposed to CBO, another source of oil (Exxon Valdez Oil Spill - EVOS), urban background petroleum concentrations and controls. Laboratory artifacts and field or laboratory variables were noted during review of data from 2008 field studies and the 2009 experimental study; these were primarily environmental stressors that appeared to be at least as significantly associated with the developmental abnormalities as CBO exposure. In 2010, these environmental variables were evaluated under experimental conditions designed to mimic the 2008 field conditions. This document draws on results from all CB studies to examine causal relationships between the CBOS in 2007 and its potential to exert adverse impact to early development stages in herring. The assessment framework (from Fox 1991; Tillitt et al. 2008) addresses six primary lines of evidence to determine the probable cause(s) of effects observed in 2008. Those lines of evidence are:

- *Probability and Time Order* – Did the oil spill occur prior to the spawning events and was there sufficient ability to identify CBO and to document its presence during the spawn events?
- *Strength of Association* – Is the CBO signature in the oil and water distinct from other sources of petroleum; is it also a distinct signature in eggs known to be exposed to CBO?
- *Specificity* - Are each of the biological responses solely associated with CBO or can other factors result in similar response patterns?
- *Consistency of Association* – Are similar effects observed in the absence of CBO?

- *Predictive Performance* – Are the observed biological responses predictable based on results from the scientific literature?

- *Coherence* – Were CBO chemical signatures observed in 2008? Were the effects predictable based on concentrations of CBO contaminants? Were biological effects observed in 2008 demonstrative of petroleum exposure or to other factors?

This interpretive report uses this causal inference framework to combine scientific data collected between 2007 through 2010 with effects-based literature summaries to arrive at the following conclusions:

- The Pacific herring eggs spawned in 2008 and collected from the intertidal areas characterized as being exposed to CBO (in 2007) were not exposed to CBO during their development. The chemical exposure signature in the developing eggs was consistent with urban background and burned wood or creosote and not with CBO. The developing eggs were instead exposed to urban San Francisco Bay PAH contamination (Peninsula Point and San Rafael) and/or urban Bay PAH contamination augmented by sources enriched with fluoranthene and pyrene that may have come from burned wood or creosote (Keil Cove and Sausalito).

- Experiments conducted with known CBO exposure in 2009 produced a diagnostic chemical signature of CBO which is distinctly different from ANS and urban sources of PAH exposure under either transmitted or blocked UV light and at all concentrations of CBO. The absence of the CBO chemical exposure signature in 2008 means that the developing eggs were not exposed to CBO and as a result were also not exposed to any of the measured or unmeasured chemical components contained in CBO.

- A distinct biological signature was developed during the 2009 and 2010 studies that separates the effects of petroleum contamination from other stressors. The biological signature associated with petroleum related effects is the incidence and severity of pericardial edema. Yolk sac edema incidence was demonstrated to occur with temperature and salinity stresses in the 2010 study at similar rates as observed in 2008. Pericardial edema was not present except in organisms that had extensive body axis defects in both the 2008 and 2010 studies. The presence of yolk sac edema, extreme body axis defects and early mortality in developing eggs observed in 2010 in the absence of petroleum was also observed in natural spawn events of 2008 and 2009. The absence of the petroleum related pericardial edema response combined with the incidence and intensity of yolk

sac edema, extreme body axis defects and early mortality of developing eggs are all consistent with effects being created by factors other than CBO exposure.

This body of data demonstrates that CBO spilled in November 2007 did not create adverse effects on the development of Pacific herring embryos that were spawned three to five months later in the intertidal environment of central San Francisco Bay.