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Coastal Response Research Center

**State-of-Science for Dispersant Use in Arctic Waters
Eco-Toxicity and Sublethal Impacts**

General Statement

The following terminology is used in this document:

- DDO is the abbreviation for Dispersants and Dispersed Oil.
- Unless otherwise noted, the word “dispersant” used in this document will refer to chemical dispersants, not other forms of dispersants such as biological dispersants.
- The term DOR is used to denote the Dispersant to Oil ratio.
- Physically dispersed oil refers to oil that is dispersed by natural (e.g., wave action) or mechanical processes.

The following caveats are important to remember when reading this document:

- The panel acknowledges that the resources accessed for this document were limited to information published (including advance publications) prior to December 31, 2015, and is aware of relevant and impactful references published since that time, that would add value for the topics discussed in this document.
- This document focuses on the toxicity of oil and dispersed oil, because:
 - The most toxic impacts are expected to come from oil and dispersed oil (NRC 2005).
 - Dispersant application targets oil, so the volume of “dispersant alone” in the marine environment is expected to be much less than dispersed oil or oil.
 - Dispersant formulations have changed since the 1970’s. This document reviews current literature on newer formulations that are designed to minimize toxicity.
 - Although trace metals are a component of oil, metals were not included in this document because they have not been implicated as primary drivers of oil toxicity and are not analysed in the majority of oil spill literature.
- This document focusses on species that could be exposed to an oil spill in the marine environment. In this review, Arctic species include those that have exclusively Arctic distributions (e.g., Arctic cod) as well as those whose distribution includes parts of the

Arctic and sub-Arctic (e.g., herring, pink salmon, calanoid copepods) (Duesturloh et al., 2002; George et al., 2009; Moulton et al., 2009; Logerwell et al., 2015).

I. Exposure and Exposure Pathways

A. General Statement:

A pathway of exposure is the direct physical course a chemical or pollutant takes from its source to the organism exposed (U.S. EPA, 2013). There are four basic pathways of exposure: Inhalation, aspiration, ingestion, as well as external contact (adsorption and absorption). There are unique features in the Arctic environment that will influence these basic pathways of exposure (e.g., physical environment, biological characteristics). In addition, the location of the release; the state of oil (chemically and physically dispersed oil vs. undispersed oil); the type of oil; and the degree of weathering, determine the most significant pathway(s). Exposure is also a function of the organism's life history, distribution and behavior.

Adverse effects vary in severity and mechanism and are a function of the exposure pathway, the degree of exposure (concentrations and duration), the inherent toxicity of the stressor (e.g., oil, oil component, dispersant and dispersed oil) to the organism, and the sensitivity of the organism (e.g., species, life stage, individual).

Knowns:

1. It is important to identify as many of the different variables as possible that interact to influence biological effects (i.e., chemical mixture they are exposed to, whether those chemicals are in dissolved or particulate phase, duration and concentration of exposure, pathway of exposure, and timing of exposure relative to life stage/natural history).
2. Oil is a complex chemical mixture with thousands of constituents that have varying toxic effects, which is not unique to the Arctic.
 - Computational models have been developed to help address this complexity (DiToro et al., 2000; French McCay 2002; Redman et al., 2012, Redman and Parkerton, 2015).
3. Measuring Total Petroleum Hydrocarbons (TPH) serves as a general assessment of the level of exposure to whole oil in water; however, measuring Total Polycyclic Aromatic Hydrocarbons (TPAH) will provide a better predictor of toxicity (NRC, 2005). TPH and TPAH concentrations are not suitable for assessing the impacts of physical interactions of oil with biota, nor the effects of metals in oil.
 - Studies that summarize existing aquatic toxicity data on physically and chemically dispersed oil indicate that the polycyclic aromatic hydrocarbons (PAHs) are the petroleum hydrocarbons that contribute the most to chronic toxicity and that the

- likelihood of exposure to PAHs increases with water solubility (NRC, 2003, 2005; Redman et al., 2012; DiToro et. al., 2000; French McCay, 2002). Acute lethality has been associated with low molecular weight compounds (e.g., monoaromatics, naphthalenes). However, PAHs cause acute effects as well (DiToro et. al., 2000; French McCay, 2002). Other petroleum hydrocarbons may contribute to aquatic toxicity (e.g. heterocyclics) (Lee et al., 2015; Barron et al., 1999; Incardona et al., 2012). Some components of dispersants may contribute to toxicity, but most toxicity is expected to arise as a result of exposure to petroleum hydrocarbons (NRC, 2005; Hook and Osborn, 2012; Adams et al., 2014a).
- Though analytic capabilities will vary, a standard suite of 50+ PAH analytes has been recommended for forensic chemistry and toxicity evaluations and has been used in the Deepwater Horizon (DWH) oil spill (NOAA, 2015) and other oil spills (Boehm et al., 1996). Many laboratories do not currently have the capability for measuring this full analyte list.
 - When comparing toxicity data on a TPAH basis, it is important to consider that PAH analyte lists may vary across studies and that the relative PAH composition varies across oils, which complicate comparability of results (French McCay, 2002; Bejarano et al., 2014).
 - Nominal concentrations or loadings provide limited data to support an understanding of toxicity and are not informative (Coelho et al., 2013; Bejarano et al., 2014).
 - TPAH alone can be misleading. The composition of the PAH mixture should be determined for all toxicity tests. The most informative laboratory studies are those that include a measure of exposure, assess the exposure (in the water or air, or at the air-water interface), assess the dose (to the organism), and conduct a complete analysis of the oil sample being tested.
 - Toxicity tests that include analysis of PAHs (including homologs), oxy- nitro- PAHs, mono-aromatics, and heterocyclics, are more informative than those with a more limited analyte list.
 - Chemical detection limits for some standard analytic methods may exceed known toxicity thresholds for some species, including those in the Arctic. Typically, lowering the detection limits makes analyses more costly, but allows for better interpretation of the chemical effects.
4. When conducting toxicity studies with oil, exposure solutions should be chemically characterized and measured because many of these compounds are volatile or poorly soluble.

5. It is important to identify and chemically characterize the fraction of oil that is dissolved versus the fraction in droplets, because this affects bioavailability and exposure (Carls et al., 2008; Carls and Thedinga, 2010; NRC, 2005). There are several methods available for characterizing dissolved and droplet oil fractions (Bennett et al., 1990; Payne and Driskell, 2003; Allan et al., 2012; Wiens, 2013; Letinski et al., 2014).
6. The exposure pathway and biology of the organism need to be considered to characterize exposure (e.g., exposure pathways that involve dissolved constituents of oil, ingestion, inhalation, or contact with whole oil or particulates contaminated with constituents of oil).
7. Dispersants change how oil partitions in the water column. Dispersants alter the exposure concentrations of dissolved components of oil and the size distribution of particulate oil.

Uncertainties:

1. Not all of the constituents in oil or all of the degradation products can be measured. Therefore, it is difficult to determine what other components of whole oil are also drivers of toxicological effects, without further development and validation of methods.
2. While there is some evidence that dispersants alter bioavailability and potential accumulation of PAHs and other oil constituents, relative to physically dispersed oil (Wolfe et al., 1997; Wolfe et al., 1998a; Wolfe et al., 1998b; Wolfe et al., 1998c; Wolfe et al., 1999; Wolfe et al., 2001; Couillard et al., 2005; Mielbrecht et al., 2005), what is uncertain is whether the presence of chemicals from dispersants alters the rates of dissolution or uptake into the organisms.
3. The role of droplets in contributing to toxicity is complicated and poorly understood. Oil constituents in droplets can dissolve into water and thus become bioavailable to aquatic organisms. This dissolution is more limited in physically dispersed systems that are characterized by lower droplet concentrations (Redman & Parkerton, 2015). Droplets can also be ingested, but the relevance of this pathway on toxicity is uncertain (Nortdug et al., 2011; Hansen et al., 2012).

II. Exposure and Exposure Pathways in Arctic Conditions

Knowns:

1. In general, the pelagic and benthic exposure pathways in the Arctic operate similarly to other marine ecosystems. In contrast, the existence of sea ice may create pathways that are unique to Polar Regions. Consequently, in this section, the focus is on these unique sea ice interactions.

2. There are potential exposures to DDO from three different pathways, i.e., via oil dispersed by:
 - Surface application in open water;
 - Surface application in ice-infested waters;
 - Subsea dispersant application;
 - Or some combination of the above.
3. Higher levels of spatial and temporal variability in physical and biological parameters in the Arctic than in other regions might influence exposure and potential effects resulting from dispersed and undispersed oil.
4. Oil and ice together create a unique biophysical environment influencing the primary pathways, which affect the degree of exposure.
 - Undispersed oil can pool under, in, or on top of ice. Pooling in between ice floes, leads, polynyas and breathing holes can change exposure duration, concentration, and pathways for organisms.
 - Under-ice communities are unique in their concentration and composition of species, some of which exist nowhere else. This habitat will be exposed to particulate, floating and dissolved contaminants
 - Under-ice communities are most similar to benthic habitats “living upside down”. Their algal communities are a seed for the water column and a food resource when pelagic production is low. When the algae slough off, those species go into the water column and eventually sink to the sea floor. Some species are ice-obligate, others ice-facultative.
 - There is a unique food web associated with the sea ice; this food web increases the diversity and density of organisms, compared to Arctic open-water and thus the number of species and individuals, which could be exposed to undispersed oil and to a lesser extent dispersed oil (e.g., polar cod, ice seals).
 - Bacteria, benthic larvae, and protozoa live in brine channels. Krill scrape algae off ice. These are potential pathways for toxic effects and incorporation of contaminants into the food web.
 - Ice undergoes seasonal cycles that can affect the fate and transport of contaminants, and may affect exposure. During freeze up, material can be encapsulated in ice (see Physical Transport and Chemical Behavior, and Degradation and Fate documents).

- Marine species tend to aggregate at interfaces where oil can collect.
 - Marine mammals are particularly vulnerable to oil in leads or holes because they breathe large volumes of air at the surface air-water interface (Reed et al., 1994; Zapol 1987), where volatile compounds or dispersed/undispersed oil can be highly concentrated (Buist et al., 2011; Nudds et al., 2013). Oil that concentrates in holes in the ice increases exposure of pinniped species that use that hole to get onto or off the ice (Englehardt, 1983; St. Aubin, 1992; Ainley et al., 2003).
 - Birds may also be susceptible to oil in leads (e.g., feeding, resting).
 - Grounded contaminated ice is a potential pathway for sediment and shoreline contamination.
 - There are sensitive environments with unique species, densities, productivity in the Arctic as in other regions (e.g., the thermal bar west of the Mackenzie River to the Beaufort Sea) (Carmack and MacDonald, 2002; ADNR, 2014).
5. Trophic transfer is not a unique exposure pathway, but in the Arctic, trophic-chain lengths can be shorter and organisms are lipid-rich. Trophic transfer in invertebrates is more efficient at low temperatures because less energy goes into respiration. This has implications for vertebrates feeding on invertebrates (Borgå, et al., 2004).
- Arctic food chains are dependent on invertebrates that metabolize oil inefficiently leading to potentially greater trophic transfer (Rust et al., 2004; Barros et al., 2014). Furthermore, Arctic invertebrate species may metabolize oil constituents more slowly at lower temperatures (Carrasco-Navarro et al. 2015).
 - Aquatic invertebrates generally show higher bioaccumulation of components of oil such as PAHs compared to vertebrates because of slower biotransformation in invertebrates (den Besten et al., 2003; Meador, 2003). More efficient metabolism of oil in vertebrates limits bioaccumulation.
6. The Arctic has extensive shallow shelves compared to most other oceans. The pelagic, benthic (pelagic-benthic coupling) and sea ice communities are tightly connected on shelves. The tightness of the connection varies seasonally and spatially (Dunton et al., 2005).
7. Undispersed oil trapped in ice weathers more slowly and remains persistent in the system in an unweathered state (see Degradation and Fate document). This is also true for oil trapped in deep or shoreline sediments. There is a prolonged and delayed potential for physical fouling and exposure because of the encapsulation of oil in ice.
8. In order to understand effects on Arctic species, the duration and variability of exposure must be considered. There are potentially longer exposure times under the ice.

9. Uptake and metabolism are slowed by lower temperatures, resulting in delayed acute toxicological responses and potentially chronic effects (Chapman and Riddle, 2005). Ultimately, the unique aspects of the Arctic environment (e.g., temperature, UV exposure, and ice) may influence the duration of exposure, dynamics of bioaccumulation, and toxicity that may alter the biological or ecological effects or impacts of a spill.

Uncertainties:

1. Exposures are dependent on fate and transport of the contaminants, however, it is unknown if there are unique transport mechanisms that occur in Arctic environments (e.g., brine channels) that are different from those in other places.
2. The effects of higher trophic web organisms ingesting low trophic web organisms that are not able to metabolize oil, have not been studied. While there have been studies for other ecosystems (Neff, 2002; Wan et al., 2007), Arctic food webs are different and therefore there is potential for exposure to be different. The effects of oil on Arctic food webs are unknown. Because Arctic food chains are lipid-rich, there may be higher rates of bioaccumulation (Meador et al., 1995).
3. On the Arctic's extensive shallow shelves, the pelagic, benthic (pelagic-benthic coupling), and sea ice communities are tightly connected (see above). The consequence of this coupling to exposure pathways is uncertain (CSESP, 2014).
4. Because of variable and changing conditions in the Arctic, the degree of exposure to oil and dispersed oil, as well as relative importance of different exposure pathways may change in the future (e.g., Carrasco-Navarro et al., 2015; Hjorth et al., 2011).
5. Low temperatures and ice are known to influence the exposure pathways, but quantifying these influences is uncertain, which leads to uncertainties about the duration and variability of exposure.
6. The effects of low temperature on chemical processes and the resulting biological effects of oil exposure have not been extensively studied and are uncertain (Chapman and Riddle, 2005).

III. Toxicity of Oil and Dispersed Oil to Arctic Species

A. General Statement:

Arctic species can be exposed to oil and oil chemicals from floating and dispersed oil in open water, on shore, and in nearshore and offshore sediments. These threats may result from physical contact with the oil or from toxicity of accumulated constituents.

Knowns:

1. The addition of dispersants to floating oil removes some of the oil from the surface and reduces the exposure of surface-dwelling species (Wolfe et al., 2001; Ramachandran et al., 2004; Carls et al., 2008; Schein et al., 2009; Carls and Thedinga, 2010; Hook and Osborn, 2012; Bejarano et al., 2014; Wise et al., 2014). However, chemical dispersion increases the rate of transfer of dissolved and particulate oil to the water column, and may temporarily increase water column concentration by orders of magnitude compared to not using dispersants; the volume of water that is contaminated to a given concentration; and the bioavailability of the constituents of oil in water. Dispersants also decrease droplet size and reduce the rate of re-coalescence and re-surfacing of oil droplets. The interaction among these changes/processes is dynamic due to weathering, dilution, and biodegradation (see Physical Transport and Chemical Behavior, Degradation and Fate, and Efficacy and Effectiveness documents).
2. Physically and chemically dispersed oil and dispersants are expected to dilute rapidly (within hours) in the ocean (e.g. Bejarano et al., 2014; see Degradation and Fate document). Dispersants reduce the amount of floating oil and therefore reduce VOC and aerosol concentrations (See degradation and fate document). However, at the air-water interface, VOCs and aerosols may persist after dispersant application at concentrations sufficient to expose air-breathing animals via inhalation and aspiration.
3. Chemicals that are present in dispersants, such as surfactants, show a mechanism of action and elicit toxic effects that are different from many of the toxic chemical components of oil. The timeframes for uptake, toxic effects, and degradation of dispersant constituents may be different than for oil hydrocarbons.
4. While current dispersant use plans in the United States have protocols to minimize application of dispersant onto wildlife and fish (Alaska RRT Oil Dispersant Use Plan, 2014), exposure to dispersants and chemically dispersed oil can occur. For example, responders would avoid large flocks of birds, but individual birds may be exposed.
5. Only a limited number of toxicity tests have been reported for Arctic species:
 - The acute toxicity of physically and chemically dispersed crude oil and the dispersant Corexit 9500A was evaluated for key Arctic species: the copepod (*Calanus glacialis*), juvenile Arctic cod (*Boreogadus sadia*), and larval sculpin (*Myoxocephalus sp.*) (Gardiner et al., 2013).
 - Some toxicity testing work has been done on Alaskan species with Arctic and sub-Arctic distributions (e.g., Moles et al., 1979; Rice et al., 1979; Barrie et al., 1992; Moles 1998; Neff and Durell, 2012; Harvey et al., 2013; Camus et al., 2015; Rhoton et al., 2001).

Uncertainties:

1. There is little information on the kinetics of accumulation, distribution, metabolism, and elimination of hydrocarbons for Arctic species living at or near freezing (e.g., +1 to -1°C).
2. While there is limited information about the toxic effects on individual Arctic species, including those that are endangered, there is less for populations, communities or ecosystems, particularly in combination with other stressors in the changing Arctic environment (e.g. climate change, industrial development, vessel traffic, noise pollution).

B. Birds***Knowns:***

1. There are several potential pathways of dermal, inhalation, aspiration or ingestion exposure: at the surface from slicks of undispersed oil, from airborne dispersant, and swimming through plumes of dispersant and dispersed oil during diving activities (which applies to birds, otters, fur seals, and all other pelagic organisms). There is also exposure through trophic transfer and parental caretaking of bird eggs and young (Leighton, 1993; Peterson et al., 2003). The relative importance of each pathway will depend on site, species and incident specific factors.
2. Undispersed oil and its constituents have a high potential to impact birds at the sea surface through direct external contact, ingestion, and aspiration/inhalation (Leighton, 1993; Helm, 2014).
3. High bird densities (e.g., breeding congregations of planktivorous seabirds) in Arctic regions increases their risk from oil spills. High densities vary with species, location, and season, but can occur at all times of year (Petersen et al., 1999; Petersen et al., 2003; Brown et al, 2007).
4. The effects of dispersants and DDO can include disruption of feather structure, hypothermia (a harmful lowering of body temperature) due to loss of water surface tension and subsequent penetration of water or oil to skin, and loss of buoyancy potentially leading to drowning or increased energy expenditure (Lambert et al., 1982; Canevari, 1984; Stephenson, 1997; Stephenson and Andrews, 1997). This can lead to illness and/or death. In addition, dispersants themselves can increase water's penetration to the skin, causing many of the same physical effects (e.g., hypothermia, loss of buoyancy) (Duerr et al., 2011).
5. A bird does not need to be completely covered by dispersants or oil in order to affect its buoyancy or waterproofing (Jenssen and Ekker, 1991; Jenssen, 1994).

6. Under controlled laboratory conditions, dispersants alone (Corexit 9527 and 9500) are toxic to bird eggs (Albers, 1979; Wooten et al., 2012) but the relative toxicity of crude oil, and DDO is dependent on chemical composition, dose, and species.
 - Corexit 9527 toxicity to mallard eggs (hatchability) was similar to North Slope fresh crude oil; 1:5 and 1:30 DOR mixtures were more and less toxic, respectively, than crude oil (Albers, 1979).
 - Albers and Gay's (1982) environmentally-relevant exposures (using Corexit 9527 and oil in penned ponds, with mixtures transferred from hens to eggs rather than applied directly to eggs as in Albers, 1979) suggested that chemically dispersed oil is similar in toxicity to fresh crude, although both treatments demonstrated more toxicity than controls.
 - Weathered crude oil from the Gulf of Mexico was less toxic to mallard embryos than a mixture of 1:50 DOR (Corexit 9500) and more toxic than a 1:10 mixture, suggesting that greater proportions of dispersant reduce oil toxicity. All treatments (weathered oil and chemically dispersed oil) in this study were less toxic than fresh crude (Finch et al., 2012).
 - Finch et al., (2012) suggest that the toxicity differences demonstrated in these studies are due to the reduction of the concentrations of volatile PAHs in weathered oil compared to fresh crude, or the greater toxicity of Corexit 9527 (containing 2-butoxy ethanol) compared to Corexit 9500.
7. Short-term repeated oral exposure to artificially weathered LSC oil altered flight paths, increased flight duration, and increased flight distance; indicating potential negative effects on migration (NOAA, 2015).

Uncertainties:

1. There are effects of dispersed oil on feather structure, but there are few publications on the extent of the effect of environmentally relevant concentrations of dispersant alone on birds. Albers and Gay (1982) demonstrated that an environmentally relevant mix of oil and dispersant (10:1 fresh crude to Corexit 9527) adhered to bird feathers similarly to fresh crude alone.
2. There is a lack of toxicity testing, in general, on birds, so we do not have a clear understanding of sub-lethal and indirect impacts of DDO such as:
 - Behavioral effects,
 - Interactions with migratory behavior,
 - Organ damage,

- Genotoxicity, and
- Reduction in prey.

C. Marine Mammals

Knowns:

1. Undispersed oil has a high potential to impact all marine mammals at the sea surface. For example, oil alone is extremely deleterious to polar bears (Ørisland et al., 1981). Further, inhalation or aspiration of oil, and dispersed oil droplets and vapors also have a high potential to impact all marine mammals, as discussed below in the live-capture Barataria Bay dolphin studies (Schwacke et al., 2013).
2. DDO can disrupt fur structures (as they do feathers) in sea otters, polar bears, and fur seals, by decreasing water surface tension. This leads to loss of "water-proofing," increased waterlogging, and hypothermia (Hurst and Ørisland, 1982; Lipscomb et al., 1993). External oiling may also lead to hyperthermia in all marine mammals (Williams and Davis, 1995), however, this is not an issue related to oil, dispersant, or DDO toxicity.
3. There are no controlled whole animal studies on the impacts of DDO on marine mammals. However, there have been studies performed on marine mammals impacted by specific oil spills (Ballachey et al., 1994; Loughlin et al., 1996; Bowyer et al., 2003; Loughlin, 2013; Schwacke et al., 2013; Lane et al., 2015; NOAA, 2015).
4. There have been studies on the impacts of oil alone in controlled conditions on cetaceans (Geraci and St. Aubin, 1988); however, the only Arctic species tested were beluga whales (Geraci and St. Aubin, 1988) and polar bears (St. Aubin, 1990).
5. Behavioral avoidance of oil by cetaceans did not appear to have occurred in subarctic/temperate environments, including during the Exxon Valdez oil spill (EVOS) (Matkin et al., 2008) and the DWH spill (Schwacke et al., 2013; Lane et al., 2015; NOAA, 2015).
6. Polar bear behavior and natural history may predispose them to be attracted to oil, or at least to locations where oil could be concentrated. For example, bears hunt along cracks that form around hydrocarbon infrastructure (Stirling 1988), and it has been observed that polar bears will deliberately ingest oil (Derocher and Stirling 1991). Polar bears also hunt along active lead systems (Stirling 1990), where spilled oil may accumulate and persist (Neff 1990).
7. There have been studies on terrestrial and marine mammals regarding the toxicity of PAHs (Albers and Loughlin, 2003; Malcolm and Shore, 2003). Following EVOS, there

were a small number of crude and Bunker C oil toxicity studies done on mink as a model for sea otters (Mazet et al., 2000 and 2001; Schwartz et al., 2004; Mohr et al., 2008) indicating both endocrine and reproductive impacts. In addition, studies of bottlenose dolphins after the DWH oil spill also found both endocrine and reproductive impacts (Schwacke et al., 2013). Of PAHs, four-, five- and six-ring compounds have the greatest carcinogenic potential (Albers and Loughlin, 2003).

8. There are studies on biological effects using spills of opportunity; however, it is often difficult in those situations to distinguish the effects of dispersants, dispersed oil, and undispersed oil and there is usually a lack of replication or proper controls for the different exposure groups. Studies of both live animals and stranded animals following the DWH oil spill found that dolphins living in the more heavily oiled areas of Barataria Bay, Louisiana and Mississippi Sound, experienced significant health impacts, poor reproduction, and decreased survivorship as compared to dolphins in non-oiled areas. Three adverse health effects were poor body condition, lung disease and abnormal stress response (Schwacke et al., 2013; Venn-Watson et al., 2013; Lane et al., 2015; Venn-Watson et al., 2015a, Venn-Watson et al., 2015b).
9. The most important but understudied effects are those on the survivorship and fecundity of the population. This has been evaluated for a few spills of opportunity (e.g., for EVOS Loughlin et al., 1996) and DWH for bay, sound and estuary bottlenose dolphins (Lane et al., 2015, NOAA, 2015). It is often difficult to apply short-term studies on individuals to what can happen over the lifetime of organisms in a population. It is difficult to differentiate effects from exposure to oil versus DDO in individuals undergoing additional environmental influences (Peterson et al., 2003; Matkin et al., 2008).
10. There is a potential for marine mammals to be affected by chronic exposure and effects to residual oil decades after a spill. For example, Bodkin (2012) documented this for sea otters with respect to Prince William Sound residual oil.
11. Two studies considered the effects of oil or dispersants on cultured marine mammalian cells. One study investigated the effects of dispersants on the cultured dermal cells of a sperm whale (Wise et al., 2014). The other study cultured lymphocytes and monocytes from sea otters and exposed them to oil (Schwartz et al., 2005).
12. Inhalation of volatile components and aspiration of undispersed and dispersed oil are key exposure and injury risks for cetaceans (Schwacke et al., 2013), pinnipeds (Smith and Geraci, 1975; Engelhardt et al., 1977), and, to a lesser extent sea otters (Lipscomb et al., 1993) all of which breathe at the air-water interface where aerosols, volatiles and oil/dispersant droplets occur. This risk is especially high for cetaceans whose respiratory anatomy and physiology are different from all other mammals. Cetaceans: 1) lack a nasal turbinate to filter air, 2) have deep lung exchange (80-90% of the lung volume with each breathe as compared to 10-20% for humans), 3) may breath hold for long periods of time,

and 4) have rich blood supplies for rapid exchange of compounds (Irving et al., 1941; Ridgway et al., 1969; Green, 1972; NOAA, 2015). In addition, their life histories require that they have a functioning lung in order to feed efficiently at depth and migrate in the aquatic ecosystem in which they live.

13. Surface air-breathing mammals may encounter chemical components of oil at the air-water interface that can cause both toxicity and injury. These components may be released into the air as volatile organic compounds (VOCs), intermediate volatile organic compounds (iVOCs), or semivolatile organic compounds (sVOCs) (de Gouw et al., 2011; Stout, 2015; Haus, 2015; Murphy et al., 2015) and may be present in the air for inhalation or aspiration. Chemical components may also associate with small seawater droplets that can become suspended in the air column (Brock et al., 2011; de Gouw et al., 2011) due to breaking waves, wind, raindrops, animals breaking the surface, or other disruptions to the air-water interface (primary aerosols). In addition, volatiles and particles in the air can undergo chemical transformations and coalesce to form suspended particulates (secondary aerosols) (de Gouw et al., 2011; Haus 2015; Murphy et al., 2015).
14. Engelhardt (1977) noted that spilled oil might be expected to interfere with feeding behavior through its effect on baleen function, since the inner aspect of the baleen plates presents a very rough surface. This concept was supported by subsequent laboratory studies (Braithwaite et al., 1983) utilizing baleen from bowhead whales that showed filtering efficiency was reduced by approximately 10 percent when coated with Prudhoe Bay crude oil and reduced by up to 85 percent when coated with an oil of higher wax content. Geraci and St. Aubin (1982, 1985) reported similar findings for fin and gray whale baleen, and a temporary inhibition of water flow. St. Aubin et al. (1984) studied baleen from seven species of mysticete whales (minke, right, humpback, gray, fin, sei, and bowhead) and subjected samples to exposure to crude oil, gasoline, and tar but found few consistent impacts on the composition and structural integrity of the plates. They suggested that it was unlikely that transient oil exposure during a spill would deteriorate baleen plates.
15. For marine mammals, oil, dispersant, and dispersed oil exposure may be incidental to feeding or may occur directly through ingestion of contaminated prey. Oil on the seafloor may be an exposure pathway for marine mammals that feed in the benthos via digging or eating benthic prey. Marine mammals may also be exposed while feeding on mesopelagic prey throughout the water column and at the surface. The exposure pathway and level is species dependent as a function of the feeding behavior (Bodkin et al., 2012).
16. With the significant proliferation in tagging technology and diet studies, foraging areas and feeding depths are known for many marine mammals. In addition, prey studies have provided information on prey for some species or stocks of marine mammals. There may be seasonal, inter-annual and decadal changes in prey consumed by these species, and some species (e.g., baleen whales) have seasonal feeding and fasting periods and

locations. Furthermore, some species and prey distributions are changing in the Arctic (Moore and Stabeno, 2015; Kovacs et al., 2011; Stabeno et al., 2012; Moore and Huntington, 2008).

Uncertainties:

1. There are no known studies on toxicokinetics of oil or dispersed oil/dispersants in marine mammals.
2. There is uncertainty about the extent to which dispersants decrease or increase the risk of exposure and impacts of the volatile oil components at the air-water interface (VOCs, iVOCs, and sVOCs). The behavior of oil and/or dispersant at the air-water interface is important to those exposure pathways. However, there remain uncertainties about the conditions (e.g. atmospheric and oceanographic) and the impact that biological activity (e.g. marine mammal surfacing and breathing) will have on the behavior of oil and/or dispersant at the air-water interface.
3. There is currently no information on dispersant effects on baleen relative to oil effects and there has been a dearth of research on oil and baleen (including dispersed oil/dispersants) between 1980 and 2015 with only two studies being published (Braithwaite et al., 1983; St. Aubin et al., 1984).
4. Another concern for baleen fouling is whether oil or dispersed oil alters the way a whale moves through the water, changes feeding efficiency, and/or alters the level of exposure. It is uncertain whether these effects are influenced by environmental parameters (e.g., temperature and turbulence).
5. The pathways of oral exposure to oil and the effects of dispersant application on that exposure pathway and dose are uncertain. It is often thought that chronic, long-term oral exposure to PAHs is through ingestion of contaminated prey or from oil/water/dispersant in the water ingested incidental to feeding (Neff and Smith, 1979). There have been limited laboratory experiments with environmentally-relevant oral exposure to oil in marine mammals (Caldwell and Caldwell, 1982, Geraci and Smith, 1976). The significance of direct exposure to oil and dispersed oil through ingestion of contaminated water and prey while feeding (e.g., cetaceans, pinnipeds and deepwater suction feeders) is unknown. The extent to which aerial and subsea dispersant application move oil into the water column and impact marine mammals and their prey is uncertain.
6. Uncertainty exists about the impact of DDO on marine mammals because there are currently no biomarkers that distinguish exposure from oil or DDO during actual oil spills.
7. There are no toxicological studies on the impacts of DDO on Arctic marine mammals. Contributing factors include:

- There are significant ethical, legal, logistical, and financial challenges to conducting dosing studies that elucidate the impacts of oil/dispersants/dispersed oil on marine mammals.
 - Targeted toxicological dosing studies are not practical for most of Arctic marine mammal species.
 - The most feasible ways to obtain data may be through spills of opportunity, surrogate whole animal studies (if an appropriate surrogate can be identified for the species of interest), and cell culture-based or ex situ studies (which are often difficult to interpret).
8. There have been some studies on the impacts of oil on skin, but few that have evaluated the effect of dispersant on transdermal uptake or transport and pathology including what role temperature may play on these processes.

D. Fish and Lower Trophic Levels

Knowns:

1. The amount of data regarding acutely lethal toxicity of dispersed oil to Arctic fish and lower trophic levels is limited compared to the data available on temperate species (NRC, 2005; de Hoop et al., 2011).
2. The available, but limited, acute lethal toxicity data do not reveal systematic differences in sensitivity between Arctic and non-Arctic fish and lower trophic levels for dispersed oil and oil components (de Hoop et al., 2011; Olsen et al., 2011; Gardiner et al., 2013; Dussauze et al., 2014; Camus et al., 2015).
3. There are a limited number of studies in other taxa and life stages that show that Arctic species behave similarly to temperate species, when exposed to toxicants (de Hoop et al., 2011; Olsen et al., 2011; Sanchez et al., 2011; Hansen et al., 2014; Camus et al., 2015;). However, there is some evidence that low exposure temperatures may cause more delayed and lower intensity toxic effects, though the underlying mechanism for this has not been established (Hansen et al. 2011; Hansen et al. 2013). See Uncertainties #6 and #7.
4. For fish, at typical application rates for dispersants during an oil spill, potential acute lethal toxicity of chemically dispersed oil is primarily associated with the dispersed oil and dissolved oil constituents and not with the chemicals in the dispersants themselves (Carls et Al., 2008; Hemmer et al., 2011; Adams et al., 2014a). The bulk of the data are associated with non-Arctic species.

5. In general, for fish, chemically dispersed and physically-dispersed oils have similar (within three- to five-fold) acute lethal and sub-lethal toxicities (NRC, 2005; Hemmer et al., 2011; Adams et al., 2014a; Incardona et al., 2014, NOAA 2015; Gardiner et al., 2013).
6. For copepods, chemically dispersed oil and physically-dispersed oil have been shown to have slightly different toxicities (Hansen et al., 2015).
7. The conclusions in statements 5 and 6 are based on actual measured hydrocarbon concentrations in the water. Toxicity based on loading rates shows greater toxicity in the presence of dispersants because dispersants increase the partitioning of petroleum hydrocarbons in the water (Bejarano et al., 2014). Toxicity data based on loading rates are not particularly informative (See Exposure and Exposure Pathways known #3)
8. Dispersants make smaller oil droplets, increasing the surface area-to-volume ratio (Brakstad et al., 2015). This increases the rate at which oil constituents are partitioned to the water column, but does not change the toxicity of those constituents (Carls et al., 2008; Schein et al., 2009; Carls and Thedinga, 2010; Hook and Osborn 2012; Adams et al., 2014a; Bejarano et al., 2014; Incardona et al., 2014; Wise et al., 2014).
9. Effects-driven chemical fractionation points to the 3-5 –ringed alkyl PAH as the predominant cause of oil toxicity to fish embryos (Hodson et al., 2007; Adams et al., 2014b; Bornstein et al., 2014).
10. Heterocyclics can contribute to the toxicity of unweathered and weathered oils. In weathered oils, their contribution may increase when polycyclic aromatics or other lower molecular weight aromatic hydrocarbons have been depleted (Rhodes et al., 2005; Sauer and Uhler, 1994).
11. Some sub-Arctic species (e.g., Pacific herring, *Calanus* copepods) can be sensitive to dispersed oil (e.g., at ppb total PAH concentrations) (Duesterloh et al., 2002; Barron et al., 2003; Carls and Meador, 2009; Incardona et al., 2012; Incardona et al., 2014).
12. Most standardized toxicity tests (e.g., LC₅₀) measure lethality of oil to older life stages (i.e., juveniles and adults). There is limited information for more sensitive larval and embryonic life stages of Arctic species (Frantzen et al., 2012; Gardiner et al., 2013; Olsen et al., 2013).
13. Data from standard acute LC₅₀ and EC₅₀ tests can miss delayed mortality and other adverse, ecologically-important endpoints that are expressed over a longer period of time (Ingvarsdóttir et al., 2012; Olsen et al., 2013).
14. Dispersants can alter the integrity and permeability of cell membranes (Cotou et al., 2001; Hook and Osborn 2012; Almeda et al., 2014). This has been shown in bacteria and

diatoms. The permeability can be increased and the surfactants can damage the integrity of the cell membrane.

15. The significance of photo-enhanced toxicity in Arctic waters is uncertain, but the following is known:
 - UV radiation is seasonally and spatially (3D) variable (Weatherhead and Morseth, 1998).
 - Studies prior to and following the DWH spill showed that UV radiation can increase the toxicity of oil to some organisms by 1-2 orders of magnitude (Barron et al. 2003; Incardona et al., 2012; Alloy et al., 2015, NOAA 2015). This has not been tested in Arctic species, though it has been assessed in sub-Arctic, Alaskan species (Pelletier et al., 1997; Barron et al. 2003; Incardona et al., 2012).
 - There are some existing data on irradiation levels and modeled water clarity in the Arctic (Weatherhead and Morseth, 1998).
16. There is literature documenting that developing fish embryos are generally very sensitive to low concentrations of crude oil as measured by exposure and internal doses of PAHs (Heintz et al., 1999, 2000; Carls and Meador, 2009).
 - Fish embryos are especially sensitive to oil, regardless of exposure pathway (NOAA, 2015).
 - The effects on fish hearts of low-dose, early life stage exposures to oil PAHs (e.g., cardiac edemas, arrhythmia) have been demonstrated in sub-Arctic fish species (Incardona et al 2009; 2015; Sorhus et al. 2015). There are effects on cardiac mitochondrial function in Arctic fish species (Dussauze et al., 2014), but effects of early life stage exposures have not been assessed.
 - Experimental evidence has demonstrated a 40% reduction in returns of adult pink salmon after their exposure as embryos to crude oil (Heintz et al. 2000). Population modeling has suggested that exposure of embryos to low concentrations of oil can have a population level effect on salmon (Heintz, 2007).
17. There have been fewer sublethal oil and dispersant toxicity studies on invertebrate embryos than on early life stages of fish (Bellas et al., 2008; Saco-Álvarez et al., 2008; Bellas et al., 2013).
18. There are many endpoints that could be ecologically important that have not been as well assessed as acute mortality. For example, sub-lethal exposures of pink salmon embryos to oil severely reduces resistance to mechanical shock (Carls and Thedinga, 2010).

- Many protocols for determining whether dispersants can be safely used rely on toxicity tests with larval fish. Protocols have been developed for embryonic stages as part of the DWH damage assessment (NOAA, 2015).
19. There is some evidence that smaller body size (greater surface area to volume ratio) may cause greater sensitivity to DDO for:
- Phytoplankton (Fan and Reinfelder, 2003),
 - Microzooplankton (Almeda et al., 2014), and
 - Fish embryos and dispersed oil (Incardona et al., 2014).
20. Recent research from Norway (Sørhus et al., 2015) on the toxicity of oil to haddock eggs indicates that oil droplets stick to the eggs, potentially increasing the exposure of embryos to hydrocarbons. Adhesion of oil to eggs has not been reported for many fish species, but may not be unique to haddock.
21. With site-specific information about the oceanographic conditions, specific species, spawning habits, and distribution of eggs and larvae, Vikebø et al. (2015) used a modeling approach to predict the exposure of a species to dispersed and undispersed oil in an Arctic region in northern Norway. Vikebø et al. (2015) model predictions estimate, that under some scenarios, a substantial portion of the young of the year could be exposed to oil in a single event, but that this portion could be moderately reduced by the application of dispersants.

Uncertainties:

1. Standard test species and life stages may not be representative of the most sensitive members and life stages of an aquatic community (Barron et al., 2013). This is true regardless of what test species/life stages are used (now or in the future), as the most sensitive species of an ecosystem may not be known given that it is not practical or feasible to test all existing species and life stages.
2. The limited available data suggest certain Arctic species can show a delayed response compared to non-Arctic species following oil exposure (Gardiner et al., 2013; Camus et al., 2015). This may be due to differences in species physiology or exposure temperatures.
3. Relative to temperate species, there is limited information about effects of DDO on Arctic species, other than acute lethality. Newer techniques (e.g., metabolomics, proteomics, and transcriptomics) can provide mechanistic insights about sub-lethal effects caused by DDO in fishes and invertebrates (Lin et al., 2009; Van Scoy et al., 2010; Hook and Osborn, 2012; Van Scoy et al., 2012).

4. Photo-enhanced toxicity occurs in other environments, but its significance in the Arctic is uncertain. It is uncertain which organisms and life stages are at risk and how much oil and UV radiation is required to produce significant photo-toxicity in Arctic waters. This uncertainty applies to both dispersed and undispersed oil. Photo-enhanced oil toxicity may be expected in the Arctic due to longer daytime hours in the summer months, as well as reduced stratospheric ozone. However, there are many factors that impact the UV irradiance in the Arctic (e.g., the angle of the sunlight is more extreme which reduces UV radiation intensity) making it difficult to predict the magnitude of photo-toxic effects (Weatherhead et al., 2005).
5. There is incomplete understanding of the exposure processes, toxicokinetics, and toxicodynamics at the low temperatures present in the Arctic.
6. Temperature, pH, and salinity affect chemical uptake and toxicity in some aquatic species (Ramachandran et al., 2006; Whitehead 2013), but it is uncertain how these parameters affect toxicity in Arctic species.
7. There are aspects of the ecological physiology of Arctic fish relating to cold temperature that may make early life stages more susceptible to the toxic effects of oil (e.g., ion channels that maintain heart rate at extreme cold temperatures, metabolic rate) (DeVries and Eastman, 1981; Incardona et al., 2014).
8. It is uncertain how the characteristics of Arctic ecosystems influence the vulnerability of species to oil. Some examples include:
 - The accumulation of oil under the ice where there is a rich community of organisms.
 - The nearshore coastal zone of the Beaufort Sea is an area rich in fisheries, marine mammals and birds due to unique oceanographic conditions created by freshwater run-off (Carmack and MacDonald, 2002; ADNR, 2014).
 - Large aggregations of multiple species at polynas, where there is a potential for oil to accumulate.
9. It is crucial, but currently difficult, to extrapolate from effects on individuals to populations for ecological risk assessment (Chapman 2002; Calow and Forbes, 2003; Van Straalen, 2003; Hendriks et al., 2005; Barnthouse et al., 2007; Forbes et al., 2008; Fodrie et al., 2014).
 - Some attempts to extrapolate effects on individuals to populations have been made, including pink salmon (Heintz, 2007); but the inputs to the models are based on limited or incomplete data sets.

IV. Overall Summary

Knowns:

1. Environmental conditions in the Arctic (e.g., low temperatures, extreme light cycles, sea ice) may affect the behavior, distribution, and fate of spilled oil, dispersant, and dispersed oil, the extent to which marine biota are exposed to oil and to dispersants, and the effects of those exposures.
2. Dispersants change exposures to oil in several ways, though these changes are not unique to the Arctic environment:
 - The amount of oil in water
 - The amount of oil on the water surface
 - Droplet size
 - Fraction of dissolved vs. particulate
 - The array and relative concentrations of petroleum hydrocarbons that are bioavailable to aquatic species
3. Most studies of biological effects on individuals have been on temperate species.
 - There are physiological differences between Arctic and temperate species.
 - The limited numbers of studies on Arctic species show that they respond similarly to temperate species when exposed to toxicants.
4. There are population- and ecosystem- level differences between Arctic and temperate species and communities. There is information on the population biology of many Arctic species and their ecosystems; however, there is little information on the resiliency of populations to oil exposure.

Uncertainties:

1. Data about oil and dispersed oil toxicity to Arctic species is limited which leads to uncertainty in predicting impacts, particularly because of the shifting baseline due to changes in Arctic environments.
2. Compared to more temperate regions, the unique ecosystems and aspects of biology/aggregation due to time of year and life history in the Arctic create uncertainty in assessments related to dispersed and undispersed oil.
3. For any environment, there will always be uncertainties and unanswered questions regarding the biological effects of oil spills.

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This panel consisted of:

Sarah Allan, Ph.D., Toxicologist, Alaska Regional Resource Coordinator, NOAA, Office of Response and Restoration

Mace Barron, Ph.D., Gulf Ecology Division, Office of Research and Development, U.S. Environmental Protection Agency

Adriana C. Bejarano, Ph.D., Research Planning, Inc.

Jewel Bennett (retired), Ph.D., U.S. Fish & Wildlife Service

Deborah French-McCay, Ph.D., RPS ASA

Michel L. Gielazyn, Ph.D., NOAA Assessment & Restoration Division

Peter Hodson, Professor Emeritus, Department of Biology and School of Environmental Studies, Queen's University, Kingston, ON, Canada

Sharon Hook, Ph.D., Commonwealth Scientific and Industrial Research Organization (CSIRO) Oceans and Atmosphere, Australia

John Incardona, Ph.D., NOAA Northwest Fisheries Science Center

Angela Matz, Ph.D., Environmental Contaminants Specialist, U.S. Fish and Wildlife Service

Teri Rowles, D.V.M., Ph.D. NOAA National Marine Fisheries Service, Office of Protected Resources

Mathijs Smit, Ph.D., Environmental Scientist, Shell Global Solutions International BV

Mark D. Sprenger, Ph.D. U.S, Environmental Protection Agency OELM OSRTI TIFSD ERT

Ron Tjeerdema, Ph.D., DABT, Professor, Department of Environmental Toxicology, College of Agricultural & Environmental Sciences, University of California, Davis

Dana Wetzel, Ph.D., Senior Scientist, Mote Marine Laboratory, Sarasota, FL.

John Pierce Wise, Sr., Ph.D., Professor, Department of Pharmacology and Toxicology, School of Medicine, University of Louisville

Michael Ziccardi, DVM MPVM Ph.D., Director, Oiled Wildlife Care Network, Co-Director, Karen C. Drayer Wildlife Health Ctr, One Health Institute, School of Veterinary Medicine, University of California, Davis

NOAA ORR Liaison for this project: Doug Helton and Gary Shigenaka

USEPA Liaison for this project: Vanessa Principe and Greg Wilson

Facilitator: Nancy E. Kinner, Ph.D., UNH director, Coastal Response Research Center, University of New Hampshire

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