Supporting Information

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SI Text

List of Polycyclic Aromatic Hydrocarbon (PAH) Chemical Indicators. Thirteen toxicologically representative PAHs and alkylated homologs were selected for critical analysis of potentially impacted seafood (1). They included 6 noncarcinogenic PAHs (naphthalene and C1, C2, C3, and C4 alkylated naphthalene homologs; fluorene and C1, C2, and C3 alkylated fluorene homologs; anthracene/phenanthrene and combined C1, C2, C3, and C4 alkylated anthracene/phenanthrene homologs; pyrene; and fluoranthene) and 7 carcinogenic PAHs [chrysene, benzo(k) fluoranthene, benzo(b)fluoranthene, benz(a)anthracene, indeno (1,2,3-cd)pyrene, dibenz(a,h)anthracene, and benzo(*a*)pyrene]. The PAH levels of concern, and factors for their derivation, were developed specifically for this particular oil spill event.

Sample Classifications for Federal Waters. Since the initiation of seafood safety sampling on April 28, 2010, a variety of samples have been collected and they are described in the National Oceanic and Atmospheric Administration *(NOAA) National Marine Fisheries Service Seafood Sampling Plan* (2):

> Surveillance samples are collected in an area before closure. These samples help provide a baseline of preoil conditions for comparison of seafood analyzed for chemical analyses.
> Surveillance-perimeter samples are collected outside the original closed area. These samples are used to provide supplemental information on the perimeter of the closed area and to account for fish movement outside the grids.
> Surveillance-closed samples are collected within a closed harvest area but are not used for the purposes of reopening and are used to monitor seafood contamination within a closed harvest area before reopening.
> Surveillance-reopened samples are collected in areas previously closed to harvest but subsequently reopened. Sampling is conducted ~1 wk after reopening and continues through two 7-d sampling periods, separated by at least 1 wk. The purpose of this sampling is to ensure the continued safety of seafood marketed from these open harvest areas.
> Reopening samples are collected within each grid in a closed fishing area for both sensory and chemical analyses and are used specifically in reopening grids closed to harvest.
> Dockside surveillance samples are purchased by the NOAA Fisheries Southeast Fisheries Science Center port samplers in major ports in Louisiana, Mississippi, Alabama, and Florida and transported to the National Seafood Inspection Laboratory for analysis. These samples help to minimize the risk of tainted seafood reaching the market.

Seafood Collection Criteria for Sensory Testing and Chemical Analyses. For a closed area to be considered for reopening, the criteria described in the protocol (1) for sensory testing included collecting up to six subsamples per seafood type (three subsamples for oysters) for each targeted depth location at each sample location in the area under consideration for reopening. A subsample consists of individual organisms for legal size finfish and multiple organisms for shrimp and shellfish depending on the seafood type (e.g., 6 blue crabs, 10 oysters, and 0.5 pound of shrimp).

For chemical analyses, the criteria listed in the protocol (1) require collecting a minimum of 15 oysters, 0.5 pound of shrimp, and up to six finfish per species (multiple species of fish were collected from some sites, in particular in areas of heavy oiling) at or near each sample location. A sample of edible crab tissue includes collecting a minimum of 10 legal size organisms from each crab sampling location for these analyses.

Sensory and Chemical Testing. Using glass Pyrex bowls with lids, raw samples were presented as caught for smaller species and fillets for larger pelagic species. A portion of raw sample was placed into a glass-covered Pyrex bowl and transferred to a microwave oven for cooking. The sample was fully cooked and presented to the panel again with the top on the bowl. In this way as much moisture would remain to keep the sample as warm as possible throughout the test. Crabs, on the other hand, were evaluated only in the cooked state. They were brought in live and then steamed prior to evaluation. Sensory testing of seafood collected in federal waters was performed at the NSIL.

The gas chromatography/mass spectrometry (GC/MS) method (3) used to measure PAHs in seafood is a reliable and sensitive analytical method that has been used to measure these compounds in seafood and other marine organisms collected after previous oil spills and natural disasters (4, 5). For the GC/MS method (3), seafood samples were extracted with dichloromethane using an accelerated solvent extractor. Polar compounds were removed from the extracts using a gravity flow silica/alumina column and followed by separation of PAHs from interfering biogenic compounds using liquid chromatography (LC) with size exclusion chromatography. PAHs were then measured on a low-resolution quadrupole GC/MS system. To increase laboratory capacity for analysis for PAHs in seafood, a method was developed by FDA to rapidly measure PAHs using liquid chromatography/fluorescence detection (LC-FLD) (6). PAHs were extracted from seafood with acetonitrile/water using a QuEChERS (i.e., quick, easy, cheap, effective, rugged and safe) extraction procedure. Each sample extract was passed through a 0.20 μm filter and was subsequently analyzed using LC-FLD.

The method to measure the dispersant component dioctyl sodium sulfosuccinate (DOSS) in seafood used the same rapid QuEChERS extraction procedure used for PAHs followed by liquid chromatography/tandem mass spectrometry (LC-MS/MS) analysis (7, 8). All chemical analyses of seafood collected in federal waters were performed at either the Northwest Fisheries Science Center in Seattle, WA or at the NSIL. Laboratories conducting chemical analyses of Gulf seafood used a number of quality assurance measures, including analyses of method blanks, National Institute of Standards and Technology Standard Reference Materials (when available), matrix spikes, incurred chemical contaminant residues in laboratory-exposed seafood, and continuing calibration verification standards, to ensure that instruments were in excellent operating condition and that the chemical data were of known and acceptable quality.

Cancer risk. To estimate the cancer risk for individual PAH compounds likely to be found in the Gulf of Mexico light crude oil, a toxic equivalency (TEQ) approach was used. TEQ approaches are often used when determining health risks associated with exposure to mixtures of compounds with similar chemical structures and biological activities (9). The concentration of benzo(*a*)pyrene equivalents (BaPE) is considered the most valid measure of the carcinogenic potency of a complex mixture of PAHs.

For the *Deepwater Horizon* (DWH) oil spill, the carcinogenic activity for each PAH relative to benzo(*a*)pyrene (BaP) was estimated as a toxicity equivalency factor (TEF) (10). Using this method, tissue concentrations of carcinogenic PAHs (other

than BaP) were multiplied by their respective TEF and added to the BaP concentration to determine the total BaPE concentration. The following TEF values were used: chrysene, 0.001; benzo(k)fluoranthene, 0.01; benz(a)anthracene, 0.1; indeno (1,2,3-cd)pyrene, 0.1; benzo(b)fluoranthene, 0.1; and dibenz(a, h)anthracene, 1.

The following equation was used to determine the public health levels of concern (LOC) (in micrograms per gram or milligrams per kilogram equaling parts per million wet weight) for carcinogenic PAH compounds (BaPE) potentially found in seafood:

$$\mathrm{LOC(BaPE)} = (\mathrm{RL} \times \mathrm{BW} \times \mathrm{AT} \times \mathrm{CF})/(\mathrm{CSF} \times \mathrm{CR} \times \mathrm{ED}).$$

Definitions, assumptions, and specific factors used in the above equation are described below.

LOC: Level of concern.

BaPE: Benzo(*a*)pyrene equivalent.

Risk level (RL): Risk-based criteria were selected to prevent consumers from being exposed to the carcinogenic components of crude petroleum in doses that exceed a RL of 1×10^{-5} (1 in 100,000). This RL is within the acceptable range of risks (1×10^{-4} to 1×10^{-6}) used by the Food and Drug Administration (FDA) and the Environmental Protection Agency (EPA) in regulatory criteria for food and drinking water (11) and is provided as an example of an acceptable risk level in the *US EPA Guidance for Assessing Chemical Contaminant Data for Use in Fish Advisories* (12).

Body weight (BW): The average adult body weight, 80 kg, was adopted from the most recent *CDC National Health Statistics Report* (13).

Averaging time (AT): The averaging time, 78 y, was adopted from the most recent *CDC National Vital Statistics Report* (14).

Conversion factor (CF): Unit conversion factor (1,000 μg/mg).

Cancer slope factor (CSF): The upper-bound estimate of the probability that an individual will develop cancer over a lifetime as a consequence of exposure to a given dose of a specific carcinogen. For the DWH seafood risk assessment, the EPA current BaP CSF value of 7.3 $(\mathrm{mg \cdot kg \cdot d})^{-1}$ was adopted (15).

Consumption rate (CR): Consumption rates for shrimp and crab (13 g/d), oysters (12 g/d), and finfish (49 g/d) were adopted from the 2005–2006 National Health and Nutrition Examination Survey (NHANES) data for high-level (90th percentile) seafood consumers adjusted for consumption frequency. The FDA adjusted the 90% meal size to account for the number of meals consumed by a 90th percentile consumer to determine the appropriate seafood consumption rate for high-level consumers. For 90th percentile consumption values, data from the 2005–2006 NHANES 2-d recall survey were used. To determine the average daily rate for these consumers, the 2005–2006 NHANES 30-d recall survey was used to determine frequency of seafood meals eaten by 90th percentile consumers,

$$\text{Grams of seafood per day} = [\text{meal frequency}/30 \text{ d in a month}] \times \text{meal size}$$

where

Meal frequency = 9.1 meals per month for finfish, 2.9 meals for oysters, and 4.4 for shrimp/crab;

Meal size = 160 g for finfish, 120 g for oysters, and 90 g for shrimp/crab;

Grams of seafood per day = 49 g for finfish, 12 g for oysters, and 13 g for shrimp/crab.

Exposure duration (ED): The exposure duration was assumed to be 5 y. This is a conservative estimate of the potential retention period of DWH oil contaminants in Gulf seafood.

Using the assumptions and equation shown above, the levels of concern for each of the seven carcinogenic PAHs for shrimp and crabs, oysters, and finfish are presented in Fig. 3. Concentrations of alkylated homologs of the carcinogenic PAHs listed above were excluded as they are found in very low levels in the Louisiana light crude oil.

Noncancer risks. Noncancer risks were also determined on the basis of the concentrations of anthracene, phenanthrene, fluoranthene, fluorene, naphthalene, and pyrene measured in seafood. Alkylated homologs of naphthalene, fluorene, and anthracene/phenanthrene were summed with the parent compounds and compared with the appropriate toxicity criterion. The alkylated homologs of pyrene and fluoranthene were not included due to the very low levels found in the Louisiana light crude oil. The following equation was used to set the public health protective LOC (micrograms per gram or milligrams per kilogram equaling parts per million wet weight) for these noncarcinogenic PAHs potentially found in seafood:

$$\mathrm{LOC} = (\mathrm{RfD})(\mathrm{BW})(\mathrm{CF})/\mathrm{CR}.$$

The following specific factors and assumptions were used in the above equation:

Reference dose (RfD): An estimate of daily human exposure to a chemical that is likely to be without significant risk of adverse effects during a lifetime, in milligrams per kilogram per day. RfDs for selected PAH compounds were obtained from the US EPA's Integrated Risk Information Service (IRIS) database (accessed June 2010; see IRIS database for specific chemicals). The RfD for anthracene was used as a surrogate for phenanthrene.

BW: The average adult body weight, 80 kg, was adopted from the most recent *CDC National Health Statistics Report* (13).

CF: Unit conversion factor (1,000 μg/mg).

CR: Consumption rates for shrimp and crab (13 g/d), oysters (12 g/d), and finfish (49 g/d) were adopted from 2005–2006 NHANES data for high-level (90th percentile) seafood consumers adjusted for consumption frequency as described above.

Using the above equation and assumptions, the noncancer public health levels of concern for individual PAHs were calculated and are shown in Fig. 4.

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