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The Science of Natural Resource Damage Assessments

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The process for conducting natural resource damage assessments (NRDAs) is supported by important scientific investigation components. The degree to which science forms the basis of NRD settlements varies by case, but all NRDAs depend to varying extents on scientific studies. While some of these investigation components have been discussed in previous papers and agency guidance, many of the scientific components remain anecdotal and poorly supported. This article presents each of several scientific components as an integrated investigation approach and gives recommendations and examples of the methodological features.

INTRODUCTION

The natural resource damage assessment (NRDA) concept was developed to ensure that the public is adequately and appropriately compensated for the loss of services provided by resources that are impacted as a result of the release of hazardous substances or oil. In the context of NRDAs, natural resources are defined as "land, fish, wildlife, biota, air, water, ground water, drinking water supplies and other such resources . . ."[1] In the United States, there are a number of laws and regulations that are relevant to the NRDA process (U.S. Department of the Interior 1996; U.S. Code of Federal Regulations 2001), but the two most prominent statutes are CERCLA and OPA.[2] CERCLA (Comprehensive Environmental Response, Compensation, and Liability Act of 1980, also known as Superfund) directed the Department of the Interior to establish

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1 43 CFR § 11.14(z).
2 http://www.darrp.noaa.gov/about/nrda.html; http://www.doi.gov/oepc/response/a13.htm

rules and guidance for NRDAs at hazardous waste sites and for emergency incidents and longer-term releases involving hazardous substances. OPA (Oil Pollution Act of 1990) assigned responsibility for rules and guidance for NRDAs arising from oil spills in U.S. navigable waters to the National Oceanic and Atmospheric Administration (NOAA). There are some differences between the OPA and CERCLA rules, but for the purposes of this discussion, overall similarities exist in the scientific investigation steps.

The purpose of NRDAs and the scientific components thereof overlap with, but are distinct from, the cleanup of hazardous waste sites and habitats affected by oil spills. However, information and data acquired while determining the extent of contamination, and during many other aspects of the cleanup process, are important inputs to an NRDA. As described by numerous authors (e.g., Barnthouse and Stahl 2002; Gala et al. 2009), ecological risk assessment for site cleanups and injury assessments for NRDAs, though related to each other, are distinct scientific efforts. While the probabilistic nature of ecological risk assessment is a central component of site cleanups, definitive measures of actual injury are central to injury assessments as part of NRDAs. Measures of risk may be very different from measures of actual injury. The scientific information needs and data collection efforts used in cleanup activities are necessary, but not always sufficient, for the conduct of NRDAs.

In the federal rules, agencies responsible for both CERCLA and OPA assessments have provided some technical guidance concerning the kinds of assessments that can be used in NRDAs. Moreover, NOAA has released additional detailed guidance for conducting injury assessments under OPA (Huguenin et al. 1996). However, these documents are somewhat outdated and do not reflect the more recent advances in assessment techniques. Many of the injury assessment and restoration components of NRDAs are well documented in the more recent literature (e.g., Barnthouse and Stahl 2002; Dunford et al. 2004; French McCay et al. 2004). However, other steps—the scientific investigations concerning the nature and timing of release(s) of the chemicals themselves, the source(s) of chemicals, and the existing physical and chemical background and biological baseline of services that frame the releases, and hence the exposure of resources to chemicals—have not been well described. Therefore, the purpose of this article is to present and discuss the basic, important, scientific components of NRDAs through the injury quantification phase, including estimation of ecological service losses. In this review, emphasis is placed on the relevant scientific literature that postdates the available regulatory guidance from agencies involved in NRDAs. This review does not include the damage determination phase of NRDAs because these final steps involve many economic considerations and models that are used in the interpretation of service losses and are, therefore, beyond the scope of this article.

OVERALL METHODOLOGY

The scientific methodology for supporting an NRDA is generally tailored to each set of releases or a discrete oil or chemical spill and progresses through a sequence of interwoven steps. Releases and NRDAs of same can vary significantly in complexity—simple and straightforward to complex—and this variation in complexity affects the applicability and intensity of the use of the following steps. These steps focus on: (1) the details of the release(s) itself; (2) the pathways of exposure of natural resources to the released chemicals; (3) measurements of injury; and (4) ultimately, the estimation of loss of services provided by those resources. Although the steps are linked, in practice they need not be strictly sequential. The steps of the NRDA are both scientifically rigorous (i.e., include hypothesis testing as inherent in the scientific methods) and generally specified in the several regulations that drive the NRDA. These assessment steps include:

Confirmation and Analysis of the Release(s): Determining the chemistry, spatial extent, and history/duration of the release(s). Determining the release in the context of other similar sources and prerelease background levels.

Confirmation of a Pathway from the Release to Affected Resources: Determining the route(s) and mechanisms of transport of the released chemicals, and collecting evidence that links the released chemicals to the resource(s) potentially impacted.

Characterization of Baseline Conditions: Determining the biological and chemical conditions in the area of exposure that would have existed if the release(s) had not occurred so as to determine the incremental and/or cumulative impact of the released chemicals on natural resources.

Measures of Exposure: Determining the types of chemicals and amounts of those chemicals to which biological and other natural resources were exposed, including the time-varying levels of that exposure by location.

Analysis of Causation: Evaluation of whether any observed injuries are caused by the release.

Measurement of Injury: Determining the effects of those exposures on habitats (soils, sediments, water, etc.), organisms, and ultimately, the exposed populations in terms of actual injury, with the actual measures of injury to be determined on the basis of the type of resources affected.

Reductions in Services: Quantifying the implications of injuries and the scale of those injuries in terms of reductions in ecological services resulting from the injuries.

The remainder of this article explains in detail the interrelationships of each of these steps and describes the kinds of scientific analyses that can be used to accomplish each of these technical steps in an NRDA.

IMPLEMENTATION STEPS

Confirmation and Analysis of Release(s)

NRDAs were intended to cover two types of chemical releases: those involving chemicals classified under CERCLA as hazardous substances, and those involving petroleum—crude oil and petroleum products—under OPA. By statute, petroleum and petroleum products are not hazardous substances under CERCLA and are excluded under a provision of CERCLA termed the "petroleum exclusion."[3] Both the types of chemicals released and the nature of the releases can vary greatly among NRDAs, and the NRDA process for each site needs to address site-specific release scenarios. These scenarios include:

- Past releases from defunct operations, termed *legacy releases*
- Older or current releases from operating fixed facilities or predecessor operations at the same location
- Spills of various chemical types and complexities—from tank failures, facility explosions, pipeline releases, tanker spills, oil and gas exploration, and production blowouts, and others

NRDAs can vary significantly in complexity, and this variation in complexity affects the application of the scientific steps in the NRDA. NRDAs involving legacy releases under CERCLA very often require more complex and detailed science to determine the sources and timing of the releases, compared to oil spills under OPA. These needs are driven by both scientific and legal issues that bear upon establishing what really happened and when, in order to determine the duration of exposures to natural resources and assign responsibilities. Legacy releases are especially relevant at industrialized single- or multiparty sites where a variety of chemicals have been released over a period of time to the soil or sediment habitat. The progress of NRDAs over the last twenty years has shown that precise definition and analysis of the release and its source(s), while often complex, are of vital interest in establishing a factual and fair assessment.

[3] http://www.epa.gov/superfund/programs/er/triggers/haztrigs/whatsub3.htm (accessed September 22, 2008). The Webpage states, "EPA interprets CERCLA section 101(14) to exclude crude oil and fractions of crude oil—including the hazardous substances, such as benzene, that are indigenous in those petroleum substances—from the definition of hazardous substance. Under this interpretation, petroleum includes hazardous substances that are normally mixed with or added to crude oil or crude oil fractions during the refining process. This includes indigenous hazardous substances, the levels of which are increased as a normal part of the refining process."

Methods for characterizing, confirming, and reconstructing past releases fall into the area of environmental science called *historical reconstruction* or *environmental forensics* (Murphy and Morrison 2007). Though largely absent in the NRDA literature, this step of investigation is routinely part of many site investigations, including NRDAs for hazardous waste releases, as well as some oil spills where the source(s) are not apparent, and sets the stage for determining specific sources, responsibility, and duration of the release. What appears to be a straightforward regulatory-driven task of confirming that a release has occurred may, in reality, involve a complex scientific effort of verification and/or reconstruction, particularly for operating and legacy sites, to fully understand the history and characteristics of the release(s). Many examples of such reconstruction are documented in the literature (e.g., Kaplan et al. 1997; Stout and Wasielewski 2004; Davis et al. 2005; Mahinthakumar and Sayeed 2007), although few are tied to specific NRDAs.

Hazardous Chemical Releases under CERCLA

Environmental forensic investigations, which support the confirmation and analysis of release(s), answer the following questions:

- What chemicals were released?
- From what source(s) were they released?
- By what mechanisms were they released?
- When did the releases occur?

The lines of investigation and types of data collections that are needed to answer these questions include chemistry, geochronology, statistics, and aerial imagery, which collectively inform the remaining steps of the NRDA and help to understand the responsibility or shared responsibility for the release(s).

Chemical Analyses. Reconstruction usually begins with an examination of data and information from remedial investigations (RIs) of both terrestrial and aquatic locations associated with hazardous waste releases, because they provide an initial characterization of the site. During the RI process, the chemicals of concern are identified, and risks to humans and ecological resources are assessed in the context of site cleanup requirements. The results from these site investigations provide knowledge of the extent—amount and delineation—of contamination, the exposure pathways, and the potential risks posed by these chemicals. The sources of the releases and their timing, however, are not always identified in detail during an RI, and both are key to any scientifically rigorous NRDA.

For example, in many CERCLA cases the sources and timing of the release are not identified in the RI; soil/sediment sampling is almost always conducted as part of the RI and used to delineate the areas of the release and characterize current risk to biological receptors, but the timing of the releases generally is not determined. As part of a subsequent NRDA, additional data are often collected, including a more complete suite of analytes, additional tests, and the use of Sanborn Fire maps (discussed below) to delineate specific releases related to historical operations within the time periods during which those operations occurred in the area where hazardous waste contamination remains. While U.S. Environmental Protection Agency (USEPA)-mandated analyses may be adequate for mapping the extent of contamination and understanding risk from released chemicals, and may suffice in simple NRDA cases, the breadth of the chemical analyses and the detection limits of analyses may be inadequate for NRDA considerations in complex cases where source, exposure, and injury characterization often requires more detailed chemical analyses and evaluations of transport and exposure pathways.

As mentioned previously, each NRDA is unique, and the scientific needs for the assessment differ. These needs can vary based on the chemicals released and the environment in which the release occurred, or the scientific needs for the NRDA can vary based on the desires of the trustees and the potentially responsible parties (PRPs) to settle the case in a fair and expeditious manner. In the latter scenario, which is more typical of simple releases with unambiguous PRPs, the scientific data needs may be satisfied by those data collected in the RI/FS (Remedial Investigation/Feasibility Study) process; however, for more complex releases, releases in poorly understood ecological environments, or sites with several PRPs, more extensive scientific data are needed to support the NRDA.

In recent years, data needs for release characterization and source identification in complex cases have come into alignment with the need for more specificity and scientific rigor in injury assessment itself. For example, measurement of the total polychlorinated biphenyl (PCB) or total dioxin concentration in a sample no longer can be assumed to have either direct toxicological or source identification relevance. Detailed congener-specific measurements are needed not only to evaluate potential toxicity and injury, but also, in many cases, to identify the source of these chemicals (e.g., Newsted et al. 1995; Drouillard et al. 2001; Johnson et al. 2006). It is well known that congener-specific evaluations can inform historical reconstruction of releases of these materials, as far as source type, industrial process type, and ultimately, associated with processes, timing, and locations of releases within a site that can be traced to these processes (Johnson et al. 2006). The same rationale holds for polycyclic aromatic hydrocarbons (PAHs), petroleum hydrocarbons, chlorinated solvents, and the relationship of metal constituents to other metal and nonmetal constituents.

For NRDAs that require more rigorous scientific support for release characterization and source identification, more specific chemical analysis must often be coupled with higher analytical sensitivity. For example, while analysis of the sixteen USEPA Priority Pollutant PAHs, which are typically measured as part of an RI for site cleanup, may suffice for characterizing the extent of contamination and human health risk from certain PAH compounds, the breadth of the chemical analyses and the detection limits of analyses may be inadequate for NRDA considerations where source, exposure, and injury usually require a more substantial list of PAHs composed of alkylated PAHs, as well as the sixteen USEPA Priority Pollutant PAHs, at lower detection limits (Douglas et al. 2004; Boehm 2006).

PAHs related to petroleum can also be evaluated rigorously only by extending the PAH analyses (Sauer and Boehm 1991). Investigations at refineries and fuel terminal sites require not only this type of analysis for reconstruction of releases, but also manufacturing- and refining-specific time stamps—chemicals such as alkylated lead, sulfur content, and other manufacturing additives and changes that were made to the products at various well-known times in the past (e.g., Kaplan et al. 1997; Oudijk 2005). Together, the extended PAH analyses and the chemical time stamps provide robust scientific data to characterize the source of the release and the time period of release, which reduce uncertainty in the NRDA and are advantageous to both trustees and PRPs seeking a fair assessment.

Geochronology. In NRDAs involving contaminated soils and/or sediments, the reconstruction of releases may be greatly facilitated by geochronology, which documents the history of releases over time through the combined techniques of sediment coring; dating the cores to determine sedimentation rates and, therefore, the age of sediments at various depths; and chemical analyses of samples taken from specific depths in specific time horizons. Such techniques have been widely applied to sediment/soil sites and have proven to be invaluable in helping to decipher chemicals, releases, historical operations, and responsible parties. Layers in sediment/soil cores can be dated using a number of techniques, including the isotopic composition of core layers and the analysis of chemicals in these dated layers, which can help to reconstruct dates or time frames of releases, as well as the rates of release (e.g., Van Metre et al. 2004). Among the numerous examples of NRDAs that have used geochronology methods to reconstruct chemical releases over time are the Hudson River NRDA to reconstruct PCB depositional history (Bopp et al. 1981), and off the Palos Verdes Shelf in California to reconstruct PCB and dichlorodiphenyltrichloroethane (DDT) releases (Eganhouse and Pontolillo 2008).

Statistics. NRDA release and source studies often rely on multivariate statistical methods such as principal component analysis (Johnson et al. 2007) and constrained least squares (Burns et al. 1997), among other statistical methods, to explain relationships among chemicals at a site and relationships between sampling stations and areas within a site. Statistical methods also help to delineate the locations and the timing of releases. While these statistical methods can be used with a reduced analytical chemistry data set, the more comprehensive chemistry analyses described above can improve the statistical models by providing more information about the characteristics of the chemicals in the environment.

Historical Records. Historical information is needed for both cleanup and NRDA allocation, including extensive access to historical information. This information may be available from a variety of sources: for example, state and local records, media reporting, fire insurance information (Sanborn Maps; for example, http://www.sanborn.com/products/fire_insurance_maps.asp), and aerial photographs (Grip et al. 2000). These are important tools in the historical reconstruction of releases that provide documentation of the industrial history of an area. Sanborn Maps help to identify the locations of specific industrial operations at sites, which in turn, can be combined with chemical data to fully understand the history of a release. When used in conjunction with aerial photographs, now readily available for many locations, not only can the land uses be identified for specific periods, but actual topographic and bathymetric changes (e.g., landfilling changes) can be discerned.

Oil Spills under OPA

Reconstructing historical releases of oil under OPA is often a more straightforward exercise. When oil is released into the environment, whether to surface water, groundwater, or land, the source of the oil is usually known, although the proximity of additional petroleum hydrocarbon sources to the affected resources (i.e., natural oil seeps, other transient or persistent oil releases from platforms, ships, etc.) can confound accurate delineation of the footprint of the release. Occasionally, the source and timing of an oil release may be unknown; however, an oil spill is typically well reported, observed, and documented. What becomes more important for NRDAs involving oil releases is delineation of the exposure zone and differentiation from the chemical background (baseline conditions), as discussed below. Obtaining a sample of source material from an oil spill is most important and a seemingly obvious step to provide the chemical signature of the released oil; however, in practice, it is more difficult to obtain. Often, the source material varies (e.g., crude oil from a tanker mixed with bunker fuel from its engines; multiple fuel oil cargos in a single barge), so it is imperative that all possible source materials be obtained directly from the release point (or as close to the release point as can be safely accessed), to obtain the most accurate samples for source characterization.

Quantification of the release is another important, and often the most complicated, aspect of reconstructing an oil release. While it is possible to establish an upper limit on the amount of oil that can be released from the damaged tanks of a ship or barge (e.g., *Exxon Valdez*), determining the amount of oil released from a blowout of oil and gas near the sea bottom is a far more difficult measurement challenge, raising issues that go beyond the scope of this article (see, e.g., Camilli et al. 2012). Multiple, independent measurements are usually needed to characterize and quantify the volume of oil released in both types of incidents.

Pathway Confirmation

In many NRDAs scientists assume that natural resources in the general area of a release have been impacted based on chemical data or anecdotal information. However, the transport pathways of chemicals from a source to a habitat or receptor (i.e., biological resource) require careful analysis, thorough measurement, and documented evidence. As stated in appendix C of Huguenin et al. (1996), as part of the NOAA guidance on injury assessment for OPA NRDAs, "[T]o conclude that a specific injury resulted from a discharge, an exposure pathway linking the incident to the injury must be identified." Stratus (2000) summarizes Department of the Interior (USDOI) guidance as follows:

> *Pathway* refers to the route or medium through which hazardous substances are transported from the source of their release to the injured resource [43 CFR §11.14 (dd)]. *Pathway determination* is a component of injury determination [43 § 11.61 (c)(3)] in that it establishes the connection between the release and the injury. Pathway determination involves consideration of (1) the chemical and physical characteristics of the released hazardous substances, (2) the rate or mechanism of transport of the released hazardous substance, and (3) the combinations of pathways that transport hazardous substances to the exposed natural resources [43 CFR § 11.63 (a)(1)]. Pathways may be determined by demonstrating the presence of the hazardous substance in "sufficient concentrations" in the pathway, resource, or through the use of models that demonstrate the exposure route [43 CFR § 11.63 (a) (2)].

Thus, the analysis of specific contaminant transport pathways represents the scientific bridge between the previous characterization of the release(s) and the subsequent exposure assessments. For oil and chemical releases, analyses of the pathways almost always involve combinations of observations and measurements, supported by mathematical modeling.

Hazardous Waste Releases under CERCLA

Documenting transport pathways as part of the NRDA process at hazardous waste sites, whether in upland soils and groundwater or in aquatic sediment, may be equally challenging, if not more so, than at oil spill sites. There are three main reasons for the increased degree of difficulty at hazardous waste sites. The first is that the releases may have occurred over a long duration, with slow releases over time accumulating to large deposits in sediments/soils or in the subsurface (groundwater) environment. The second is that physical changes in the receiving environment (e.g., rerouting of streams, filling in of land, etc.) may have resulted in changed hydrology and flow direction over the history of the releases. Third, contaminants may be buried, and while possibly released in the past, such contaminants may no longer be available for transport to receptors and natural resources; thus, they do not represent any ongoing exposure pathway.

As described previously, the Department of the Interior regulations for conducting NRDAs address pathway determination specifically [43 CFR§11.63]. A pathway is defined in the regulations as “the route or medium through which oil or a hazardous substance is or was transported from the source of the discharge or release to the injured resource” [43 CFR §11.14(dd)]. A pathway is determined by either (1) demonstrating the presence of the oil or hazardous substance in sufficient concentrations in the pathway resource or (2) using a model that demonstrates that conditions existed in the route and in the oil or hazardous substance, such that the route served as the pathway [43 CFR §11.63(a)(2)] (Beltman and Cacela 1999). Examples include pathway analyses conducted for the Fox River NRDA (Beltman and Cacela 1999) and the Coeur d’Alene River NRDA (Stratus 2000), among others.

Pathway analyses at hazardous waste sites usually begin with a conceptual site model, wherein the transport pathways and movement of chemicals are hypothesized. The conceptual model is usually developed as part of the RI process; however, if transport pathways are not established and confirmed in the RI process, confirmation of these pathways in NRDA requires actual measurements to establish the exposure pathways. Transport pathways of chemicals from waste sites to biological receptors involve several media—surface water, groundwater, air, soils, and sediment—each one requiring consideration, with appropriately proportionate data collection, possible mathematical modeling, and confirmation. Often, the presence of a chemical of concern in a receptor (e.g., PCBs in birds) may be used as a short-cut to determining a pathway (i.e., because the chemical is present, a pathway is established). However, this approach may lack rigor, because in reality, there are often a multiplicity of possible chemical sources, especially for PCBs, PAHs, and many metals, at industrialized NRDA sites. Also, mobile receptors such as birds may have been exposed to the same chemicals at other sites. These multiple potential sources can lead to possible misinterpretation of a chemical signal to represent the confirmation of a pathway from the release. Consequently, the documentation of transport and exposure pathways requires a careful application of many disciplines and data collection, including hydrogeology/groundwater modeling; detailed environmental sampling and rigorous analytical chemistry; historical reconstruction of deposition via techniques such as geochronology; and surface water and groundwater transport modeling with measurement confirmation, along with measurements of chemicals in various media. The Fox River NRDA (Beltman and Cacela 1999) and other NRDAs involving PCBs and other complex organic compound classes, such as PAHs and dioxins, have relied on evaluating concentrations, concentration gradients, and isomer/congener patterns in pathway resources as an integral part of the exposure transport pathway analysis.

Oil Spills under OPA

In the case of most oil spills on the surface of water, aerial surveillance from multiple platforms (fixed-wing aircraft equipped with sensors flown at various altitudes; satellites) represents a critical component for tracking oil from a source to aquatic and shoreline receptors. However, under some conditions, some oil components and some chemicals that are spilled may mix into, dissolve, and/or sink into the water column after release. While crude oil is lighter than water and tends to float on the water surface, oil may tend to sink if it is mixed with suspended sediments or other materials that are heavier than water (National Research Council 1999) or is burned so that residues have a higher density than water. For example, this may occur in some rivers or other marine environments where high turbulence creates a mixing zone for suspended sediment and oil. In these and other cases where oil is combined with material that may sink, aerial surveillance and documentation of surface transport pathways should be paired with direct measurements in the water (e.g., sampling and survey methods such as acoustic profiling) and potentially on the seafloor or river bottom. The purpose is to track the extent and location-specific aspects of any subsurface movement and evaluate potential sedimentation of released chemicals.

In cases of surface water or sub(water)surface oil releases from pipelines, production platform accidents, or wellhead blowouts, surveillance and tracking from the water surface is accompanied by spill-specific water tracking using water current measurements (direction and speed), in situ measurements such as those based on fluorometry deployed with instrument packages, and discrete sampling of the water. These data may be used to evaluate potential scenarios for hypothetical fate and transport pathways for oil.

Oil spills may involve a range of different petroleum products, from light crude oils or fuel oil with abundant water-soluble constituents, as in the M/V *Braer* and M/V *North Cape* spills, respectively (Kingston 1999; Reddy and Quinn 1999), to very heavy fuels with few water-soluble constituents, as in the M/V *Prestige* spill (Diéz et al. 2007). The type of oil, the release scenario, and response actions, including the application of chemical dispersants at or below the water surface and in situ burns, affect the distribution, composition, and persistence of oil constituents in the water column, which in turn, directly affects the investigation priorities and scientific strategies employed in the pathway evaluation.

During an active oil spill, while real-time fluorometry may provide evidence for transport pathways, or lack of a pathway, actual quantitative sampling of the water and analysis of those samples for chemical constituents is needed to confirm the transport pathway and establish the spatial extent and magnitude of chemical exposures that feed into the injury assessment process. Pathways of transport to and potential exposure of resources on the bottom of a water body should, of course, be tested with direct sampling and analysis of the sediments.

A variety of mathematical models have been used to simulate transport pathways of released chemicals, both in surface waters and in groundwater. These models are especially useful for very short-duration events, for which measurements may be difficult to obtain or sparsely collected. Historically, most oil spills in the aquatic environment occur over a very short duration, so there may be limited opportunity to collect sufficient field samples to quantify exposure and document injury. Consequently, these models have become increasingly prominent in recent years, and trustees often use them to estimate potential oil fate and transport, exposure, and associated injury in oil spills. A widely used exposure assessment/injury assessment model is the *Spill Impact Model Application Package* (SIMAP) (French McCay et al. 2004), which includes a hydrodynamic model coupled with chemical fate and transport models to predict the probable chemical concentrations within an exposure area over time.

Important outputs of models, such as water-column concentrations over time and space, should be validated with empirical data to assess the accuracy and precision of the model. In this regard, models require empirical field data for model calibration, and then a separate set of empirical field data for model validation. Thus, media sampling is very important, even where models will play a significant role in injury assessment. The main weakness when modeling is conducted in lieu of empirical measurements centers on the lack of field data to validate the model results. The use of fate and transport modules in the absence of incident-specific data on biodegradation and partitioning of chemicals into dissolved and particulate components can overstate exposure and, in the case of biodegradation, potentially expand the spatial and temporal transport beyond the reality of exposure. A combination of empirical measurements to confirm pathways of exposure, along with the appropriate use of fate and transport modeling to predict the movement of water and chemicals, may provide a stronger, more technically defensible approach to exposure assessment and subsequent injury assessment than provided by modeling alone.

Characterization of Baseline Conditions

The estimation of baseline is simple in concept (i.e., conditions but for the release), but can be highly complex in actual NRDAs. In some situations, baseline can be determined in a relatively straightforward manner by the use of upstream reference areas (for riverine sites) or when prerelease data are available for a site. However, estimation of baseline for large, complex sites, especially those involving historical releases occurring over many decades, can be problematic and contentious. Determination of baseline services is especially difficult when many natural and anthropogenic stressors have historically influenced a resource in combination with the effects of a release being assessed in an NRDA (e.g., see Iannuzzi et al. 2002). In all situations, estimation of baseline should involve the use of multiple reference areas in conjunction with the analysis of biological conditions occurring over a gradient in chemical concentrations. Multivariate statistical techniques are especially useful for analysis of these kinds of data. When collecting reference-area data, the most fundamental requirement is that the reference stations should reflect, as closely as possible, the environmental conditions in the assessment area, except for the substances included in the release being assessed. Before and after sampling at locations may be part of the baseline assessment, but site-specific prespill information may not always be available or obtainable. In some cases, there may be a tendency to collect data from nearby areas that are recognized as being the least contaminated from a regional perspective. For many NRDAs, however, such data may be problematic, in that the results do not reflect the range of stressors that may be present in the assessment area absent the effects of the release.

Chemical and Other Nonbiological Conditions in Baseline Characterization

Releases that occur as a result of oil and chemical spills, and releases from operating or legacy industrial sites, often occur in locations that have been impacted by other releases (Boehm et al. 2003). In addition, other environmental factors create a dynamic range of changing abiotic baseline conditions in terrestrial and aquatic environments, conditions that need to be assessed from the literature, from government databases, and from measurement programs as part of the injury assessment studies of an NRDA. Whether a release occurs in a lake or river, or in an industrialized harbor, organic compounds and heavy metals are found that are unrelated to the release. Both CERCLA and OPA regulations recognize this fact and require that the baseline be assessed so that the addition of released chemicals can be assessed accurately.[4] Because the natural environment is always changing as a result of both large- and small-scale shifts in abiotic and biological factors, the baseline and the return to baseline conditions that define recovery do not pertain to a prerelease condition, but instead, to conditions that would have existed in the affected area

[4] CERCLA: Baseline is the "condition or conditions that would have existed at the assessment area had the discharge of oil or release of the hazardous substance under investigation not occurred" (43 CFR 11.14(e)). OPA: Baseline "means the condition of the natural resources and services that would have existed had the incident not occurred" (15 CFR 990.30).

if the release had not occurred. Thus, both chemical and other environmental changes over time are part of the baseline or background conditions that need to be documented and measured in NRDA investigations. The determination of the chemical background or baseline is accomplished through the sampling and analysis of reference sites that are unimpacted by the releases. Sampling of the chemical background includes natural and other anthropogenic sources, as well as—in the case of sites in the midst of heavy industrialization—areas upstream or otherwise removed from the influence of the site. In the case of oil spills, sampling needs to include other petroleum sources, both natural and anthropogenic. This is especially true in areas of natural oil seeps, such as the Santa Barbara Channel and the Gulf of Mexico, and in areas with erosion of high-organic, hydrocarbon-laden shale material, such as southern coastal Alaska (Kennicutt et al. 1987; I. R. MacDonald et al. 1993; Page et al. 1998; Kvenvolden and Cooper 2003). There is a wealth of information in the literature (e.g., Bradley et al. 1994; Lauenstein and Daskalakis 1998; Rabideau et al. 2007) and in U.S. government reports on inland and coastal water through the U.S. Geological Survey, USEPA-EMAP, and NOAA Status and Trends programs that can be used to inform the determination of chemical background at a site.

Differentiating background chemicals from those contained in the release not only requires an adequate number of carefully chosen reference locations, but also employing techniques of chemical fingerprinting, for which there are many approaches applicable to the variety of chemicals found in urban and remote background environmental media (Morrison and Murphy 2006; Murphy and Morrison 2007).

Beyond the chemical background, other nonbiological and biological features of the unimpacted baseline environment are important. Ocean currents, circulation patterns, and/or temperature regimes are examples of factors that may change over time, as may river flows and/or the trophic status of a lake. Weather patterns, occurrences of severe storms, droughts, and other phenomena are important aspects of the assessment of services but for the chemical releases. Included in this category are specialized large phenomena, such as the regular occurrence of low oxygen conditions, known as hypoxia, in the coastal Gulf of Mexico from the influence of the Mississippi River (Rabalais and Turner 2001). Also of particular importance are large-scale storm events that cause massive redistribution of sediments and associated contaminants.

Biological Conditions in Baseline Characterization

Baseline conditions for biological resources are frequently characterized by sampling at one or more reference areas that are presumed to be similar to the assessment site, except for exposure to the discharge or release. Although this fundamental requirement for reference areas is clearly identified in the CERCLA and OPA regulations and in relevant publications (e.g., Barnthouse and Stahl 2002), NRDA cases frequently involve significant disagreements concerning the appropriateness of reference stations used for baseline comparisons. These disagreements generally center on the degree to which the reference site is similar to the assessment site for all natural and anthropogenic factors other than the substances being assessed. This is an important issue, because if the reference area(s) are not similar in all such respects, the resultant statistical comparisons may indicate the presence of injury in the assessment area, when in fact the difference was caused by some unrelated stressor. Efforts should be made to identify the biological features (such as indicator species) that may serve as the basis for accurate and defensible comparisons.

Evaluation of the biological baseline can be similar or very different for OPA and CERCLA sites. For example, for a current oil spill, relatively recent biological studies of the impacted area or nearby areas may be available that can serve as baseline data. Even if the affected area has not been studied in the past, it may be relatively straightforward to conduct current studies of nearby unaffected areas to collect baseline data for comparative purposes. The situation for CERCLA sites can be much more complex, especially for sites where the releases being investigated may have occurred many decades ago. In such cases, there may be no comparable biological data that can be used for comparative purposes. Even if recent reference-area data are available, they may be useful only for describing current baseline conditions, which may be very different from the historical baseline. Many CERCLA sites are located in highly industrialized areas, the presence of which further complicates baseline issues because baseline must consider all other physical and chemical influences on the biological populations in addition to the release being investigated. When determining approaches to define the biological baseline, three important factors must always be considered:

1. Baseline is a dynamic condition with continuously changing influences on the structure and function of biological populations.
2. There may not be a definitive reference area that is similar to the assessment area in all respects, but for the effects of the release.
3. With the exception of some small, localized assessments of current releases, the use of a single reference station (or area) will often not be adequate to define baseline conditions.

From an optimal perspective, the use of a before-after-control-impact (BACI) study design would enable the most precise evaluation of any significant differences in biological conditions from reference areas. In such studies, data are collected at reference and impact stations before and after the release. However, with the possible exception of current discharges at sites that were monitored historically, this option is not usually available. This sampling approach also involves the appropriate accounting for how factors other than the incident being assessed have been operating through time on both the control and assessment areas. In statistical terms, this means that there is no interactive effect between treatments. For a large assessment area with relatively remote reference areas, this assumption may not be valid. Therefore, for CERCLA assessments involving historical releases, the determination of baseline conditions is best approached using multiple lines of evidence (MLOE) that potentially include the use of multiple reference areas, analyses of biological responses over gradients in chemical and physical conditions, analyses of historical data and other information, and data analyses involving multivariate statistics and modeling of past or future conditions if the data are strong enough to support reasonably accurate modeling.

Measures of Exposure

While the investigation and documentation of transport and exposure pathways is a vital link in the NRDA process, it is the actual documentation of exposure that feeds into the injury assessment process described below. In order to conclude that natural resource injuries resulted from the incident under investigation, scientists need to consider the following (Huguenin et al. 1996):

- The pathway(s) of the oil (or hazardous chemicals) from the point of discharge to the injured natural resources;
- Whether injured natural resources were exposed, either directly or indirectly, to the same oil (or hazardous chemical) that was discharged;
- The geographic and temporal nature of the exposure; and
- Whether exposure to the discharged oil (or hazardous chemicals) caused the injury.

Further, as stated in appendix C of Huguenin et al. (1996):

> Demonstrating exposure is an important step in determining injury, but evidence of exposure alone is not sufficient to conclude that injury to a natural resource has occurred (e.g., the presence of petroleum hydrocarbons in oyster tissues is not in itself an injury). The purpose of the exposure portion of an injury assessment is to determine whether natural resources came into contact, either directly or indirectly, with the oil and to estimate the amount or concentration of the oil and the geographic extent of the oil. This information is necessary to design, interpret, and extrapolate the results of the injury studies.

Exposure assessment is central to all NRDAs, and it is the purpose of all NRDA plans to determine the extent of exposures. Exposure assessment is the quantitative determination of the extent and degrees of environmental exposures of resources resulting from the released hazardous chemicals or oil components. The exposure assessment involves detailed measurements of chemicals in habitats (sediment, soil, air, or water) and/or in biological resources themselves. These chemical exposure investigations must be rigorous, in that the existence of a chemical in an environmental medium does not necessarily define its bioavailability and, hence, exposure of one or more natural resources (Neff et al. 2006; Boehm and Page 2007). Contemporary measurements may need to be paired to estimates of exposures from past releases using concentrations in media (e.g., soils and sediments) coupled to partitioning (e.g., sediment water partitioning) and other availability factors such as solubility principles (e.g., McGroddy et al. 1995; Chiou et al. 1998; Thomann et al. 2009). Additionally, exposures need to be measured over time to track the cumulative history, whether or not the return to baseline conditions (e.g., Page et al. 2005) in terms of potential injury.

Strategies for measuring exposure vary greatly, depending on the nature and duration of the releases. In oil spills, strategies for the measurement of exposures during the various phases of a spill have been documented extensively (Boehm and Page 2007). Exposures are determined: (1) from direct measurements in water, (2) from body burdens in the receptors themselves or in surrogate or passive samplers (e.g., Huckins et al. 1993, 2002), or (3) inferred from indirect measurements. Exposures may also be inferred and estimated from computer models.

There are two basic types of exposure assessments—direct and indirect. Direct measurements involve sampling and analysis of environmental media to determine the concentration of chemicals. Such measurements rely on high-quality samples and chemical analyses by laboratories that can achieve both the sensitivity (low detection limits) and specificity (analytical targets) required. The sampling and analysis specifications for direct measurements are largely dependent on two aspects: (1) the detailed chemistry of the chemicals themselves and the components therein that are potentially responsible for toxicity and, ultimately, injury; and (2) the types and locations of resources at risk through the assessment of the pathways, described above. Direct measurements should be focused not only on chemical concentrations in various media but especially on the accessibility and bioavailability of those chemicals. Chemicals that are deeply buried in soils or sediment, and chemicals that are tightly bound to a matrix such as shale rock, coal, or other forms of “black carbon” (e.g., Gustafsson et al. 1996; Accardi-Dey and Gschwend 2002) are not generally available to organisms and, therefore, will not produce injury. The mere presence of chemicals in a sample does not directly relate to exposure or injury. It is the availability of the chemicals to organisms that is relevant to exposure and injury. Indirect measures of exposure, such as enzyme induction or other biomarkers (e.g., Lee and Anderson 2005; Forbes, Palmqvist, et al. 2008; and discussed below), are sometimes used where chemical measurements are not possible or in conjunction with chemical measurements. However, such biochemical biomarker measurements, without other measurements directly related to measures of injury, are nothing more than measures of potential exposure to the chemicals associated with the release(s) in question or to similar sources unrelated to the release.

For hazardous chemicals released at waste sites, measures of exposure use the same types of approaches and techniques as those used for oil and chemical spill studies—sampling and analysis of water, sediments/soil, and biota. As is the case for oil and chemical spills, hazardous waste site studies almost always focus on potential release from sediment and soils into water bodies (i.e., overlying water or groundwater), followed by uptake of bioavailable components by receptors, and ultimately, exposure of wildlife to released chemicals directly or through plant and animal food sources. These prey species themselves acquire released chemicals from soils or sediment pore waters, through groundwater transport to surface waters, and/or through direct contact. Thus, exposure assessment studies focused on wildlife usually involve applying a detailed understanding of the feeding behavior of wildlife species to the chemical assessment of habitat and prey species in order to assess bioavailability and potential injury (e.g., Neff et al. 2006).

Analysis of Causation

Analysis of causation in an NRDA is closely related to the assessment of baseline conditions and services discussed previously. A causation analysis refers to the systematic and organized evaluation of the relative associations of various stressors (as part of baseline or the release or discharge) to the resources being assessed. Without a causal analysis, measured changes in the status of a resource cannot be adequately interpreted with regard to compensable service losses. Therefore, a demonstration of causal relationships is critical to the development of a scientifically sound NRDA, for the following reasons:

1. This analysis forms the basis for showing that the release caused an adverse effect in a natural resource.
2. The analysis can be refined to help define the magnitude of damage done to the resource.
3. The various causal relationships associated with non-anthropogenic and anthropogenic stressors that are not associated with the release (i.e., are part of baseline) must be understood and be apportioned reliably to develop an estimate of baseline services for comparative purposes.

Analysis of causal relationships for NRDAs at hazardous waste sites typically involves analysis of many chemicals as potential causal agents, usually involving many potential releases of those chemicals that may be separated in space and time. In all situations, there are many potential physical, chemical, and biological stressors (limiting factors) on local resources that are not associated with releases being assessed at the site (e.g., part of baseline conditions). Even in the case of oil spills, where the release being assessed may be the dominant potential cause of any injuries, other sources of petroleum hydrocarbons from natural or anthropogenic sources may be affecting local resources. Because of these complexities, the assessment of causation in NRDAs represents an important scientific step in the overall process of identifying injuries and determining potential damages. Moreover, the NRDA process is fundamentally a legal claim by trustees for monetary damages, which may culminate in a trial in federal or state courts. Thus, there may be high scientific standards associated with demonstrating that an injury was actually caused by the release of substances specified in the claim.

The use of various criteria for assessment of causation in ecoepidemiology has been advocated for some time, and as indicated previously, the CERCLA rule contains very general criteria associated with causal relationships. NOAA guidance for OPA (Huguenin et al. 1996) discusses specific criteria (from Fox 1991) that trustees should use in evaluating the causal pathway between the incident and a particular injury:

- *Probability.* Is there a statistically significant correlation between exposure to the incident and an adverse change in a natural resource?
- *Time Order.* Did the adverse change occur after the incident? What was the time frame for the incident, the exposure, and the adverse change to the natural resource?
- *Strength of Association.* What is the severity, frequency, or extent of the adverse change in light of exposure?
- *Specificity.* How precise is the adverse change relative to baseline? Does the adverse change also occur in nonexposed populations?
- *Consistency on Replication.* Has the association been observed repeatedly under different conditions?
- *Predictive Performance.* Is the association between the exposure and the adverse change strong enough that it can be predicted?
- *Coherence.* Is there a plausible mechanism that explains the exposure and the resultant adverse change?

These kinds of criteria are generally associated with human epidemiological studies, but are also relevant to ecological investigations. Most of these criteria are based on those originally proposed by Hill (1965) and are also referenced in USEPA's *Ecological Risk Assessment Guidance for Superfund* (U.S. Environmental Protection Agency 1997). More recently, U.S. Environmental Protection Agency (1998) published its *Guidelines for Ecological Risk Assessment* and identified four criteria associated with evidence for causality associated with chemicals:

- The injury, dysfunction, or other putative effect of the toxicant must be regularly associated with exposure to the toxicant and any contributory causal factors.

- Indicators of exposure to the toxicant must be found in the affected organisms.
- The toxic effects must be seen when organisms or communities are exposed to the toxicant under controlled conditions.
- The same indicators of exposure and effects must be identified in controlled exposures as in the field.

Although these criteria are general in nature, they indicate that a combination of field and controlled laboratory studies is required to provide a valid scientific confirmation of causation.

Epidemiological approaches to evaluation of causation, such as those proposed by USEPA or NOAA have also been identified by other authors (e.g., Fox 1991; Beyers 1998; Suter et al. 2002; Adams 2003). All of these authors recommend similar scientific frameworks for assessment of causation and share many of the important criteria for demonstrating causal relationships. Of the available criteria, four criteria based on site-specific information form the strongest basis for causal inference: co-occurrence, temporality, biological gradient, and complete exposure pathway (Suter et al. 2002).

Co-occurrence is the spatial relationship between the cause and effect and implies that, in a flowing system, the initial effect will occur only downstream from the cause. The definition of *downstream* may be complex, e.g., in tidal systems. Temporality means that the cause must precede the effect. Biological gradient indicates that the level of effect is expected to increase with increasing duration and magnitude of exposure to the causative agent. Biological gradient is a fundamental relationship in ecotoxicology that involves the quantitative relationships between stressors and the magnitude of adverse effects. Complexities in such relationships can occur, however, when populations adapt to the stressor. The evaluation of these relationships is useful for interpretation of causal relationships and for the closely related topic of baseline (Preston 2002). Even though the determination of baseline conditions may be somewhat uncertain because of complex interactive factors, the multivariate analysis of spatial gradients in biological responses relative to chemical concentrations may provide considerable value in defining a baseline envelope. Complete exposure pathway refers to the requirements for pathway determination discussed previously, and includes a demonstration that the causal agent actually reaches a receptor so that a direct effect can be manifested. Again, complexities can occur in the form of secondary and/or indirect (e.g., food web) effects.

In addition to the generic causal criteria discussed above, several authors have recommended specific approaches to assess causal information for specific kinds of ecological studies. For example, a U.S. Fish and Wildlife Service manual (Meyer and Barclay 1990) provides descriptions of the forensic techniques that should be used to investigate the cause(s) of fish kills. For sediment quality triad studies, in which synoptic data are collected on sediment chemistry, sediment toxicity, and benthic community structure (Long and Chapman 1985; Chapman 1990, 1996), the use of weight-of-evidence (WOE) approaches for assessing ecological impairment has been reviewed by Burton, Chapman, et al. (2002). For such studies, it is important to evaluate causal relationships and to document causation in accordance with field measurements (Chapman et al. 2002). As noted by Culp et al. (2000), the use of laboratory tests, field experiments (e.g., mesocosms), and field studies substantially strengthens the ability to draw firm conclusions concerning causation. Causal relationships derived from field studies should be confirmed by laboratory or field experiments.

Recent advances in causal analyses have included approaches involving multiple lines of evidence (MLOE) using a WOE approach (Menzie et al. 1996; Suter et al. 2000). Such approaches represent considerable advances beyond the relatively simple specifications of causal criteria by enabling the weighting of alternative causal explanations. Most recently, Suter et al. (2010) described a framework that includes detailed descriptions of how site-specific data and other information can be used in a WOE framework to identify potential causal agents. The framework is derived from criteria identified originally in Susser (1986) and Fox (1991). Although this framework has been developed for use by USEPA in various regulatory programs, it is highly relevant for the kinds of causal assessments that are encountered in NRDAs. In this framework, seventeen kinds of scientific evidence are scored and evaluated for the degree to which they demonstrate a causal relationship between stressors and effects.

Although various authors and agencies have identified criteria that can be used in demonstrating causal relationships, the development of specific decision-making frameworks has been proposed only recently. Such frameworks represent an important aspect for demonstrating causal relationships in a WOE approach because they can be consistent with a priori standards and involve transparent and objective approaches. For a comprehensive evaluation of causation, the framework described by Suter et al. (2010) represents a useful and robust approach for use in NRDAs.

As indicated in this review, the establishment of causal relationships is essential to form the basis of baseline determinations and demonstrate that a release actually caused injuries to a natural resource. A rigorous assessment of causality requires the use of an established framework that should be incorporated into the assessment plan. Moreover, establishing causal relationships and conducting reliable analyses of injuries necessitates the use of MLOE and an associated interpretive framework. Ideally, this framework should include a priori specification of the MLOE that will be used to evaluate causal relationships, the individual weight given each LOE, and the decision framework for interpretation of the combined results, especially if contradictory results are indicated for the individual analyses. In this manner, an iterative framework for evaluation of causal relationships can form an important part of the assessment plan and be useful for guiding the kinds of data that should be collected in the overall assessment.

Measurement of Injury

A key step in an NRDA is to determine whether injury to biological resources has occurred as a result of a release of oil or a hazardous substance. A variety of scientific methods can be used to determine whether an injury to biological resources has occurred. The selected method for determining injury should be based on the capability of the method to demonstrate a measurable biological response and relate the response to the presence of oil or a hazardous substance. Many kinds of biological responses can be used in field or laboratory studies to demonstrate injury. Examples include mortality, reduced growth or reproduction, and a variety of physiological or behavioral responses. However, not all measurable biological responses necessarily represent injury to biological resources in the context of an NRDA. According to the assessment regulations for CERCLA assessments (43 CFR § 11.62), injury can be demonstrated in a valid manner if the biological response satisfies all of the following criteria:

1. The biological response is often the result of exposure to hazardous substances
2. Exposure to hazardous substances is known to cause the biological response in free-ranging organisms
3. Exposure to hazardous substances is known to cause the biological response in controlled experiments
4. The biological response measurement is practical to perform and produces scientifically valid results

In a general sense, these criteria are related to issues of causation between substances associated with a release and a biological effect. However, these criteria are very general in nature and provide little useful information relative to a rigorous analysis of causal relationships when compared to the more specific criteria identified in the previous section.

Under CERCLA the injury assessment process includes two distinct steps: injury determination and injury quantification. For determining that a biological injury exists, the rule provides that a wide variety of biological responses can constitute an injury, including statistically significant changes in histopathological lesions, external malformations, reproductive performance, cholinesterase enzyme inhibition, behavioral abnormalities, and laboratory toxicity tests. Injury to surface-water resources can be determined by exceedance of an applicable water quality criterion if the criterion is based on an effects level rather than just exceedance of a no-effects level. However, the CERCLA rule also provides that any resource determined to be injured should be quantified according to the reduction in services when compared to baseline. Moreover, the rule indicates that, for biological resources, the difference from baseline conditions should be analyzed at the population, habitat, or ecosystem level (43 CFR § 11.71[l]). This difference between the kinds of response metrics that can be used to determine injury versus those that are meaningful for injury quantification represents a significant area of controversy among NRDA practitioners. A possible solution to this controversy is to refine the injury quantification at higher biological levels such as the population (as is discussed in a later section).

NRDAs conducted under both CERCLA and OPA include the concept of services as a fundamental metric for use in ultimately estimating the magnitude and spatial extent of any losses of a resource. Notwithstanding the guidance provided in the CERCLA rule, for biological resources, a fundamental question for many NRDAs includes the level of biological organization that is appropriate for quantification of any injuries and associated service losses. It is well known that, as the level of biological organization increases from cellular to community levels, the cost, complexity, and overall uncertainty in the assessment will also increase. Thus, a study showing significant effects at the cellular level may be cost-effective and sensitive for demonstrating a response to a hazardous substance or oil and may fit under the acceptance criteria for injury determination under CERCLA, but the central question may remain concerning whether the response represents a loss of ecological or human services that warrants compensatory damages. The demonstration of adverse effects at the population or higher level of biological organization represents an unequivocal demonstration of service losses and, in such cases, much of the controversy concerning the significance of individual losses and cellular changes would not exist. However, it is recognized that assessments should also be flexible based on the availability of existing data and the size and complexity of the site or release being assessed.

NRDAs have used a wide range of techniques for assessing injury to abiotic and biotic resources. One of the simplest approaches for assessing injuries to both abiotic (e.g., water or sediments) and biological resources (e.g., benthic macroinvertebrates) is the comparison of environmental chemistry data to published chemical benchmarks that are designed to predict the presence or absence of adverse effects on sensitive species. Such numerical values actually represent simple models that are intended to relate a chemical concentration to an adverse effect on an exposed organism. These values may also involve various assumptions concerning the relative protectiveness of the chemical concentration, such as protection of sensitive species or prediction of adverse-effect levels. The use of these benchmarks in NRDAs, especially for larger sites with historical releases, has been controversial. The issues associated with their use center on several key uncertainties, including the following:

- Does the benchmark represent an accurate threshold for an adverse response for indigenous organisms being assessed on a site-specific basis?
- If a benchmark is exceeded, is there a population- or higher-level response?
- What is the degree of service loss associated with increasing levels of exceedances of a benchmark?

Although limited attempts have been made to validate some benchmarks (see D. D. MacDonald et al. 2000 for sediment quality values), large uncertainties remain regarding the predictive abilities of such comparisons. Moreover, the validity of interpreting benchmark exceedances from a population or community perspective has not been determined. A review of the use of sediment quality guidelines (SQGs) by expert panels (Wenning and Ingersoll 2002) acknowledges some of the uncertainties associated with various methodological approaches and also concludes that these values can be used to identify concentration ranges where adverse effects are likely or unlikely to occur. However, the authors also conclude that "SQGs should be incorporated into a larger WOE framework to better evaluate the degree of adverse biological effects in sediments that fall within the transition zone of the concentration–response model." Chapman et al. (2002) found that sediment quality values (SQVs) had generally limited utility, and that the relative weighting of any SQVs used in a WOE framework should be based on a demonstration of the reliability of those values in predicting adverse effects in the field. In a specific evaluation of one group of SQGs for PCBs, Becker and Ginn (2008) showed that so-called consensus values for sediment quality had significant conceptual flaws and were not predictive of adverse effects on benthic macroinvertebrate communities under actual field conditions.

Chemical benchmarks are available for water (aquatic organisms), sediments (benthic communities), and soils (plants, invertebrates, mammals, and birds). Some of these benchmarks have been incorporated into regulatory programs and, in fact, may provide important screening-level information on the ranges of potential adverse effects of chemicals in these environmental media. However, given the substantial uncertainties associated with their derivation and interpretation, these benchmarks have limited utility for use in the injury quantification phase of NRDAs. For NRDAs with large potential liabilities and multiple causal agents associated with water, soil, or sediment contamination, quantification of injuries should be based on standardized and accepted empirical measurements of site-specific biological conditions. In such cases, there is little need for screening-level or predictive relationships as the sole indicators of injuries. Their potential use in NRDAs would be restricted (1) to the injury determination phase; (2) for initial screening assessments to identify areas for more detailed study (e.g., using laboratory or field assessment techniques); or (3) for small, localized releases where they may form the basis for settlement discussions between PRPs and trustees.

The lowest organizational level of biological measurement that has been used in NRDAs is at the sub-cellular or cellular level, and many of these measurements are commonly referred to as biomarkers. Examples of biomarkers include measurements of neurotransmitter activity (e.g., acetylcholinesterase), aryl hydrocarbon receptor (AhR) binding, vitellogenin activity, and aminolevulinic acid dehydratase (ALAD) activity. Biomarkers are intended to demonstrate either an exposure to a substance (biomarker of exposure) or an effect caused by exposure to a substance (biomarker of effect). In some cases, biomarkers have been recommended for assessment of environmental risks. However, Forbes, Palmqvist, et al. (2008) question the use of biomarkers for assessment of adverse effects at the population or higher levels and indicate that putative biomarkers of higher-level effects "must be tightly and consistently linked to responses at these higher levels." A comparison of biomarker DNA damage and oxidative stress for marine species exposed to oil indicated that biomarkers were up to fifty times as sensitive as whole-organism toxic responses (Smit et al. 2009). Results such as these indicate that, although biomarkers may provide early screening indicators of organism exposure to toxic substances, for injury determination in an NRDA they may not be predictive of effects on survival, growth, or reproduction of exposed organisms. They are also highly uncertain for use in predicting any quantifiable injuries at the population level or service losses associated with a release.

A recent area of increased emphasis in the scientific literature is the development of tissue residue benchmarks that are presumed to represent thresholds above which biological effects at the organism level would be expected. An example for fish is the benchmarks developed by Steevens et al. (2005) for dioxins and dioxin equivalents. In developing these benchmarks, the authors used rigorous data screening criteria that have not been used for some proposed benchmarks, and developed species sensitivity distributions for the resultant concentrations. Notwithstanding this approach, the derived toxicity thresholds could be calculated only for a limited number of fish species, and the threshold values covered approximately two orders of magnitude in dioxin toxic equivalents (TEQs) for individual fish species. Therefore, such values may be useful for screening exercises, but they provide little meaningful information on potential service losses that may be associated with any exceedances of the thresholds at a specific site. Recent approaches for the development of tissue residue guidelines for metals in freshwater invertebrates have shown promise for developing statistical relationships between tissue residues and population-level effects in natural systems (Luoma et al. 2010; Schmidt et al. 2011). At the current state of development, these approaches show considerable promise for use in risk assessments, especially for predicting population-level risks associated with changes in water quality resulting from remedial alternatives. However, the documented statistical uncertainties are sufficiently high that application of these predictive relationships to NRDA for the purpose of estimating service losses would not be warranted, especially given the reliability of site-specific community-level assessment techniques for aquatic invertebrates.

Although considerable controversy may be associated with the use of chemical benchmarks or biomarkers for projecting higher-level injuries, few ecologists would argue with the conclusion that measurements of responses associated with survival or reproduction provide meaningful and interpretable endpoints for assessing potential injuries that may be linked to populations or communities. However, the key question facing NRDA practitioners is associated with estimating a quantifiable service loss that corresponds to an observed effect for one or more of these kinds of responses. In other words, if a 10 percent reduction in the production of fledged young at a contaminated site when compared to baseline is observed in a breeding-bird study, what would be the population-level consequence (or service loss) of that injury? Various approaches to population-level assessments, including modeling and empirical approaches, are reviewed in Barnthouse et al. (2008). In a critical review of this issue, which is referred to as *extrapolation*, Forbes, Calow, et al. (2008) suggest that population modeling has advantages over other methods for interpretation of individual-level responses in a population context. In discussing potential linkages between organism-level responses and population-level effects, Kramer et al. (2010) indicate that organism-level responses must have two characteristics:

1. A dose-response relationship to endpoints related to survival or reproductive success; and
2. Integration of a laboratory-generated dose-response function with a field-generated demographic endpoint.

These reviews and others indicate that, although reliable population models have been developed for some species at some sites, there are important issues associated with the parameterization and application of such models, and the development of these models is expensive and time consuming. Given these limitations, the use of population models for NRDAs would most likely be restricted to large, complex sites. However, as is discussed in a later section of this review, a wide variety of empirical field approaches can be used in lieu of population models to assess injuries at the population and community levels.

Quantification of Reductions in Services

Prior to determining monetary damages or restoration requirements, the final step in an NRDA is to quantify the amount of services that are lost as a result of the release. The concept of ecological services is explicitly included in both CERCLA and OPA frameworks for both the quantification of losses from the release and identification of restoration projects that will compensate for any lost services. For the purposes of an NRDA, services are described as the physical, chemical, or biological functions that a resource may provide to another resource or to humans. In the case of ecological services, general examples include such functions as providing habitat or food, but the regulations do not provide explicit examples of how ecological services are quantified or how services are inferred from various kinds of field or laboratory data. At least one article has concluded that services are not a quantity that can be measured directly per se (Cacela et al. 2005). A review of the scientific literature is of little help in developing precise recommendations for measuring ecological services because this term is used in NRDAs. Although there is an increasing body of literature associated with the concept of *ecosystem services*, the emphasis of such discussions is almost always ultimately focused on the human use or valuation of ecosystems (e.g., National Research Council 2004). Other papers focus on the economic valuation of important ecosystem services (e.g., Barbier and Heal 2006). Recently, the potential approaches to value ecosystem services have been evaluated for the Deepwater Horizon oil spill in the Gulf of Mexico (National Research Council 2012). In cases involving the concept of ecosystem services, authors generally refer to specific kinds of individual services that are commonly assessed in NRDAs (e.g., tidal marshes as nursery habitat for important fish species).

The concept of ecological services is a critical input variable when habitat equivalency analysis (HEA) is used to assess interim losses and required restoration. However, there are generally significant amounts of uncertainty and professional judgment used in conjunction with site-specific scientific information to estimate proportional service losses per unit area for NRD sites. The amount of professional judgment is different when resource equivalency analysis (REA) is used, because this method may involve site-specific estimates of numbers or biomass of organisms lost. Examples of these kinds of estimates of injuries to salt marsh habitats, aquatic fauna, and birds are provided in Penn and Tomasi (2002). However, caution is required because of the often subjective and uncertain nature of equivalency estimates.

A different kind of framework is proposed by Cacela et al. (2005), in which percent service losses are derived from several characteristics of toxicological information, including the following:

- The type and severity of effects;
- The degree of observed effects for individual organisms;

- The extent of the effects within a population; and
- The organizational level at which the effect occurs (i.e., subcellular to ecosystem).

The authors present an example application that relates assumed service losses to varying toxicological responses, including lesions and reproductive impairment in demersal fish and several sediment quality values related to invertebrate species. Although the criteria for evaluation of toxicological information proposed by Cacela et al. (2005) are appropriate for evaluating potential service losses, the relationships between contaminant concentrations and presumed service losses involve considerable professional judgment. Moreover, the relationships between the magnitude of a particular response (e.g., a prevalence of lesions in fish or exceedance of an SQV) are not obvious and must be assigned by the investigator based on an arbitrary scale. A comprehensive review of potential proxies for ecosystem services for tidal marshes was conducted by Peterson et al. (2008). For marshes dominated by monospecific stands of plants (e.g., *Spartina* spp.), the large number of ecosystem services provided by this component of the tidal marshes can be characterized by simple metrics such as stem density and plant height. Measurements of taxa richness, diversity, and similarity to reference marshes may be useful supplementary and more appropriate metrics for tidal marshes. It is also recognized that studies of services associated with resources of special significance (threatened or endangered species) or economic importance may require individual assessment rather than reliance on plant characteristics as a proxy.

Therefore, although the regulatory definition of ecological services is general and does not identify specific associated metrics, the ecological basis for the importance of services in an NRDA is sound. The underlying concept is that healthy populations of organisms depend on adequate habitat for growth, reproduction, and feeding. Reductions in these functions below baseline conditions can be understood as losses of services provided by that habitat or organism group. For example, contamination of sediment resources by toxic substances may affect the services provided by those sediments to benthic infaunal organisms and ultimately to fish feeding on those organisms. In the case of injury assessment, the loss of services can sometimes be quantified by studying the structure and function of baseline infaunal communities, rather than a prediction of the potential adverse effects of chemical levels in the sediments themselves. Such assessments can be accomplished by comparisons to reference conditions and evaluation of chemical response gradients at the assessment site.

For individual NRDAs, it is recognized that a wide variety of assessment approaches is available, ranging from relatively simple comparisons with chemical criteria to studies of free-ranging populations. For relatively small releases of oil or hazardous substances, the site assessment may involve qualitative or semiquantitative observations when combined with professional judgment to estimate service losses. However, larger, more complex sites frequently warrant definitive studies of local populations for injury quantification because of potentially large monetary liabilities and the inherent uncertainties associated with simple predictive assessments. The use of population-level assessments for determining service losses may be necessary and appropriate, although it has been criticized because of difficulty in detecting changes at a time scale relevant to an assessment (Cacela et al. 2005). It is recognized that some populations, such as pelagic marine fish or wide-ranging birds or mammals, may be very difficult to assess. However, the recent literature provides examples of successful population-level studies in relation to site assessments. The following sections describe the kinds of population-level assessment approaches available for commonly studied biotic groups at NRDA sites.

Examples of Population-Level Studies

As indicated above, reliable quantification of service losses in an NRDA generally requires comparisons of adverse effects on biological resources at the habitat, population, or higher level. Alternatively, measurements can be made at the organism level if such metrics can be interpreted in the context of population-level effects (e.g., fledging success in reproducing birds). The following sections provide examples, for the major categories of biological resources, of field and laboratory assessment techniques that are available to determine effects at the population or community levels.

Benthic Invertebrates. Almost all NRDAs conducted in freshwater, estuarine, or marine environments include assessment of the effects of sediment contaminants on benthic macroinvertebrate communities. The general concept is that sediments may be contaminated, and therefore the services provided to benthic invertebrates may be diminished. There are well-established methods for quantification of benthic communities, using combinations of field and laboratory assessments. Since the 1980s the most widely accepted interpretive framework for both freshwater and marine surveys has been the sediment quality triad (triad) approach, described earlier, in which synoptic data are collected on sediment chemistry, sediment toxicity, and benthic community structure (Long and Chapman 1985; Chapman 1990, 1996). The concept of reference-site comparisons is an integral part of the triad approach; appropriate selection of reference sites, consistent with the concept of baseline, is essential for an accurate and reliable interpretation of study results. In reviewing the use of reference sites in sediment quality assessments, Burton, Batley, et al. (2002) conclude that "reference sites must represent the full range of conditions expected to occur naturally at all other sites to be assessed."

Since the original description of the triad approach, various decision frameworks have been proposed to use the information in an MLOE assessment (Burton, Chapman, et al. 2002; Chapman et al. 2002; Grapentine et al. 2002). The structure and characterizations used in such a decision framework are key parts of the overall triad approach, because the end result is usually an overall narrative description of relative risks for that location. The individual tests of significance for triad endpoints are usually quantitative comparisons, frequently using statistical comparisons with reference-area data for sediment toxicity and benthic community characteristics. These are, therefore, objective results that can be characterized as either being significantly different from reference or not significantly different from reference. However, the characterization of these results in the decision framework and the method of combining the results in an overall assessment frequently involve subjective decisions and may have a major effect on how a given data set is interpreted.

While triad data are generally considered the gold standard for freshwater and marine sediment assessments, controversy can arise regarding the details of the toxicity tests—specifically, whether acute or chronic tests are the most relevant to the specific NRDA. The appropriate test, however, is not necessarily a subjective decision; it should be driven by the potentially toxic persistence of the release and the estimated environmental exposures at the site. For example, if the chemical release is rapidly degraded or weathered in the environment such that the potential toxicity decreases over time, the relevant time period for the laboratory toxicity studies should reflect the release-specific chemical changes in light of the site-specific initial exposures.

Fish. As is the case for benthic invertebrates, fish resources are commonly assessed for both oil spills and hazardous waste sites in freshwater and marine environments. In the past, most NRDAs have addressed potential injuries to fish using laboratory toxicity tests or comparisons of water exposures or tissue residues to literature values. Although population and community assessment techniques for fish have been accepted as standardized approaches by fisheries scientists for many decades, the application of such techniques to NRDAs is relatively recent. For some important sport fish in the United States, there is a wealth of information on organism-level parameters that can be interpreted in the context of population-level injuries. For example, Reiser et al. (2004) sampled largemouth bass (*Micropterus salmoides*) in the Housatonic River in areas where fish are exposed to PCBs and measured five metrics: reproductive activity, young-of-year (YOY) relative abundance, YOY growth, adult growth, and adult condition. These metrics were then compared to 116 available data sets for largemouth bass populations to evaluate any adverse effects of PCBs on the local populations. In another study of the effects of PCBs on fish, Barnthouse et al. (2009) statistically evaluated relationships between maternal tissue concentrations and metrics associated with abundances of several life stages and early-life-stage survival rates over a thirty-year period. Both of these studies demonstrate that results of laboratory toxicity tests on individual organisms may not be reliable predictors of population-level effects, because of potential influences of adaptation, compensation, and other processes that occur in wild fish populations.

Birds. NRDAs conducted at sites involving bioaccumulative substances typically include assessments of injuries to piscivorous birds for aquatic sites and terrestrial (especially insectivorous) birds at upland sites. Site-specific assessments to evaluate injuries may include studies of the reproductive performance of birds in a potentially injured area when compared with reference areas. The evaluation of reproductive performance is appropriate, because this response can be related to population-level effects, and many bioaccumulative substances (e.g., PCBs, dioxins/furans, chlorinated pesticides, lead) can cause reproductive effects by the route of trophic exposure.

For aquatic sites, tree swallows have been shown to be valuable monitors of potential injuries to passerine species that may be exposed to sediment-related contaminants (Custer et al. 1998, 2003; Echols et al. 2004; Neigh et al. 2006; Fredricks et al. 2011). Advantages of using tree swallows for monitoring such sites include:

- Feed primarily on emergent aquatic insects
- Have limited foraging ranges over and near aquatic environments
- Readily use nest boxes placed in the exposure area
- Are relatively insensitive to disturbance, thus enabling nest-box monitoring

Although tree swallows are valuable for monitoring exposure to a wide variety of chemicals at sediment sites, the species has been criticized as being less sensitive to organic chemicals than other birds (McCarty 2001–). Experience at a variety of sediment sites has indicated that behavioral and developmental endpoints may be more sensitive than reproductive performance for this species.

In tree swallow studies, exposure to target substances can be monitored in prey items, eggs, and nestlings, and reproductive performance can be assessed by metrics such as nest abandonment, clutch size, hatching success, fledging success, and overall productivity. However, nest box studies should be interpreted with caution for use in estimating potential injuries for indigenous birds. For example, it is important to evaluate responses over multiple years that cover varying environmental conditions. Hallinger and Cristol (2011) found that effects of elevated mercury exposures on tree swallow reproductive success were weather dependent, and the results of the study could be very different if the sampling was conducted during average or extreme weather conditions for a particular year.

In upland areas, nest boxes may be used to assess reproductive performance for other passerine species, including western blue birds, ash-throated flycatchers, eastern bluebirds, and house wrens (Fair and Myers 2002; Fair et al. 2003; Fredricks et al. 2010). Nest boxes have also been used for assessment of effects of contaminants on wood ducks (White and Seginak 1994). These kinds of studies have involved metrics such as hatching success, fledging success, and overall productivity of young, as well as measurements of exposure by analysis of contaminants in prey items, eggs, and young of the nesting birds.

Mammals. NRDAs for both aquatic and terrestrial sites commonly include assessments for injuries to mammalian populations. Because of presumed or documented sensitivity and potential high exposure to bioaccumulative substances, the species most frequently encountered in such assessments are mink and shrews for aquatic and upland sites, respectively. Assessments of injuries to mink have involved both field investigations and controlled feeding studies using contaminated prey. Because they cannot be maintained under controlled laboratory conditions, studies of shrews must be conducted in the field.

Small mammals are commonly used for population studies at contaminated sites because of their small home ranges and potentially high exposure rates through multiple exposure routes (food, water, and direct contact during burrowing). For small mammals in upland or floodplain habitats, studies can be conducted to evaluate overall population demography, as well as relevant metrics for individual organisms. For example, Boonstra and Bowman (2003) evaluated effects of PCBs on populations of the short-tailed shrew. The study involved intensive live-trapping using mark-recapture techniques to estimate population density and the measurement of sex ratios, reproductive rates, body mass, and growth rates. A similar demographic study (Phelps and McBee 2009) was conducted at a Superfund site with metals contamination using white-footed deer mice *(Peromyscus leucopus*) as the target species. Capture-recapture methods have also been used to measure demographic parameters of deer mouse populations at a site contaminated by dieldrin (Allen and Otis 1998). An extensive study of mink populations in the Tittabawassee River, which has elevated concentrations of polychlorinated dibenzofurans, was compared with populations in nearby upstream reference areas (Zwiernik et al. 2008, 2009). Mink were trapped in the assessment and reference areas and evaluated for morphological and histological endpoints. Mink abundances were also compared with habitat suitability models for each area. For any small mammal study using mark-recapture methods it is important to consider study design and model interpretation factors such as individual capture heterogeneity and behavioral responses following initial capture (Hammond and Anthony 2006)

Integration of Injury and Service Losses

Selection of the kinds of biological data that are used to quantify injuries and determine service losses should involve consideration of the availability of pre-existing data, the magnitude and spatial extent of the release, potentially injured resources, and the individual substances constituting the release. In general, smaller and localized releases may be adequately assessed using available data, semiquantitative or qualitative information, and professional judgment. However, larger oil spills and complex CERCLA sites may require the collection of relatively large amounts of new data to enable a reliable assessment of injuries and associated damages. Because of the complexity of such cases and the difficulty of measuring baseline conditions and actual causal relationships, there may be a temptation to rely on simple comparisons of chemical concentrations in abiotic media (water, sediments, or soils) with off-the-shelf chemical criteria or analogous threshold values. Although such comparisons may meet the requirements for injury determination as described in the USDOI NRDA regulations, they do not provide reliable measurements, by themselves, for the quantification of biological injuries or service losses. Comparisons to such thresholds may be useful, however, for screening purposes or for narrowing the range of uncertainties concerning causal agents and the potential for injuries to biological populations.

Of course, it is recognized that such comparisons may be used, and agreed to by both trustees and PRPs, in some relatively straightforward assessments. Such situations essentially involve consideration of the tradeoffs between potentially large assessment and/or litigation costs and the benefits of timely resolution and restoration of injured resources. This review has demonstrated that scientifically based assessment techniques are available to assess population or community-level injuries to many diverse resources, including invertebrates, fish, birds, and mammals. In some cases, these studies may involve the collection of data at the organism level that may be interpreted in a population context (e.g., fledging success for nesting birds). In other cases, there are reliable means of directly assessing population or community-level effects using a combination of field and laboratory approaches. For all such assessments, the most reliable injury assessments will most likely involve the use of an MLOE approach, recognizing that all of the aforementioned assessment techniques differ in reliability on a case-by-case basis. For many assessments, one LOE may be associated with toxicity-test information, which may be site specific or literature based. However, the endpoints for any toxicity tests should be related in a quantifiable manner to the key functional characteristics of survival and reproduction.

SUMMARY

This review of the underlying scientific issues associated with important steps in the NRDA process has demonstrated that the scientific literature provides useful examples and appropriate analytical framework for conducting important parts of the assessment process ranging from confirmation of a release to estimation of ecological service losses. Although the regulatory framework for NRDAs provides some guidance on the kinds of assessments that may be appropriate for use in various steps of the overall processes, the specific kinds of scientific evaluations necessary to complete the assessment process are not prescribed in the regulations. This lack of specificity enables NRDAs to be flexible in nature and, in many cases, produce expedited assessments that can lead to timely and cost-effective resolutions and implementation of restoration projects. However, for larger cases that involve much more complex scientific issues (e.g., historical CERCLA sites), this lack of specificity in the regulations can lead to highly contentious issues that are based on different interpretations of alternative scientific approaches used by various parties. Examples of such issues include the definition of baseline conditions and estimation of service losses. For NRDAs of sufficient scope or complexity, this review provides guidance on the kinds of scientific study approaches that can be used to address these issues. Moreover, this review illustrates the wide variety of examples from the literature that can provide alternatives for the kinds of scientific assessments necessary to resolve key issues in large, complex NRDAs.

It is important to note that NRDAs consist of assessments other than the scientific issues discussed herein that are associated with losses of human-use services and the calculation of monetary damages associated with both ecological and human-use losses. There are large uncertainties associated with the application of various methods for estimation of losses for human services and the application of economic models to calculate monetary damages and/or the need for compensatory restoration projects.

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