

SETAC Workshop on Whole Effluent Toxicity Tests

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Session 4: Predicting Receiving System Impacts from Effluent Toxicity:

A Marine Perspective

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Introduction

The purpose of this workshop session is to critically examine case studies conducted to evaluate effluent toxicity and related receiving system impacts. One difficulty in this evaluation is that no single marine case study has been designed with the goal to comprehensively evaluate that relationship. Whole Effluent Toxicity (WET) tests originally were not designed to predict receiving system impacts. As the name states, they detect toxicity in whole effluents. However, this lack of predictability of "instream" effects was an early criticism of EPA's Complex Effluent Toxicity Testing Program (CETTP), and the freshwater component of the program undertook several case studies to successfully show a reasonable correlation between whole effluent toxicity and instream impacts (Mount, et al., 1984; 1985; 1986; Norberg-King and Mount, 1986.). Results of field studies to show cause and effect in all but single discharger situations are difficult to interpret largely because of the difficulty in showing a correlation with exposure. Correlation is a critical step in determining cause and effect. This difficulty is particularly true for saltwater because the hydrology of estuarine sites does not promote the formation of simple, linear gradients of effluent concentration by distance as in the case with streams. However, the weight-of-evidence derived from freshwater studies and from receiving water monitoring for saltwater dischargers, suggests a strong possibility that the discharge of toxic effluents will have an impact on a receiving system.

The lack of an ideal case study for the marine environment does not invalidate the value of WET tests of discharges into that environment. Field data in numerous state monitoring reports show that an effect is present, although it may be very difficult to show a cause-and-effect

linkage to a particular discharger. On the other hand, field data can never fully guarantee the absence of an effect because you can never rule out flaws in sampling design (Chapman, 1995). Direct measurement of the toxicity (or lack thereof) of an effluent allows a cause-and-effect linkage to a particular discharger. In addition, if an effluent being discharged has no measurable toxicity then it is reasonable to expect that particular effluent may not cause receiving system impacts (although we can never rule out that some ecosystem response may be more sensitive than the current suite of WET tests). Case studies have value in helping to sort out the relationship between the magnitude of WET test responses and potential field effects (as will be brought out later), but case studies have limited value as a direct, routine, regulatory tool. In this presentation we emphasize several aspects of WET tests as they relate to case studies. First, we review some of what has been done in the marine environment relative to linking effluent toxicity to the receiving system. Second, we propose a purpose for case studies. Finally, we present a discussion of how to improve the use of effluent toxicity data.

What has been done to link effluent toxicity to the receiving system?

We have discovered no case studies in the scientific literature that describe a detailed analysis of the toxicity of an effluent discharging to the estuarine or marine environments that also describes a corresponding impact on the water column and benthic communities of the receiving system. There are several plausible reasons for this, including: the extremely high costs associated with such a study; the difficulties in ascribing cause and effect due to the uncertainty of the exposure regime; and the problem of historical discharges and its effect on the contamination and/or enrichment of the benthos. However, there have been several studies that

shed some light on: a) documenting the existence of *in situ* toxic effects that can be attributed to point source discharges to estuaries; b) the relationship of effluent toxicity to receiving water toxicity within a mixing zone; and c) the documentation of benthic community impacts and chemical contamination in areas adjacent to major population centers having numerous point source discharges.

Two of the older studies examined surface waters (Woelke, 1968; Cardwell et al, 1977), using the gametes and subsequent fertilization of the pacific oyster (*Crassostrea gigas*) to discern the toxicity of ambient waters in the vicinity of pulp and paper mills in Puget Sound. These studies documented in-situ toxicity due to paper mill discharges, but also detected the rapid elimination of ambient water column toxicity when effluent was interrupted due to a mill strike. No detailed toxicity analysis of the effluent itself was conducted by authors in either study, nor was there any analysis of the condition of the benthic community at any of the discharge sites.

We could verify only one study that investigated the direct relationship of effluent toxicity and estuarine/marine receiving water toxicity (Schimmel et al, 1989). In that study, five estuarine toxicity test methods currently in use within the NPDES permitting process were evaluated at seven sites along the Gulf and Atlantic Coasts. Among the conclusions drawn from these data were that: the precision of the estuarine toxicity tests were consistent with those reported for freshwater tests; the effluent toxicity test methods reliably detect toxicity in a wide variety of effluents and receiving waters; the species most sensitive to effluents were those most sensitive to the corresponding receiving waters; and that, when detected, receiving water toxicity was (with one exception) generally near-field in nature and within the zone of initial dilution.

Much of the historical published effects data associated with the impacts of specific point source discharges to the estuarine environment deal with benthic impacts (e.g., Pearson and Rosenberg, 1978), and relatively few attempt to trace the effects from the discharge to the water column. Benthic community structure generally is a good indicator of environmental conditions in estuaries. Benthic organisms live in direct contact with the sediment and pore water, and have limited mobility. Benthic communities integrate the effects of multiple stresses over time, and are, therefore, a reasonable and effective indicator of the extent and magnitude of pollution impacts in estuarine environments (Bilyard, 1987; Holland et al., 1988 and 1989). Benthic invertebrates also are a critical link in the aquatic food chain, serving as food for a wide variety of fish species and larger benthic invertebrates such as lobsters and crabs. However, relating effluent toxicity to benthic effects is difficult. There simply is no verification of exposure, since the plume is frequently at or near the surface of the water (Baumgartner, et al., 1994; E. Dettmann, U.S. EPA, Atlantic Ecology Division, Narragansett, RI).

Although there are many examples in the literature of benthic impacts and chemical contamination of sediments in the vicinity of point source discharges, perhaps the most informative examples of the extent of these anthropogenic impacts lie in regional monitoring programs. The National Status and Trends (NS&T) Monitoring Program has been in existence since 1986 and, predictably, results indicate that the areas of highest sediment contamination are in embayments and harbors with the highest populations and point source discharges (O'Connor and Ehler, 1991; Wolfe et al, 1994). The Environmental Monitoring and Assessment Program's Virginian Province study from 1990-1993 indicated similar results. Long Island Sound, one

region within the Virginian Province, exhibited significant hypoxia in bottom waters closest to New York City and smaller embayments surrounding the Sound had the highest levels of chemical contaminants and sediment toxicity (Schimmel, 1995). In addition, these enriched, contaminated and toxic sediments were in closest proximity to many major point source discharges (Schimmel and Morrison, 1995). Although it would be impossible to discern the effects from point and non-point source discharges in these environments, there can be no reasonable doubt that historic toxic contamination of coastal environments is due, at least in part, to point source discharges. It is this contamination that the WET tests are designed to help prevent in the future.

What is the purpose of case studies?

In many estuaries, the combination of multiple discharges with the complex movements of estuarine waters, raises significant uncertainty that any toxicity (or field effects) detected in the vicinity of a specific discharge can be attributed solely to the discharge in question. As mentioned earlier, however, case studies can be designed to minimize the above uncertainty, namely by selection of an area with a single discharger. Whereas relatively uncomplicated sites can be selected for case studies, field sites mandated for direct regulatory purposes (i.e., to decide if an effluent is toxic or not) are usually complicated by factors such as multiple discharges.

If we are not going to use field data for regulatory purposes, then why conduct case studies at all? Clearly there is a need to consider ecological significance when we make a

judgement as to how big an effect on survival, growth, or reproduction (depending on the particular WET test) is important. Just because we can measure a statistical effect does not necessarily mean that it is biologically significant. WET tests are biological "meters" measuring something we choose to call toxicity. Toxicity is essentially a continuous value from 0 to 100% effect. Conducting case studies that are designed specifically to evaluate the relationship between effluent toxicity and receiving system impact can be very valuable in making the judgement as to what percentage effect along that continuum should be considered significant. This will likely be different for each test species.

Two things must be taken into account when designing a case study for evaluating effluent effects on receiving systems. First, case studies traditionally have centered on the measurement of ecosystem structure not function. Although changes in attributes such as numbers and types of species and standing crop are important to consider, it is just as important to consider such functional aspects of an ecosystem as productivity, and energy and material flows (Odum, 1985). It is possible that significant impacts on ecosystem structure can take place with little or no noticeable impact on function; the reverse may also be true. In addition, using community structure as an indication of the impacts of a discharge may only provide information on the accumulated history of contamination and/or enrichment. This history of contamination may exist from the same industry or municipality before an upgrade in effluent treatment, and not reflect the current improved state of the discharge. Worse, the history of contamination may be from an industrial site attributed to a previous tenant, totally unrelated to the present occupant. Indeed, due to the beneficial effects of the Clean Water Act, it is likely that a discharge from a

previous tenant (or earlier discharge from the same tenant) was more heavily contaminated or was laden with more particulates and nutrients than the present discharge.

Secondly, even if we are able to accurately account for all possible field effects, the current suite of WET tests are not designed to detect all impacts (e.g., eutrophication). The EPA's Environmental Monitoring and Assessment Program for the Virginian Province has clearly demonstrated that, on an aerial basis, potential impacts to the coastal environment from low dissolved oxygen (presumably from eutrophication caused, in part, by domestic waste dischargers) exceed those from toxic substances (Strobel et al., 1995); eutrophic effects are not discernable from WET tests. Several researchers also have reported on the abundance of genotoxic effects in domestic and industrial effluents (Meier et al., 1987; Stahl, 1991; White and Rasmussen, in press). If we are going to judge the direct applicability of WET tests to receiving system impacts, we must design the tests to account for all potential types impacts, including eutrophic effects, bioaccumulation of toxic substances, and genotoxic effects.

Once we have conducted the "ideal" series of case studies, and have determined the ecological significance percentage effect for each WET test (no small task in time, money, or degree of difficulty), we need to remind ourselves of the appropriate perspective on WET tests as they relate to receiving system impacts. Results of a case study should only be used to evaluate the power, biological significance (or whatever term you want to use) of the percentage effect chosen as representing "toxic" results from a WET test. If WET test are going to be protective rather than reactive, they must be more sensitive than the receiving system. By current use, if we see toxicity in an effluent we are predicting that the potential for a receiving system impact

exists. However, predictable field effects from laboratory results can only be validated by pursuing an exposure scenario that ultimately results in a manifestation of the predicted effects. "This is not a particularly useful course of action if one is trying to avoid pollution" (Chapman, 1995). Ecosystems can tolerate a variety of stresses without much outward sign of injury, then reach a disruption threshold at which the cumulative consequences finally reveal themselves in critical proportions (Myers, 1995). The problem with monitoring the receiving system for impacts is that we do not know how far we are from that threshold, nor are there any reliable ways to measure the location of the threshold. The problem with using toxicity alone is how are we going to define toxicity. This leads us to the next section.

What can we do now to improve the use of effluent toxicity data?

We have already established our position that whole effluent toxicity is a useful indicator for preventing impacts in fresh waters. However, we need a clearer understanding of how we are going to define toxicity. There is variability associated with the results of any WET test. Once a test method has been developed and the power of that test demonstrated, then there should be no need for field validation (Chapman, 1995). The emphasis should be placed on how we are going to define toxicity of the effluent itself. To demonstrate the power of a given WET test we need to deal with variability. There are two primary sources of variability associated with effluent toxicity; that attributed to the tests and that from the effluent. Variability is a real aspect of anything we do. We cannot rid ourselves of this phenomenon, but we can characterize it, decide what level is "acceptable" or typical, and use that knowledge in our decisions concerning the potential for an effluent to be toxic. Since there is a workshop session devoted to variability, we

will not go into detail here, but just touch on what we consider to be the main issues.

Test Variability-- We have not been particularly good at incorporating statistical variability of test data into the decision making process for effluent toxicity. When we conduct a hypothesis test (analysis of variance, *t*-test, etc.), we need to remember that the statistical test just gives us an **estimate** of reality. Whatever decision we make, either that an effluent is toxic or that it is not toxic, there is an error associated with that decision. If we decide, based on the statistics, that an effluent is toxic, there is a certain probability that it is not toxic. This probability is usually set at 5% (fixed by the *alpha* level chosen--generally 0.05). If we decide that an effluent treatment is not toxic there is a probability that it is in reality toxic. This probability (referred to as *beta*) is not fixed and varies depending on the magnitude of the response by the organisms exposed to the effluent (relative to the control) and the variability of that response (variance)¹. *Beta* often seems to be forgotten, with all error assumed to be "5%". Our confidence (e.g., power = one minus *beta*) in a decision that there is a statistical difference depends on the value of *beta*. However, *beta* error can vary greatly. The smaller the difference in response relative to the control and the greater the variability in that response, the greater the *beta* error. It is important that we incorporate an understanding of all sources of statistical error into our decision making process.

If a test procedure could be exactly duplicated, then there would be no variability among results either within a single test (among replicates for a given control or treatment), among tests within a single laboratory (intra-laboratory variability), or among different laboratories (inter-

¹*Beta* also is a function of the *alpha* level chosen, however, *alpha* is usually set at 0.05.

laboratory). We know that this is not possible. There is inherent "random error" in all three aspects of test variability mentioned above. Thus, we must characterize each of these sources of variability and use that characterization to decide what kinds of differences each test was designed to detect. The current WET methods have acceptance criteria that permit a wide range of variability among replicates for both controls and treatments. This does not mean that the test methods are flawed, just that we must consider variability in our decision making criteria about effluent toxicity. The greater the variability in results the greater the difference required between a control and treatment before the treatment is declared statistically different from that control. Thus, there exists the potential for rewarding sloppy data with a higher probability of passing a WET test. On the other hand, there is no incentive for dischargers to obtain "better" (less variable) data. There should be in place a system of data interpretation that rewards using better methods and better laboratories. The ideal data interpretation system would reward extra effort (more treatments, more replicates, more organisms, more tests, etc.) with a higher percentage acceptable effluent--not a system that potentially rewards high variability and a low "n". By now there should be ample data for each of the WET tests that decisions concerning acceptable variability also can be incorporated into acceptance criteria for each test.

Effluent Variability--The second primary source of variability is with the effluent itself. This is true variability (as opposed to the above random error). The EPA's Technical Support Document (TSD--EPA, 1991) requires that effluent variability be characterized as a part of the permitting process. The more variable an effluent, presumably the more frequently WET tests are to be performed. However, the TSD does not give guidance on how many replicate samples one

is to take for a given sampling event. Not only does the large scale temporal variability of the effluent need to be characterized, but also the small scale temporal and spatial variability. The greater the variability the more individual replicate samples that must be taken during a given sampling event. Currently there is no guidance on how many grab samples or composites must be taken during each sampling event. If we are going to continue to place the regulatory emphasis on preventing toxic effluents from entering the nation's waters, then we must have an effective means to incorporate all of the sources of variability into a decision concerning toxicity. However, as with test variability, we need to have in place incentives that reward more data from a discharger.

We propose the following as a procedure for improving the use of effluent toxicity data as it relates to a single effluent sample.

1. Use power curves and intra- and inter-laboratory precision data to decide what difference from the control each test was "designed to detect". This difference also should be evaluated via case studies. That is to what degree might the detectable difference be over or under protective. Depending on the test species this difference might increase or decrease in the light of case studies.
2. Incorporate confidence intervals (e.g., 95%) in our effluent toxicity data evaluations. If, for example, we used the lower confidence limit for a given endpoint (e.g., the EC25 or EC50, NOECs or LOECs--endpoints and their method of calculation are the topic of a separate workshop session here), then dischargers would have an automatic

incentive to get the best data possible. The more replicates used, the less variability among the replicates (i.e., the use of a well qualified laboratory with good QA track record), etc. the tighter the confidence limits and the greater the percentage effluent represented by the lower confidence limit. This is analogous to the "benchmark dose" of Crump (1984) as discussed in Hoekstra and van Ewuk (1993).

Summary

Case studies relating effluent toxicity to receiving system impacts are very valuable in evaluating the biological significance of percentage reductions in WET test endpoints. No one marine case study exists, however, that allows such an evaluation. In the absence of such a case study we should proceed with a logical next step, the incorporation of variability into the decision making process that leads to labeling a given effluent toxic or non-toxic. If we are going to protect against negative impacts on receiving systems, then that decision-making process also must be a conservative one. We cannot insist on a demonstrated field effect before declaring a given effluent toxic and in need of alteration.

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