

The Role of Traditional and Novel Toxicity Test Methods in Assessing Stormwater and Sediment Contamination

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Abstract: Traditional effluent and ambient water column toxicity tests have been used widely for evaluating the contamination of stormwaters and sediments. These assays consist of a routine bioassay exposure design of 1 to 9 days using freshwater and marine/estuarine species known to be sensitive to a wide range of toxicants. While effluent toxicity may be indicative of sediment or stormwater toxicity in the receiving system, the exposure is different, and therefore toxicity cannot be readily predicted. Traditional, standardized, whole effluent toxicity (WET) test methods have been used effectively and also misused in evaluations of whole sediments, pore (interstitial) water, elutriates (extracts), and stormwaters. Results show these methods to be very sensitive to sediment and stormwater toxicity. These traditional toxicity tests are predictive of instream sediment or stormwater effects where significant contamination exists or where exposure concentrations are similar. Modifications of these standardized test methods to include sediments or pore waters have been shown to be as sensitive as short-term, whole sediment toxicity tests using benthic species. However, the added complexity of sediments and stormwaters (e.g., partitioning, high K_{ow} compound bioavailability, suspended solids, sporadic exposures, multiple exposure pathways) dictates that traditional toxicity test applications be integrated into a more comprehensive assessment of ecologically significant stressors. The limitations of the WET testing approach and optimized sample collection and exposure alternatives are frequently ignored when implemented. Exposure to sporadic pulses of contaminants (such as in stormwaters) often produce greater toxicity than exposure to constant concentrations. Lethality from short-term pulse exposures may not occur for weeks after the high flow event due to uptake dynamics. Pore water and elutriate exposures remove sediment ingestion routes of exposure and alter natural sorption/desorption dynamics. Traditional toxicity tests may not produce reliable conclusions when used to detect the adverse effects of: fluctuating stressor exposures, nutrients, suspended solids, temperature, UV light, flow, mutagenicity, carcinogenicity, teratogenicity, endocrine disruption, or other important subcellular responses. This reality and the fact that ecologically significant levels of high K_{ow} compounds may not produce short-term responses in exposures dictates that additional and novel assessment tools be utilized in order to protect aquatic ecosystems. This inability to predict effects is largely a result of the complex biological response patterns that result from various combinations of stressor magnitudes, duration, and frequency between exposures and also the interactions of stressor mixtures, such as synergistic effects of certain pesticides, metals, and temperature. In watersheds receiving multiple sources of stressors, accurate assessments should define spatial-temporal profiles of exposure and effects using a range

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of laboratory (such as WET tests) and novel *in situ* toxicity and bioaccumulation assays, with simultaneous characterizations of physicochemical conditions and indigenous communities.

KEY WORDS: stressors, indigenous communities, *in situ*, bioassays.

I. INTRODUCTION

Should standardized effluent toxicity test methods be used in evaluations of sediment and storm water contamination? A growing body of literature suggests they are useful assessment tools when used correctly, and in a multicomponent assessment approach. The most widely used effluent toxicity tests are the acute and chronic toxicity test methods currently used by the U.S. Environmental Protection Agency (USEPA) (USEPA, 1991a,1993a, 1994a). They comprise one of three primary approaches (including chemical-specific and bioassessments) used for water quality-based toxics control (USEPA 1991a). While these methods were designed for whole effluent toxicity (WET) testing, they are, in fact, simplistic assays that measure the toxicity responses of surrogate species. The exposure media they reside in during the assay (whether it be effluent, ambient water, pore water, elutriate, whole sediment, or stormwater) is perhaps of less importance in a discussion of their relevance than the manner in which they are exposed is and its applicability to the receiving water system. The traditional WET test, when properly conducted, measures the chemical toxicity of a media during constant conditions and can provide useful information on various compartments of the receiving water system, such as mixing zones, sediments, pore waters, and stormwaters. The following discussions present the critical issues, strengths, and weaknesses of using these traditional toxicity test methods and novel alternatives for use in assessments of stormwater and sediment contamination.

II. ASSUMPTIONS OF WET TEST METHODS

The assumptions, strengths, and limitations of the USEPA's WET test methods have been thoroughly documented (e.g., Grothe et al., 1996; USEPA, 1991a). Briefly, the primary assumptions and methodological parameters are as follows:

- Acute toxicity = \leq 96 h lethality
- Chronic toxicity = 7 to 9 days (except *Selenastrum capricornutum* and *Arbacia punctulata*)
- Effluents are collected (grab or composite) and placed in test chambers in a defined dilution series
- Dilution water is laboratory water or receiving water
- Acceptability, performance criteria must be met (e.g., \geq 80% survival)

- Threshold effect levels are calculated with associated statistical significance from the dilution series responses
- Standardized test methods and test species must be used
- Quality assurance and quality control guidelines must be met (e.g., organism age, health, water quality monitoring)
- Usually static-renewal; however, static or flow-through exposures used
- Assume surrogate test species are protective of 95% of resident species and recommend 3 species be tested (fish, invertebrate, and plant); however, usually a fish and/or invertebrate tested
- Acute-to-chronic ratio used to extrapolate to a chronic toxicity concentration

The USEPA (1991a) acknowledges several key limitations to the WET tests that also apply to their application to stormwaters and sediments. Some of the limitations more relevant to this manuscript include: (1) testing of only 1 to 3 test organisms may not detect toxicants with a specific mode of action; (2) bioaccumulative or “downstream” cumulative toxicity is not measured; (3) receiving water interactions with chemical and physical conditions may enhance or remove toxicity; the causative agent is difficult to identify; and (4) variable effluent toxicity requires dynamic characterization methods. Unfortunately, these limitations are often ignored.

Odum (1992) stated that stress is usually first detected in sensitive species at the population level. Natural population and community responses are not measured directly with WET tests (La Point et al., 1996; La Point et al., 2000). The traditional surrogates (*P. promelas* and *C. dubia*) may not be as sensitive as indigenous species (Cherry et al., 1991). Indirect effects of toxicity on species, population, and community interactions can be important (Clements et al., 1989; Clements and Kiffney, 1996; Day et al., 1995; Fairchild et al., 1992; Giesey et al., 1979; Gonzalez 1994; Hulbert 1975; La Point et al., 2000; Schindler, 1987; Wipfli and Merritt, 1994), and may not be detected by WET testing. A huge ecological database exists showing the importance of species interactions in structuring communities (e.g., Dayton, 1971; Power et al., 1988; Pratt et al., 1981).

It is less likely that strong relationships will exist between WET test responses and indigenous communities at sites where there are other pollutant sources, effluent toxicity is low to moderate, or dilution is high. Based on fish and benthic invertebrate responses, several studies suggest that WET tests are not always predictive of receiving water impacts (Clements and Kiffney, 1994; Cook et al., 1999; Dickson et al., 1992, 1996; Niederlehner et al., 1985; Ohio EPA, 1987); however, many studies have shown WET tests to be predictive of aquatic impacts (e.g., Birge et al., 1989; Diamond et al., 1997; Dickson et al., 1992, 1996; Eagleson et al., 1990; Schimmel and Thursby, 1996; Waller et al., 1996). These differences should not be surprising however, as it is likely a result of WET test organisms and field populations experiencing different exposures (USEPA, 1991a). In an effluent-dominated system, the in-stream exposure may be relatively similar to the

constant effluent exposure characteristic of a laboratory WET test. A less degraded watershed, one dominated by pulse non-point source (NPS) inputs, one that is not dominated by point source effluents, and/or exposures that are highly variable may have indigenous populations that are exposed to toxicity not detected in a WET test. If sensitive species have already been lost from a watershed, a toxic effluent may be inhibiting their return. In highly degraded sites, virtually any traditional assessment tool (acute toxicity testing, chemical concentrations, indigenous communities) can demonstrate a pollution problem exists with strong statistical relationships. The WET tests were not developed to evaluate all natural and anthropogenic stressors or to show all biological responses (such as, mutagenicity, carcinogenicity, teratogenicity, endocrine disruption, or other important subcellular responses). In addition, highly nonpolar compounds may be elicit an effect in short-term exposures. These issues dictate that additional assessment tools may be utilized in order to protect aquatic ecosystems (Waller et al., 1996).

In order to determine the ecological significance of a WET response, it must be related to the responses and interactions of species, populations, and communities *in situ*. This requires consideration of stressor(s) interactions, and dynamics of exposure (magnitude, frequency, and duration). This dictates the need for a weight-of-evidence approach that describes indigenous community responses, *in situ* exposures, and physical-chemical stressors. More realistic assessments of instream conditions (such as biosurveys of indigenous biota, exposures of caged organisms, or studies of mesocosms) provide essential information that the WET tests cannot (La Point et al., 1996).

III. COMPONENTS OF A HOLISTIC SYSTEM

There is a natural tendency to compartmentalize aquatic ecosystems in routine water quality assessments, only focusing on effluent, ambient water, sediment, or stormwater. This tendency is accentuated by the “media-based” design of most environmental regulatory programs. In addition to this focus on individual media, most water or effluent quality monitoring designs usually consist of a small number of samples, collected from 1 to 4 locations, covering time periods of seconds (one grab) to hours (24 h composite), at a frequency of 1 to 12 times a year. Even with the maximum level of sampling that may be encountered (*i.e.*, monthly, 24 h composites), this would equate to 12 samples that determine the presence or absence of effluent toxicity only 3.3% of the year. In addition to this minimal characterization are the uncertainties of using only one to two species as surrogates of all resident species, the unknown exposure to stress from the other unmeasured media, the fluctuating stressor exposures occurring during high flow or loading events and stressor interactions that occur in aquatic ecosystems. All of these uncertainties and assumptions suggest that a more comprehensive, holistic assessment is needed of which laboratory-based effluent or ambient water column testing is but one component.

There are several reasons why the “water column” species used in WET tests are useful for assessments of sediments. Aquatic organisms rarely exclusively inhabit one media during their life cycle. Many “pelagic” organisms may graze on surficial sediments and even encounter pore waters. For example, the often used “water column” surrogate, the fathead minnow (*Pimephales promelas*) is an omnivore, ingesting a mixture of detritus and invertebrates (Lemke and Bowan, 1998) and frequently feeds on sediment surfaces. The zooplankton, *Daphnia magna*, grazes on surficial sediments in whole sediment toxicity assays. The responses of WET tests have been highly predictive of indigenous benthic community responses at many sites (Dickson et al., 1996; Eagleson et al., 1990). Many vertebrate and invertebrate species have some link to sediments and have been shown to be adversely affected by sediment contamination through toxicity and effects of bioaccumulation (e.g., Baumann and Harshbarger, 1995; Benson and Di Giulio, 1992; Burgess and Scott, 1992; Burton, 1989; Burton, 1991; Burton, 1992ab; Burton, 1995a; Burton, 1999; Burton and Scott, 1992; Burton and Stemmer, 1988; Burton et al., 1987ab; Burton et al., 1989; Burton et al., 1992; Burton et al., 1996; Chapman et al., 1992; Lamberson et al., 1992; Landrum and Burton, 1999; Lee, 1992; Lester and McIntosh, 1994; Ludwig et al., 1993; Mac and Schmitt, 1992; Maruya and Lee, 1998).

Contaminated sediments tend to be the greatest threat to organisms that reside in or on the sediments, or feed either directly on the sediments or on benthivorous organisms. The resuspension of sediments and release of sediment-associated contaminants can be primarily attributed to bioturbation, diffusion, and hydraulics. Standardized effluent or ambient water column assays and other sediment toxicity tests can be used to ascertain whether sediment contaminants are toxic in short-term exposures. However, these relatively short-term assays may not detect high K_{ow} compound effects that are more slowly desorbed and bioaccumulated. Nonetheless, PCB and chlorobenze uptake and toxicity have been observed at low mg/kg concentrations in exposures of less than a week (Burton et al., 1999)..

Fish consumption advisories have been steadily increasing in recent years (USEPA, 1997, 1998). Over 2000 waterbodies had fish advisories in 1996 and most identified sediments as a fish contamination source. It is interesting to note that, based on the huge USEPA water quality criteria toxicity database, benthic and water-column organisms often have similar sensitivity ranges. This observation has supported the justification for using an equilibrium partitioning-based approach for sediment quality guidelines (Di Toro et al., 1991; USEPA, 1989a). Other results have shown water-column organisms to even be more sensitive than benthic species to sediment contamination (Burton et al., 1996; USEPA, 1994b).

The release of the USEPA Contaminated Sediment Management Strategy and Sediment Quality Inventory compiled the limited sediment data (only 4% of monitored sites had toxicity data) and stated that adverse effects are probable from sediments at 26% (>5000) of sites surveyed (USEPA, 1997). A recent random survey of sediments in North Carolina’s estuaries found from 19 to 36% had

contaminant levels known to cause toxicity and 13% had few to no living organisms (Pelly, 1999). These areas are dominated by agricultural watershed inputs. The paucity of sediment toxicity information and the focus of past sediment surveys on industrialized waterways raises the question of whether the extent of sediment contamination is actually much greater than envisioned.

Another essential stressor compartment of the aquatic ecosystem that must be considered in any water quality assessment is that of stormwater runoff. Nonpoint Source Pollution (NPS) is estimated to degrade more than half of the waterways of the United States (Anon., 1996), with other estimates ranging from 30 to 76% (ASIWPCA, 1984; Iivari, 1992). Moderate flows following wet weather events account for the majority of the loading in most waterways (Pitt et al., 1999). The primary loading of nutrients, solids, and anthropogenic chemicals originate from nonpoint sources (Anon., 1996). The dynamics of stream biota are tied closely to abiotic factors (Power et al., 1988) and strongly affect species richness, nutrient cycling, and decomposition processes (Minshall, 1988; Pringel et al., 1988; Resh et al., 1988). Aquatic ecosystems are open nonequilibrium systems in which the frequency and magnitude of disturbance events cannot be predicted (Carpenter et al., 1985; Pringel, 1988; Resh et al., 1988). Despite these realities, water quality monitoring assessments (physicochemical or biological) rarely characterize the role of high flows as either a source of stressors or as a loading component. Rather, permit limits and exceedances are more closely tied to periodic low flow conditions (e.g., 7Q10).

In general, monitoring of urban stormwater runoff has indicated that the biological integrity, and the beneficial uses of urban receiving waters are often affected by habitat destruction and long-term pollutant exposures (especially to macroinvertebrates via contaminated sediment). Documented effects associated with acute exposures of toxicants in the water column are being reported with increasing frequency. As seen with the recent increase in fish consumption advisories, a lack of data does not necessarily imply a lack of contamination. The primary stressors associated with most NPS runoff events include stream power, biochemical oxygen demand (BOD), suspended solids, ammonia, metals, and synthetic organic chemicals (Burton, 1994; Burton, 1995b; Burton and Pitt, 2000; Horner, 1991; Horner et al., 1994; Pitt, 1995; Pitt et al., 1995; Pitt et al., 1999). The levels and exposure of pollutants and toxicity varies orders of magnitude over brief periods (Hall and Anderson, 1988; Katznelson et al., 1995; Mancini and Plummer, 1986). A myriad of potential stressor combinations are possible in waters that receive significant NPS pollutant loadings. In the laboratory, it would be impossible to evaluate even a small number of all combinations of stressors, varying the magnitude, frequency, and duration of each stressor. Therefore, predictive modeling of NPS-related toxicity in receiving waters will be difficult to validate from a single chemical and total maximum daily load (TMDL) perspective. These realities require that innovative *in situ* approaches integrated with traditional methods for measuring and regulating effects be attempted to reduce the uncertainties of current approaches.

IV. STORMWATER QUALITY: RECEIVING WATER IMPACTS

Stormwater runoff is a major cause of receiving water quality degradation (Burton and Pitt, 2000). The effects are most severe for receiving waters draining heavily urbanized watersheds (Horner, 1991; Horner et al., 1994; Pitt, 1995). However, some studies have shown important aquatic life impacts for streams in watersheds that are less than ten percent urbanized where agriculture predominates (Kuivila and Foe, 1995). A wealth of literature exists documenting a strong relationship between degree of urban and agricultural runoff and degradation of aquatic life (Benke et al., 1981; Cook et al., 1983; CTA, Inc., 1983; Dreher, 1997; Ebbert et al., 1983; Ehrenfeld and Schneider, 1983; Garie and McIntosh, 1986; Gast et al., 1990; Handova et al., 1996; Heaney and Huber, 1984; Heaney et al., 1980; Klein, 1979; Lenet and Eagleson, 1981; Lenet et al., 1981; Maltby et al., 1995ab; Masterson and Bannerman, 1994; Moore and Burton, 1999; Mulliss et al., 1996; Pedersen, 1981; Perkins, 1982; Pitt and Bissonnette, 1983; Pitt and Bozeman, 1982; Pratt et al., 1981; Richey, 1982; Richey et al., 1981; Schueler, 1996; Scott et al., 1982; Spawn et al., 1997; Stein et al., 1995; Tucker and Burton, 1999; Weaver and Garman, 1994; Willemsen et al., 1990). However, most of these studies were not comprehensive and simply measured indigenous biological communities and related degradation to urban storm flows. Runoff impacts were found to most likely to be associated with small- to moderate-sized receiving waters, while most of the existing water quality monitoring information exists for larger bodies of water (Heaney et al., 1980). A study of over 40 northeastern Illinois small to moderate-sized streams and rivers found that nearly all streams in urban and suburban watersheds having population densities greater than about 300 people per square mile showed signs of considerable impairment to their fish communities (being in fair to very poor condition) (Dreher, 1997). Acute toxicity to *Daphnia pulex* showed the following land use relationships: commercial > industrial > residential > open space (Hall and Anderson, 1988).

A number of water-quality characteristics dominate as stressors in stormwater runoff (e.g., suspended solids) and must be considered in the use of standardized laboratory toxicity test methods. These characteristics include low dissolved oxygen (e.g., Heaney et al., 1980; Keefer et al., 1979; Lammersen, 1996; Seidel et al., 1996), high turbidity and pathogens (Bolstad and Swank, 1997; Pitt and Bozeman, 1982), ammonia (Widera and Podraza, 1996), bioavailable metals, pesticides and polycyclic aromatic hydrocarbons (PAHs) (Boudries et al., 1996; Estebe et al., 1996; Field and Cibik, 1980; Handova et al., 1996; Kuivila and Foe, 1995; Maltby et al., 1995ab; Morrison et al., 1993; Mulliss et al., 1996) and flow (Borchardt and Sperling, 1997). Recent studies have detected the highly toxic organophosphate diazinon in virtually 100% of stormwaters at levels ranging from 0.5 to 5 µg/L, and was acutely toxic to *C. dubia* (Connor, 1995; Schueler, 1995; Waller et al., 1995). Chlorpyrifos was acutely toxic in several runoff samples at ng/L levels (Connor, 1995; Vlaming et al., 2000). Another problem chemical in stormwater is zinc,

particularly in commercial and industrial areas. Concentrations during wet weather events were often above toxicity threshold levels in a Fort Worth, Texas, survey, but were highly variable (Waller et al., 1995). The primary source of zinc appears to be galvanized metals, with roof gutters producing highly toxic runoff (Pitt, 1995; Pitt et al., 1995). Amphipod uptake of PAHs from sediment extracts in urban waterways was directly related to exposure and sediment manipulation identified hydrocarbons, Cu and Zn as potential toxicants (Maltby et al., 1995b).

The stream habitat itself (e.g., refugial space, bed stability) was seen to play a major role in the degree of effect and stressor interaction (Borchardt and Statzner, 1990). Suspended sediment and depressed dissolved oxygen concentrations produced strong synergistic effects in some fish species, as did contaminants and temperature (Burton and Rowland, 1999; Cairns et al., 1978; Horner et al., 1994; Moore and Burton, 1999), none of which would be predicted in traditional stormwater quality or standardized toxicity assessments.

Long-term biological impacts in receiving waters affected by stormwater must also be considered. Snodgrass et al. (1998) reported that ecological responses to watershed changes may take between 5 and 10 years to equilibrate. Therefore, receiving water investigations conducted soon after disturbances or mitigation may not accurately reflect the long-term conditions that will eventually occur. They found that the first changes due to urbanization will be to stream and groundwater hydrology, followed by fluvial morphology, then water quality, and finally the aquatic ecosystem. They also reported that it is not possible to predict biological responses from stream habitat changes or conditions, although habitat changes may be the most severe stressor in urban waterways.

The NPS loading of contaminants is highest during storm events and occurs as both dissolved and suspended solids fractions. For this reason, downstream depositional zones will tend to accumulate contaminants that may result in chronic exposures to contaminated sediment. Contaminated sediments have often been linked to point sources; however, nonpoint sources are likely a greater source of contamination (as discussed above). So, investigations of stormwater contamination should always be linked to assessments of sediment quality.

Relationships between observed receiving water biological effects and possible causes have been especially difficult to identify, let alone quantify. It is expected that all of these above stressors are problems, but their relative importance varies greatly depending on the watershed and receiving water conditions.

A. Water-Quality Criteria Comparisons

The results of stormwater quality analyses have commonly been compared to water quality criteria in order to identify potentially toxic waters, and likely problematic pollutants. This has led to numerous problems with the interpretation of the data, especially concerning the “availability” of the toxicants to receiving

water organisms and the exposure durations in receiving waters. The water quality of stormwater, or of ambient waters immediately following high flow events, has been shown to be degraded in many studies with chemical concentrations, which may exceed toxicity thresholds (e.g., Horner et al., 1994; Makepeace et al., 1995; Morrison et al., 1993; Waller et al., 1995). Stormwater toxicants are primarily associated with particulate fractions and are typically assumed to be “unavailable”. Typically short and intermittent runoff events can also not be easily compared to the “long” duration criteria or standards. Chemical analyses, without biological analyses, would have underestimated the severity of the problems because the water column quality varied rapidly, while the major problems were associated with sediment quality and effects on macroinvertebrates (Lenet and Eagleson, 1981; Lenet et al., 1981).

To address magnitude and duration issues, the USEPA developed “Criterion Maximum Concentration” with an exposure period assumption of 1 h and “Criterion Continuous Concentration” with an averaging period assumption of 4 days. Yet, these assumptions do not accurately describe most wet weather runoff exposures. Tests with pentachloroethane (Erickson et al., 1989; Erickson et al., 1991) showed that with intermittent exposures, higher pulse concentrations were needed to affect growth, and when averaged over the entire test, effects were elicited at concentrations lower than when under constant exposure. The simplest toxicity model (with first-order, single-compartment toxicokinetics and a fixed lethal threshold) could not completely describe the data. Erickson et al. (1989) concluded that kinetic models that predict mortality were reasonable; however, chronic toxicity effects were much more complicated and no adequate models existed. Hickie et al. (1995) describe a one compartment first-order kinetics, pulse exposure model for residue-based toxicity of pentachlorophenol to *P. promelas*. Pulse exposures were of 2 min to 24 h with durations of 2 to 24 h repeated 2 to 15 times. A comparison of three models (Cxt, Mancini, Breck 3-dimensional range repair) showed reasonable prediction of fish toxicity following 1 to 4 monochloramine pulses (2 h pulse, 22 h recovery). However, predictive capability decreased with greater than four pulses (Meyer et al., 1995). Beck et al. (1991) examined the transient nature of receiving water effects associated with stormwater, stressing the weaknesses associated with more typical steady-state approaches. They felt that there were still major misconceptions associated with modeling these effects.

V. ASSESSING STORMWATER TOXICITY WITH TRADITIONAL TOXICITY TESTS

A. General Applications

Traditional toxicity testing (e.g., WET) testing has been shown to be useful for evaluating stormwaters. The use of toxicity tests on stormwater and receiving

waters, especially *in situ* and side-stream tests that also reflect changing conditions for extended periods, have added greatly to our knowledge of toxicant problems associated with stormwater. While some stormwaters may not be toxic, there is a large body of evidence that suggests many are toxic. Laboratory testing of runoff samples has shown acute and chronic toxicity to a variety of species (Bailey et al., 2000; Connor, 1995; Cook et al., 1995; Dickerson et al., 1996; Hatch and Burton, 1999; Ireland et al., 1996; Katznelson et al., 1995; Kuivila and Foe, 1995; McCahon and Pascoe, 1990; McCahon and Pascoe, 1991; McCahon et al., 1990; McCahon et al., 1991; Medeiros and Coler, 1982; Medeiros et al., 1984; Mote Marine Laboratory, 1984; Tucker and Burton, 1999; Werner et al., 2000; Vlaming et al., 2000). Pesticide pulses have been followed through watersheds, remaining toxic for days from agricultural runoff (Kuivila and Foe, 1995; Werner et al., 2000). Diazinon has been implicated as the primary toxicant in runoff causing acute toxicity to *C. dubia*, *P. promelas*, and *in situ* *Corbicula fluminea* assays (Bailey et al., 1997; Kuivila and Foe, 1995; Connor, 1995; Waller et al., 1995; Cooke et al., 1995). *C. dubia* reproduction and growth of *C. fluminea in situ* closely paralleled the health of the indigenous communities (Dickson et al., 1992; Waller et al., 1995). A simulation of farm waste effluent (increased ammonia and reduced dissolved oxygen) found amphipod precopula disruption to be the most sensitive indicator of stress (McCahon et al., 1991). Mortality only occurred when D.O. fell to 1 to 2 mg/L and feeding rates recovered after exposure to ammonia (5 to 7 mg/L) ended. Elevations of major ion concentrations were toxic to *C. dubia* and *P. promelas* in some irrigation drainage waters (Dickerson et al., 1996).

Toxicity may also be reduced in runoff. When turbidity increased during high flow, photo-induced toxicity of PAHs was reduced *in situ*, when compared with base flow conditions (Ireland et al., 1996). A recent study of the chronic toxicity of fenoxycarb to *Daphnia magna* showed a realistic single pulse exposure resulted in a MATC of 26 µg/L, as compared to 0.0016 µg/L from a standard, constant exposure study (Hosmer et al., 1998).

WET tests have also been used to evaluate the toxicity of effluents from stormwater runoff treatment systems. An evaluation of an urban runoff treatment marsh found strong relationships between *C. dubia* time-to-death, conductivity, and storm size, and time from storm flow initiation (Katznelson et al., 1995). Airport runoff containing glycol-based deicer/anti-icer mixtures was toxic to *P. promelas* and *D. magna* during high use winter months; however, during summer months runoff toxicity only coincided with fuel spills (Fisher et al., 1995). Anti-icer was more toxic to *P. promelas*, *D. magna*, *D. pulex* and *C. dubia* than deicer. Additives were more toxic than glycols (Hartwell et al., 1995). Stormwater detention ponds reduced *P. promelas* and Microtox™ toxicity 50 to 90% when particles greater than 5 µm were removed (Crunkilton et al., 1997; Pitt et al., 1999).

Medeiros and Coler (1982) and Medeiros et al. (1984) used a combination of laboratory and field studies to investigate the effects of urban runoff on fathead minnows. Hatchability, survival, and growth were assessed in the laboratory in

flow-through and static bioassay tests. Growth was reduced to one-half of the control growth rates at 60 % dilutions of urban runoff. The observed effects were believed to be associated with a combination of toxicants.

B. Pulse Exposures

Seasonal pulses of toxicity (e.g., metals in snow melt), observed during extended wet weather conditions, may be reflected in benthic communities (Clementa, 1994) and likely detected in traditional laboratory toxicity tests (Liess, 1996; Crunkilton et al., 1997; Tucker and Burton, 1999). However, localized stormwater events may only produce short-term exposures (minutes to hours) to toxicants and therefore are more difficult to assess (Burton and Pitt, 2000).

Some have suggested that relatively short periods of exposures to the toxicant concentrations in stormwater are not sufficient to produce the receiving water effects that are evident in urban receiving waters, especially considering the relatively large portion of the toxicants that are associated with particulates (Lee and Jones-Lee, 1995ab, 1996). Lee and Jones-Lee (1995b) suggest that the biological problems evident in urban receiving waters are mostly associated with illegal discharges and that the sediment bound toxicants are of little risk. This opinion, however, is not supported by field studies. Others have found sediments to be frequently contaminated at toxic levels (Burton and Pitt, 2000; Burton and Moore, 1999; EPA, 1997). Mancini and Plummer (1986) have long been advocates of numeric water quality standards for stormwater that reflect the partitioning of the toxicants and the short periods of exposure during rains. Unfortunately, this approach attempts to isolate individual runoff events and does not consider the accumulative adverse effects caused by the frequent exposures of receiving water organisms to stormwater (Davies, 1986; Davies, 1991; Davies, 1995; Herricks, 1995; Herricks et al., 1996).

A growing preponderance of data, however, is showing that toxicity is commonly observed during stormwater runoff events and that short-term pulse exposures can be more toxic than long-term continuous exposures (e.g., Brent and Herricks, 1998; Crunkilton et al., 1997; Curtis et al., 1985). Short pulse exposures in stormwater produced lethality several days to weeks later (Abel, 1980; Bascombe et al., 1980; Bascombe et al., 1989; Brent and Herricks, 1998; Ellis et al., 1992; Liess, 1996). Some of this apparent response delay may be a result of uptake and accumulation kinetics (Bascombe et al., 1989; Bascombe et al., 1990; Borgmann and Norwood, 1995; Borgmann et al., 1993). Recent investigations have identified acute toxicity problems and the importance of an adequate post-exposure observation period in side-stream studies with *P. promelas* in urban streams (Crunkilton et al., 1997), and in laboratory spiking studies (Cd, Zn, phenol) with *Ceriodaphnia dubia*, *Pimephales promelas*, and *Hyalella azteca* (Brent and Herricks, 1998; Van Der Hoeven and Gerritsen, 1997). Other laboratory studies have also shown acute

and chronic toxicity of short-term exposures using fish and amphipods exposed to chloroamines, metals, and pesticides (Abel, 1980; Abel and Gardner, 1986; Holdway et al., 1994; Jarvinen et al., 1988ab; McCahon and Pascoe, 1991; Meyer et al., 1995; Parsons and Surgeoneer, 1991ab; Pascoe and Shazili, 1986). In general, it appears that exposure to higher concentrations of toxicants for brief time periods is more important than exposure to lower concentrations for longer time periods (Brent and Herricks, 1998; Liess, 1996; McCahon and Pascoe, 1990; Meyer et al., 1995). However, increased amphipod depuration or metallothionein induction in the presence of Zn allowed greater tolerance (Borgmann and Norwood 1995; Brent and Herricks 1998).

Griffin et al. (1991) state that traditional toxicity testing is inappropriate for time-scale studies of runoff effects due to the exposure design of constant toxicant concentrations. Even the traditional exposure time used in toxicity tests may be inadequate to predict long-term effects. Lifetime *C. dubia* reproduction was unrelated to water quality conditions and more related to food-related factors (Stewart and Konetsky, 1998). This suggests assumptions of the short-term chronic toxicity tests may be questionable in some situations.

Several other studies have shown that fluctuating pulse exposures produce greater uptake and toxicity than continuous exposures, the magnitude of which were dependent on interactions with other stressors (Abel, 1980; Abel and Gardner, 1986; Brent and Herricks, 1998; Borchardt and Statzner, 1990; Curtis et al., 1985; Holdway and Dixon et al., 1986; Ingersoll and Winner et al., 1982; Jarvinen et al., 1988ab; Kallander et al., 1997; Mancini and Plummer, 1986; Siddens et al., 1986; Siem et al., 1984; Thurston et al., 1981), thus pointing to the inadequacy of current water quality criteria. Several of these studies showed significant toxicity effects occurring from exposures of 0.25 to 5 h that equated to continuous LC₅₀ level effects.

However, not all pulsed exposures are more toxic. If there is adequate time for organism recovery between pulsed exposures to toxicants, then the effects of the pulsed exposure of some toxicants are diminished (Brent and Herricks, 1998; Kallander et al., 1997; Mancini, 1983; Wang and Hanson, 1985). This difference may be attributed to the mechanism of toxicity. For example, organophosphates are relatively irreversible inhibitors of acetylcholinesterase (AChE), while carbamate inhibition may be reversible (Kuhr and Dorough, 1976; Matsumura, 1985). So little difference is observed between continual exposures and pulsed exposures (Kallander et al., 1997). Trout were observed to acclimate to ammonia if pulsed exposures were below their toxicity threshold (Thurston et al., 1981). Fenoxycarb was four orders of magnitude less toxic in a single pulsed exposure to *Daphnia magna* as compared to a standard WET exposure (Hosmer et al., 1998). Complicating predictions of effects are synergistic interactions that occur between some contaminants such as pesticides and metals (Forget et al., 1999) and between herbicides and insecticides (Pape-Lindstrom and Lydy, 1997). Organisms recovered to varying degrees given adequate time in clean water following pulsed exposures to phenol, permethrin, fenitrothion, and carbamates (Brent and Herricks,

1998; Green et al., 1988; Kallander et al., 1997; Kuhr and Dorough, 1976; Parsons and Surgeoneer, 1991ab).

Fluctuating pulse exposure issues carry particular significance in the assessment of pesticide (agricultural) and urban runoff. It is apparent that risk from brief toxicant exposure cannot be adequately predicted from standard continuous exposures (e.g., Abel, 1980; Anderson and Shubat, 1984; Hosmer et al., 1998; Jarvinen et al., 1988ab; Kallander et al., 1997; Kleiner and Anderson, 1984; Thurston et al., 1981). Toxicity testing in single events may not be predictive of long-term effects in receiving waters. It was concluded that multiple-event analyses provides necessary information of sources and variability of toxicity that is needed for many aspects of watershed management programs (Herricks et al., 1994; Herricks et al., 1997).

C. Toxicity Identification Evaluations (TIE)

After toxicity is identified in receiving waters, researchers commonly attempt to identify the toxicants responsible for the observed effects through TIE studies. Diazinon was shown to be the primary toxicant in stormwater samples using *C. dubia* (Ohio EPA, 1987; Bailey et al., 2000). Anderson et al. (1991) compared numerous stormwater outfalls in the lower San Francisco Bay, California. They found that non-polar compounds in the most toxic stormwater found (from a small heavily industrialized drainage area) were the most important components of the toxicity, with lesser effects associated with suspended solids, metal chelates, and cationic metals. In another study, stormwater (from large parking areas surrounding an airport and industry) toxicity was most strongly influenced by cationic metals. Diazinon and chlorpyrifos in urban stormwater showed additive toxicity to *C. dubia* in a TIE (Bailey et al., 1997).

Jirik et al. (1998) also used selected phase 1 TIE studies to identify the toxicants most responsible for stormwater toxicity in the Santa Monica Bay area. Sea urchin fertilization tests indicated EC_{50} values of stormwater of about 12 to 20%. Santa Monica Bay receiving waters were also found to be toxic, with the level of toxicity generally corresponding to the amount of stormwater in the receiving water. EDTA addition removed virtually all of the toxicity, implying that divalent metals were the likely toxicant component. Spiking studies showed that zinc, and sometimes copper, were the most likely metallic toxicants. Further studies using EDTA vs. sodium thiosulfate for toxicity removal also strongly implicated zinc as the likely cause of toxicity.

D. In Situ Methods

It is apparent in some situations that the complex exposure dynamics and interactions of stormwaters cannot be mimicked in the laboratory. By exposing

standard test species *in situ*, exposures are more realistic. *In situ* testing using caged organisms has been shown to be an effective monitoring tool. Numerous studies have demonstrated the approach in studies of runoff, base flow, and sediments (e.g., Burton, 1999; Burton and Rowland, 1999; Burton et al., 1996; Chappie and Burton, 1999). Studies of marine systems have primarily used mussels (Salazar and Salazar, 1997) with limited testing of amphipods (DeWitt et al., 1999; Fleming et al., 1997). Freshwater studies have consisted of a wide range of organisms, such as fish, cladocerans, amphipods, midges, bivalves, mayflies, hydra, bryozoa, and oligochaetes (e.g., Brooker and Burton, 1998; Burton and Rowland, 1999; Burton et al., 1996; Hatch and Burton, 1999; Ireland et al., 1996; Lavoie and Burton, 1998; Liess, 1996; Moore and Burton, 1999; Morgan et al., 1981; Morgan et al., 1986; Rowland et al., 1997; Sasson-Brickson and Burton, 1991; Schulz, 1996; Tucker and Burton, 1999; Waller et al., 1995). Exposure periods range from 48 h to weeks. Measurement endpoints range from lethality to sublethal biomarkers and tissue residues. Toxicity has been observed to increase and decrease during high flow events using *in situ* studies (Connor, 1995; Hatch and Burton, 1999; Ireland et al., 1996; Moore and Burton, 1999; Tucker and Burton, 1999) and better revealed which stressors were dominating, for example, suspended solids, flow, photo-induced toxicity of PAHs, PCBs, sediments. More specifically, *in situ* toxicity tests in receiving waters (Burton et al., 2000; Greenberg et al., 2000; Ireland et al., 1996; Moore and Burton, 1999; Sasson-Brickson and Burton, 1991; Stemmer et al., 1990; Tucker and Burton, 1999) have illustrated the direct toxic effects associated with exposure to contaminated sediments, stormwaters, and suspended solids. Exposures *in situ* are obviously different from those in traditional bioassays, so responses often differ between the two when compared (Sasson-Brickson and Burton, 1991; Tucker and Burton, 1999).

A variety of automated *in situ* response systems have been used in natural waters (e.g., Morgan et al., 1981, 1986; Sloof, 1979; Sloof et al., 1983). Recently, the sublethal responses of bivalve gape has been used as a continuous monitor of water quality *in situ* (Allen et al., 1996; Borcharding, 1992; Herricks et al., 1997; Sloof et al., 1983; Waller et al., 1995). Bivalves react to poor water quality conditions by closing their shells. The opening and closure of their shells (or gape) can be monitored by attaching a proximity electronic sensor to the outer shell surface. This provides method for biologically monitoring water water on a real-time basis through the use of telemetry methods (Allen et al., 1996; Borcharding, 1992; Herricks et al., 1997; Sloof et al., 1983; Waller et al., 1995). Each bivalve will have a unique gape response signature and the current challenge is to statistically characterize these response patterns and determine when significant water quality impairment is occurring. Another promising method, on-site toxicity testing, was conducted with side-stream flow systems using several species, lab and field biological assessments, and chemical measurements (Burton and Rowland, 1999; Crunkilton et al., 1997). Toxicity varied through time and ranged from acute to chronic effects, some of which peaked 25 days after exposure. The biological

and physical habitat assessments also supported a definitive relationship between degraded stream ecology and urban runoff (Crunkilton et al., 1997). Similarities were observed between side-stream and *in situ* toxicity response patterns between sample stations. Elevated temperatures accentuated site water and sediment toxicity (Brooker and Burton, 1998; Burton and Rowland, 1999; Lavoie and Burton, 1998). Sublethal indicators of toxicity have also been used. DNA strand length in the Asian clam was found to be a very sensitive indicator of stormwater contamination (Black and Belin, 1998).

VI. SEDIMENT QUALITY: TOXICITY ISSUES

Single species toxicity testing with sediments has been conducted with increasing frequency since the 1970s. Testing has involved a wide range of organisms (microbial to amphibian) covering a wide range of trophic levels (Burton 1991). Many of the sediment contamination assessments have shown water column species to be sensitive and effective tools (Burton, 1991; Burton, 1992b; Burton et al., 1996; Burgess and Scott, 1992; Carr et al., 1996; Kemble et al., 1994; Padma et al., 1998). While responses measured generally have focused on lethality and growth, more sensitive sublethal effects are the focus of more recent studies (e.g., Burton, 1991, Day et al., 1997; Ingersoll et al., 1998). Standard methods exist for several marine and freshwater species (ASTM 1995a-c; USEPA 1994c-d). Test phases have included whole sediments, pore waters, and elutriate phases primarily (Burton, 1991).

Pore waters have been shown to be a dominant exposure pathway of sediment contaminants to many benthic invertebrates (Di Toro et al., 1991). Some have suggested that pore water is a reasonable surrogate test fraction for whole sediments (Carr and Chapman, 1995; Giesy and Hoke, 1989). However, others have disagreed, pointing to the importance of sediment and overlying water consumption (e.g., Hare and Shooner, 1995; Lee et al., 2000). Pore water toxicity evaluations with WET test methods have shown relationships with benthic communities (Ankley et al., 1992). A three-phase partitioning model predicts the distribution of hydrophobic chemicals between sediment organic matter, pore water dissolved organic carbon, and freely dissolved aqueous phases (Mitra and Dickhutt, 1999); however, a large fraction of the most nonpolar high-molecular-weight organics present in pore water are colloidally bound and not truly dissolved (Burgess et al., 1996). In addition, the partitioning of hydrophobic chemicals into pore water has been shown to increase when anoxic pore waters are oxidized, allowing for greater bioavailability (Hunchak-Kariouk et al., 1997). The sediment collection process has also been shown to increase ammonia concentrations in pore water (Sarda and Burton, 1995). These factors have tremendous implications for the interpretation of WET and TIE test results that evaluate pore waters. The commonly used methods of pore water collection and testing may significantly decrease or increase

organism exposure by allowing exposure to colloid and altering bioavailability due to sediment disruption and/or oxidation. These issues suggest the role of pore water exposure and uptake *in situ* may be less than many have suggested and difficult to assess accurately.

Several studies have attempted to determine dose-response relationships of contaminants either with field collected contaminated samples or through spiking (dosing) at multiple concentrations (Giesy et al., 1988; Nelson et al., 1993). This has proven to be quite difficult, with nonlinear responses frequently resulting no matter which dilution method is used (Nelson et al., 1993). This is likely due to the disruption of contaminant partitioning to sediments and colloidal materials with the introduction of the clean diluent (whether it is water or sediment).

Toxicity testing of sediments has several advantages that chemical characterization and biosurveys do not. Toxicity testing provides unique information on whether adverse levels of chemicals are bioavailable. Toxicity testing is unaffected by habitat or natural disturbances such as high flows, temperature, or turbidity or D.O. sags. Testing is relatively inexpensive, can be conducted with less expertise, and can be conducted throughout the year. The results are relatively easy to interpret, particularly if the responses of the test organisms are severe and occur in multiple species. There are numerous publications that have shown WET tests to be predictive of benthic macroinvertebrate effects (e.g., reviews in Grothe et al., 1996; Ingersoll et al., 1997). Benthic macroinvertebrates are typically exposed to both sediments and overlying waters. Most of the studies showing benthic community and WET test response correlations did not focus on sediment contamination; however, undoubtedly it was a factor at many sites. WET and other sediment toxicity tests results have been “validated” by some and shown to be predictive of population and community-level responses (Canfield et al., 1994, 1996; Clements, 1997; DeWitt et al., 1992; Giesy et al., 1988; Hickey and Clements, 1998; Maltby and Crane, 1994; Swartz et al., 1985, 1994; Wentzel et al., 1977a-b, 1978). However, as discussed above, WET tests have been shown to be both predictive and nonpredictive of benthic effects.

It is also important to be aware of the uncertainties of sediment toxicity testing (Day et al., 1997; Ingersoll et al., 1997; Solomon et al., 1997). Sublethal effects or subtle interactions that are not measured in traditional short-term sediment or standardized water toxicity tests may occur even if toxicity is not detected (Luoma, 1995; Schindler, 1987). But sublethal “biomarker” responses have a substantial degree of uncertainty when it comes to predicting significant ecological impacts (Benson and Di Giulio, 1992; Luoma, 1995; Schindler, 1987). Sample collection and manipulation may produce artifacts that either increase or decrease toxicity, thereby leading to false positive- or -negative results (Burton, 1991). The principal sampling and testing artifacts that may decrease the accuracy of the results include: oxidation of sediments altering metal availability; desorption of contaminants increasing availability; initial increase in ammonia concentrations; mixing of vertical gradients altering contaminant exposure; nonequilibrium conditions; re-

removal of large organic material via sieving thereby altering exposure; increased predation; and/or alteration of exposure due to overlying water renewal rates. In addition, these sediment toxicity assays have some of the same limitations as identified above for WET testing and others, including laboratory extrapolation to population/community effects; exposure conditions; unknown toxicokinetics and influence of natural factors (e.g., organic matter, grain size, salinity) and stressors (e.g., food availability, predators, flow, temperature, UV), spatial heterogeneity (patchiness), errors in exposure and contaminant fate and transport models; and inability to evaluate indirect effects (e.g., Burton, 1991; Burton et al., 1996; Day et al., 1997; Hulbert, 1975; Solomon et al., 1997).

While there are many uncertainties, their impact can and has been minimized allowing for effective use of toxicity testing to evaluate both sediments and stormwaters. Ingersoll et al., (1997) weighted the uncertainty associated with the various measurement endpoints and test phases of sediment toxicity tests and are discussed in detail in other reviews (Burton, 1991).

VII. UNIQUE SEDIMENT ISSUES AND METHOD MODIFICATIONS

WET testing has been widely used and proven to be an effective environmental regulatory tool for protecting water quality. A very simplistic modification of the standardized WET test (adding a few milliliters of sediment to the test beaker) results in a very sensitive assay for sediment contamination, as evidenced by numerous studies using *C. dubia*, *D. magna*, and *P. promelas* (ASTM, 1995a; Burton, 1991; Burton and Stemmer, 1988; Burton et al., 1987ab; Burton et al., 1989). A massive comparison of all peer-reviewed sediment toxicity test methods (n = 24) at three "Areas of Concern" in the Great Lakes showed "water-column" toxicity test organisms (*C. dubia*, *D. magna*, and *P. promelas* 7-day short-term chronic toxicity tests) to be among the most sensitive to sediment contamination (Burton et al., 1996). A test battery consisting of representative species from four different response pattern groups were recommended to best detect sediment toxicity. The primary species from these groups included *C. dubia*, *D. magna*, *P. promelas*, *H. azteca*, *C. riparius*, *C. tentans*, *Hexagenia bilineata*, *Diporeia*, *Hydrilla verticillata*, or *Lemna minor*. So it is rather surprising that this simple method modification has not been utilized by the USEPA at sites where WET testing is already being used.

Recent assessments of contaminated sediments demonstrated why both laboratory and field toxicity exposures were essential to adequately identify key stressors and characterize exposure dynamics (Ireland et al., 1996; Sasson-Brickson and Burton, 1991; Stemmer et al., 1990; Burton et al., 1999; Burton et al., 2000; Greenberg et al., 2000). Sediment-associated toxicity increased in the laboratory exposure of *P. promelas*, *C. dubia*, *D. magna*, and *H. azteca* when compared with *in situ* exposures, whereas toxicity decreased in overlying waters. Photoinduced

toxicity from PAH and UV interactions and sampling-induced artifacts accounted for these laboratory to field differences. Toxicity was also reduced significantly in the presence of UV when the organic fraction of the stormwater was removed. Photo-induced toxicity occurred frequently during low flow conditions, but was reduced during high turbidity associated with high flow conditions. Toxicity was also higher in sediment or overlying waters near the contaminated sediment surface as opposed to waters several centimeters above the sediment-water interface.

An elevation in temperature of Des Plaines River water accentuated the toxicity of the water and of sediments using both water column and benthic species (Brooker and Burton, 1998; Burton and Rowland, 1999; Lavoie and Burton, 1998). Responses were replicated in laboratory, *in situ*, and artificial, side-stream exposures. The laboratory exposures helped define exact threshold temperatures, critical exposure times, and interactions with ammonia. Field exposures, on the other hand, better defined fluctuating exposures and interactions with other stressors such as suspended solids and fluctuating temperatures. Conclusions based on laboratory exposures would have underestimated stream effects.

An urban site receiving large loadings of residential, commercial, and industrial storm runoff was assessed using an integrated low and high flow assessment (Moore and Burton, 1999). A survey of sediment quality during base flow conditions found one depositional area where sediments were acutely toxic and contained elevated levels of contaminants. An *in situ* toxicity assessment found that low flow water was not toxic, but high flows were toxic and suspended solids and flow contributed significantly to overall stress. However, indigenous communities appeared to be affected more strongly by contaminated sediments than high flow conditions.

A TIE of pore water from a stormwater detention pond using *C. dubia* 48 h exposures showed ammonia to be the primary toxicant with some effects from metals (Zn, Fe, and Cu). The high level of ammonia may have obscured the metal toxicity (Wenholz and Crunkilton, 1995).

WET testing is particularly useful in dose-response (spiking) studies of sediments and pore waters. These have included objectives such as PAH criteria development (Swartz, 1999), equilibrium partitioning criteria development (Di Toro et al., 1991), partitioning dynamics and bioavailability (Landrum and Burton, 1999; Stemmer et al., 1990), and determination of field effect thresholds (Ankley et al., 1992; Giesy and Hoke, 1989; Swartz, 1999; Swartz and Di Toro, 1997).

Several studies have used WET testing to identify dominant sediment contaminants in TIE type approaches (Ankley and Schubauer-Berigan, 1995; Schubauer-Berigan and Ankley, 1991; Swartz and Di Toro, 1997; USEPA, 1989b, 1991b, 1993b). The TIE methods commonly used were originally designed for effluents (USEPA, 1989b, 1991b, 1993b) and were easily adapted for pore water testing using the same WET test species (Boucher and Watzin, 1999). Through fractionation procedures, various classes of common sediment contaminants (such as cationic metals, nonionic organics, ionics, volatiles, and pH-dependent [e.g., am-

monia] compounds) are separated followed by toxicity testing. However, in some cases the WET test methods are not sensitive enough and toxicity is lost through the fractionation process. Artifacts produced or contaminant interactions preclude confirmation of any toxicant. Newer TIE methods include whole sediment manipulations, exposure to UV (Kosian et al., 1998), or *in situ* exposures with various stressor partitioning methods and substrates (Burton et al., 1998; Greenberg et al., 1998; Burton and Moore, 1999) and may reduce the likelihood of artifacts.

VIII. INTEGRATED APPROACHES FOR SEDIMENTS AND STORMWATERS

The extent of sediment or stormwater pollution and its source(s) cannot be reliably determined in most areas without characterizing toxicity (laboratory and field) and indigenous communities while considering the role of low and high flow conditions. In order to best identify and understand these impacts, it is necessary to include biological monitoring, using a variety of techniques, and sediment quality analyses, in a monitoring program. Water column testing alone has been shown to be very misleading

Field surveys rarely can be used to verify simple single parameter laboratory experiments (Johnson et al., 1996). Watershed approaches integrating numerous databases in conjunction with *in situ* biological observations help examine the effects of many possible causative factors. Significant hydraulic disturbance of aquatic life may occur in watersheds with greater than 2 to 5% impervious areas (Burton and Pitt, 2000). The relative importance of short-term and delayed impacts depended on local conditions and was primarily related to unionized ammonia, oxygen depletion, and shear stress (Borchardt and Statzner, 1990). Recent studies (discussed above) have combined chemical-physical characterizations of water and sediment, with biosurveys and laboratory and *in situ* toxicity surveys (low and high flow) effectively characterized major water column and sediment stressors and their interactions (Burton and Rowland, 1999; Burton et al., 1998, 1999, 2000; Burton and Moore, 1999). Suspended solids, ammonia, sediments, temperature, fluorene, sediment, and/or stormwater runoff were each observed to be primary stressors in these test systems. These primary stressors could not have been identified without low and high flow and sediment quality assessments both in the laboratory and field. It is apparent that in order to determine the role of chemicals as stressors in the receiving waters, the role of other stressors (both natural and anthropogenic) must be assessed under varying stream conditions.

Johnson et al., (1996) and Herricks, et al., (1996, 1997) describe a structured tier testing protocol to assess both short-term and long-term wet weather discharge toxicity. The protocol recognizes that the test systems must be appropriate to the time-scale of exposure during the discharge. Therefore, three time-scale protocols were developed, for intraevent, event, and long-term exposures.

As discussed above, there is now a wealth of literature that documents there is ecologically significant exposure to stressors occurring for short time periods during high flow conditions. Some studies have even shown a diurnal to seasonal flux of metals from sediments during base flow conditions (Brick and Moore, 1996; Von Gunten et al., 1994). This should not be surprising given the role of temperature and light on benthic activity. Given these fluctuations, *in situ* testing using caged organisms provides greater environmental realism than laboratory exposures (Barbour et al., 1996; Burton et al., 1996; Clements and Kiffney, 1996; Dickson et al., 1996; Waller et al., 1995). However, the predictive capability of laboratory-based standard toxicity tests could be improved with well-designed studies that better characterize stressor exposures and benthic community spatial-temporal dynamics, use common (lab vs. field) assessment endpoints, or employ demographic/individual-based models for inferring population-level effects (e.g., Burton et al., 1996; Day et al., 1997)

There is a natural tendency in the popular “weight-of-evidence” or “sediment quality triad”- type approaches to look for “validation” of one assessment tool with another. For example, matching a toxic response in a WET test with that of an impaired community gives a greater weight of evidence. This does not, however, necessarily “validate” the results (or invalidate if there are differences) (Chapman, 1995b). Natural temporal changes in aquatic populations at different sites within a study system need not be the same (Power et al., 1988; Resh et al., 1988; Underwood, 1993); therefore, predictions of effect or no-effect from WET testing of reference sites may be in error. Each monitoring tool (i.e., chemical, physical and indigenous biota characterizations, laboratory and field toxicity, and bioaccumulation) provides unique and often essential information (Burton, 1995b; Chapman et al., 1992). If responses of each of the biological tools disagree, it is likely due to species differences or a differing stressor exposure dynamics/interactions. These critical exposures issues can be characterized through a systematic process of separating stressors and their respective dynamics into low and high flow and sediment compartments using both laboratory and field exposures. Then a more efficient and focused assessment can identify critical stressors and determine their ecological significance with less uncertainty than the more commonly used approaches. The chronic degradation potential of complex ecosystems receiving multiple stressors cannot be adequately evaluated without a comprehensive assessment that characterizes water, sediment, and biological dynamics and their interactions.

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