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velops an exceptionally large dorsal crest when exposed to kairomones released by predatory backswimmers (Notonectidae) (Grant and Bayly 1981). In both species of *Daphnia*, the crest reduces their vulnerability to predation by increasing capture and handling difficulties for the predators.

Predator	Prey	Induced Response
Asplanchna (rotifer)	*Branchionus* (rotifer)	Spines
Daphnia	*Scenedesmus* (algae)	Morphology
Notonecids (backswimmer)	*Daphnia*	Crest, life history, behavior
Chaoborus (phantom midge larvae)	*Daphnia*	Crest, life history, behavior
Fish	*Daphnia*	Life history, behavior
Fish	*Chironomus*	Body size
Dragonfly	Tadpoles	Morphology, color
Lembadion lucens (protozoa)	*Euplotes octocarinatus* (protozoa)	Cell shape and size
Doridella steinbergae (nudibranch)	*Membranipora membranacea* (bryozoan)	Spines

Chemical communication between species became a subject of toxicological interest when Hanazato (1991) reported that *Daphnia* exposed to certain pesticides developed crests that were morphologically identical to the structures induced by kairomones from invertebrate predators. The inducing pesticides included carbaryl, BPMC, temephos, diazinon, fenthrothion, and fenthion. He found no effect with the herbicides thiobencarb and oxadiazon or the fungicide IBP. In his initial experiments, Hanazato found that the pesticides were only effective during a window of development that lasted from late embryogenesis to the second neonatal instar.

The expression of inducible defenses usually carries some form of energetic cost which reduces the maximal potential reproductive rate of the induced organism. Hanazato and Dodson (1992) hypothesized that the addition of a toxic stress may synergistically increase the negative impact of the kairomone. They found evidence of a synergistic increase in toxicity when they co-exposed *D. pulex* to kairomones of *Chaoborus* and carbaryl.

The common factor unifying all the chemicals which Hanazato and his collaborators found could induce crests in *Daphnia* was their mode of action as acetylcholine esterase inhibitors. These chemicals, which belong to the carbamate and organophosphate classes, all act by inhibiting the degradation of the neurotransmitter acetylcholine, therefore increasing stimulation at cholinergic synapses and nerve terminals. Recently, I found that the organochlorine pesticide, endosulfan, could induce and increase crest development in *Daphnia longicephala* (Barry 1998). Endosulfan inhibits the binding of d-aminobutyric acid (GABA) to the GABAA receptor. GABA is normally an inhibitory neurotransmitter that depolarizes membranes by increasing the flux of chloride ions. By inhibiting the binding of GABA to its receptor, endosulfan also stimulates neural transmission. These effects occurred at concentrations of endosulfan at least two orders of magnitude lower than those that caused significant impairment to growth and reproduction. In contrast, high concentrations of endosulfan inhibited crest induction in *D. longicephala*. These findings led me to hypothesize that crest development in *Daphnia* is regulated by the release of target-specific hormones from neuro-endocrine glands for which acetylcholine is the main stimulatory neuro-transmitter and GABA the main inhibitory transmitter. This model was confirmed by exposing *Daphnia* to a range of cholinergic and GABAergic agonists and antagonists, in the presence or absence of the kairomone. I also found that most chemicals could inhibit the expression of inducible defenses at concentrations that caused observable toxicity. This is probably a generalized stress response in *Daphnia* and not a direct endocrine-mediated effect.

The effects of pesticides such as carbaryl and endosulfan on *Daphnia* fit the broader definition of endocrine disruptors, in that they are anthropogenic substances which stimulate (or inhibit) the production of hormones in target organisms. It appears that many chemicals which stimulate or inhibit neural transmission at cholinergic synapses may alter the expression of inducible defenses, at least in *Daphnia*. Chemical communication between planktonic species was recently characterized as a low energy, information rich network which parallels the flow of materials and may help maintain ecosystem structure and function. It remains unclear whether the effects of pesticides on inducible defenses of *Daphnia* are of ecological significance. For these effects to be of major concern, they should occur at pesticide concentrations that are likely to be found in the natural environment and at levels that are below those which directly reduce growth or fecundity. Clearly this is a field that warrants further investigation.

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Setting Water Quality Guidelines—A Statistician's Perspective

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Ecological risk assessment (ERA) has been defined as "the characterization of the adverse ecological effects of environmental exposures to hazards imposed by human activities" (National Research Council, 1993). For statisticians such as myself, the key aspect of this "characterization" is the assessment of *uncertainty*. I would argue that in the absence of uncertainty, there is no risk—

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only deliberate actions and known events having totally predictable consequences.

It is evident that the concept of "risk" means different things to different people. One popular definition is *risk = hazard* x *exposure* although this deterministic formulation overlooks the uncertainty aspect. In the context of environmental monitoring and assessment I define risk to be the *probability* of an adverse outcome.

It is clear that after decades of prosaic comparisons of an environmental concentration of a contaminant with a relevant guideline, the stage is set for assessing environmental data in a fairer and more meaningful way. By this I mean the use of methods which embrace and acknowledge the related concepts of variability and uncertainty, i.e., a more statistical approach. Having already declared my statistical background, this is not a gratuitous call to ensure the ecologically sustainable development of my profession. Indeed, in my statistical quality assurance role for a number of large environmental studies I have often times been critical of an over-reliance on classical statistical inference (that is, general linear models based on normal distribution theory). While the statistical approach will become increasingly prevalent in all aspects of environmental science, we must be careful to ensure that the tools we adopt are (i) relevant; (ii) robust; (iii) fit for the purpose; and (iv) perform better than the conventional methods they are displacing. If these criteria cannot be met then the added complexity of a more sophisticated statistical approach is not only unwarranted, but potentially dangerous.

There are numerous aspects to the risk assessment paradigm, but I will restrict my comments to a few issues that are pertinent to the establishment of water quality guidelines with particular reference to the popular method of Aldenberg and Slob (1993). The reader who is interested in learning more about the role of statistics in setting environmental standards is advised to consult the excellent monograph by Barnett and O'Hagan (1997).

Aldenberg and Slob's (1993) highly statistical method aims to identify threshold levels for contaminant concentrations such that some high fraction of all species will be protected with a high degree of confidence. Notwithstanding the rather convoluted expressions that are generated (eg. 90% of species are protected with 95% confidence), I believe the mechanics of the approach warrant closer inspection from statisticians and ecologists. Aldenberg and Slob have done a good job in raising the awareness of the risk-based framework. A genuine attempt has been made to replace the prosaic comparisons referred to above with methods that are more tolerant of aberrant observations that occur even when no action is required. However, we need to perform a reality check along the lines of points (i) to (iv) above. In this particular case a lack of robustness could be an issue. Myself (and co-workers) have looked at the statistical aspects of the Aldenberg and Slob method in detail. Our concerns with this and related methods are essentially two-fold and these relate to (i) methods used to determine individual NOECs; and (ii) the subsequent statistical modeling of these NOECs.

Concerns have previously been raised in relation to the suitability of NOECs for regulatory purposes (eg. Chapman et al. 1996) and I will not re-visit this issue in detail except to note that the two common methods have inherent difficulties. The "assessment factor" approach relies on a rather arbitrary scaling of other numerically derived quantities such as LC50s and EC50s. The statistical approach determines a NOEC using multiple comparison procedures within an Analysis of Variance (ANOVA) framework. The concern with this method is that the resulting NOEC is constrained to be one of (subjectively chosen) discrete concentrations used in the toxicity experiment. However derived, the NOEC data are the starting point for Aldenberg and Slob's more sophisticated modeling. Unfortunately, no account is taken of the uncertainty associated with these input data and thus the impact on the final results is unknown.

The Aldenberg and Slob method is predicated on the assumption that the population (that is *all* species data) of NOECs is described by a logistic distribution. (The assumed distribution is more correctly a *log*-logistic distribution since the logarithms of NOECs are modeled using a logistic distribution.) While not an unreasonable choice, it should be realized that there are other candidate models that may also provide a reasonable description of such data (for example the Weibull, double exponential, Laplace, extreme-value, and Pareto distributions, just to name a few). It would appear that part of the appeal for Aldenberg and Slob was that the logistic distribution has "some nice mathematical features that make certain calculations relatively easy." While analytical tractability is an important consideration, it should not drive the model selection process—the key is achieving model parsimony. That goodness-of-fit tests, such as the Kolmogorov-Smirnov test, fail to reject the hypothesis of a (*log*-) logistic model for a given sample of NOECs, should not in itself be taken as a validation of the model choice. Given the typically small sample sizes involved in this type of analysis, the power of any goodness-of-fit test will be very low. In such cases it would not be surprising to find other candidate distributions being accepted by the statistical test. This is a worrying aspect of the Aldenberg and Slob method, particularly if different (although equally legitimate) statistical models result in substantial discrepancies in the computed threshold values. It can be shown that the *log*-logistic distribution is a special case of a Burr family of distributions (Burr 1942). Using some NOECs for a range of toxicants in marine and freshwaters myself and colleagues investigated the utility of both the *log*-logistic and Burr distributions. Our rationale was that the Burr distribution should always do as well, if not better than the *log*-logistic by virtue of (a) an additional parameter which would allow greater flexibility in the range of shapes to be fitted; and (b) that the *log*-logistic is one of the Burr cases anyway. In most instances the resulting threshold values were in reasonable agreement, although there were a few instances where the results differed by up to a 3-fold factor. I believe this is sufficiently worrying to warrant a more comprehensive assessment of the methodology lest the results from one particular method become written into legislation.

In conclusion, I believe *contemporary* statistical methods have an important role to play in environmental sciences. However, before we all jump on the risk-assessment bandwagon, a preliminary safety inspection of the vehicle should be performed.

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