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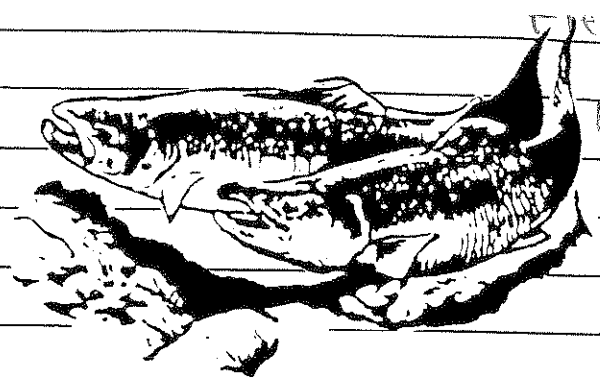

Freshwater Mussel Die-off Attributed to Anticholinesterase Poisoning

In August 1990, a die-off of more than 1,000 freshwater mussels was discovered in Swift Creek, Nash County, North Carolina. Among the dead and moribund mussels of several species, there were about 111 Tar spinymussels (*Elliptio steinstansana*), a federal- and state-listed endangered species. No dead or moribund fish or other vertebrates were found at the die-off site.

Chemical Pollution of the Watershed

Water quality in Swift Creek is generally good. At the time of the die-off, the dissolved oxygen level was 8.4, the pH was 6.64, and river flow was at a yearly low. The die-off occurred in a stream reach dominated by forestry and agriculture, with few industrial or urban discharges. The principal crops in the area were sweet potatoes, cotton, tobacco, corn, and soybeans. The county extension agent reported little use of agricultural chemicals in the vicinity during August. Lannate and Orethene were the two most commonly used insecticides in Nash County during 1990.

Deaths Were Acute

The dead mussels had normal fleshy feet with no evidence of glycogen depletion when examined at the site. Subsequent examination showed that at the time of death, gamete production was ongoing, and organs were of normal size and color. We believe the deaths resulted from acute causes rather than chronic stresses that produce a gradual wasting of body condition. There was no indication of the involvement of infectious agents or parasites.

Analytical Chemistry Results Negative

Mussels of several species, representing living and dead individuals at the time of collection, were analyzed for a variety of contaminants. None of six mussel samples contained detectable concentrations of organophosphorus or carbamate pesticides. A large metal drum located upstream of the die-off contained water and an oily substance that was analyzed for aliphatic hydrocarbons and organophosphorus and carbamate pesticides. Chemical analysis confirmed the presence of oil in the drum, but no organophosphates or carbamate pesticides. None of five mussel samples analyzed harbored biologically significant concentrations of aliphatic hydrocarbons. Samples were not analyzed for metals or organochlorine compounds because they were not suspect chemicals.

Cholinesterase Activity Determined

We determined cholinesterase activity in Eastern elliptio mussels (*Elliptio complanata)* collected upstream (10 km), downstream (7 km), and at the main die-off site in August 1990. We used standard techniques modified only slightly from those used for

diagnosing vertebrate poisonings by cholinesterase-inhibiting pesticides. Adductor muscle proved to have the richest source of cholinesterase activity in the upstream "reference" samples and was used as the tissue of choice for the determinations.

Cholinesterase Activity Was Inhibited at Kill Site

Cholinesterase activity in adductor muscle from specimens collected at the kill site and downstream was depressed 73 and 65%, respectively, compared with reference samples (Figure). The depression is consistent with a diagnosis of anticholinesterase poisoning commonly produced by organophosphate and carbamate pesticides. Confirmation of organophosphate and carbamate poisoning requires identification of these chemicals in tissues or intestinal contents. Analytical chemistry did not detect either pesticide group in the mussels. However, failure to detect these pesticide groups in poisoned, moribund animals is common because organophosphates and carbamates are rapidly metabolized and excreted. Analytical chemistry detection limits (organophosphates = 0.5 ppm; carbamates = 1.0 ppm) applied to these samples were 2 to 5 times above concentrations of pesticides in water that we subsequently have found to significantly inhibit cholinesterase activity in *E. complanata*. We believe it unlikely that analytical chemistry would have detected these compounds in mussel tissues.

We conclude from the cholinesterase determinations, acute nature of the deaths, and absence of other identified causative factors that the mussel die-off was caused by an anticholinesterase agent, most likely an organophosphate or carbamate pesticide. This is the first case reported in which cholinesterase-inhibiting compounds have been implicated in a die-off of natural populations of freshwater mussels.

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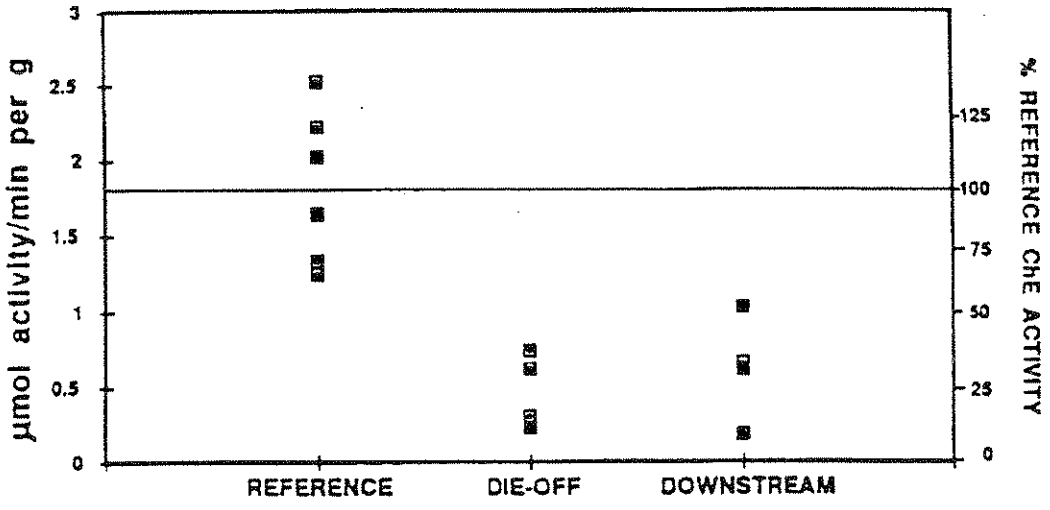

Figure. Cholinesterase activity of adductor muscles of moribund Eastern elliptios (*Elliptio complanata*) collected at the site of an ongoing freshwater mussel die-off in Swift Creek, Nash County, North Carolina, August 1990. Samples were also collected upstream (10 km, reference samples) and 7 km downstream for comparison. Some dead mussels were also found at the downstream site, indicating that the die-off occurred on ≥ 7 km of the creek.