Something's Fishy

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Marine epidemics may signal environmental threats to the immune system

In the North and Baltic Seas over several months in early 1988, some 25,000 harbor seals – about 60 to 70 percent of those residing there – abruptly perished. Subsequent investigation would trace this, the largest seal die-off in history, to a novel germ.

Dutch virologist Albert D.M.E. Osterhaus of Erasmus University in Rotterdam helped identify the source of the lethal epidemic: a distemper virus – one that he describes as "quite similar" to a microbe that can cause serious respiratory, gastrointestinal, and central nervous system problems in dogs.

But having solved one mystery – what killed the seals – he had found another: What had rendered these mammals so vulnerable? Now, he has data that illustrate the unintended injury that can result from industrial activities.

While the seals were dying, Osterhaus recalls, "There was a lot of controversy about pollution being the major cause." And even after his group successfully indicted the viral perpetrator, he says, "We held open the possibility that this particular virus – or its effects – might have been aggravated by preexposure of the animals to certain pollutants."

Had this epidemic been an isolated event, it might have entered the annals of biology as just another curiosity. But in fact, notes Susan D. Shaw, executive director of the New York City-based Marine Environmental Research Institute (MERI), this die-off was but the most tragic manifestation of a worrisome trend.

Over a 5-year period beginning in 1986, green sea turtles began appearing with massive tumors known as fibropapillomas. MERI reports that up to half the world's population of this endangered species may now bear such tumors, which have been linked to infection with a herpeslike virus.

During 1987 and 1988, some 750 bottlenosed dolphins died along beaches from New Jersey to Florida – many having succumbed to pneumonia and severe, exfoliating skin lesions that resembled acid burns. Where tested, the blubber of these animals often contained very high concentrations of chemicals called polychlorinated biphenyls (PCBs).

In 1987, seals in Siberia's Lake Baikal died in large numbers from a distemper virus. A year later, white-sided dolphins experienced a mysterious and lethal epidemic in the waters off Lubec, Maine.

Two years later, the remains of 274 bottlenosed dolphins were recovered along the U.S. shores of the Gulf of Mexico – many of them covered in a mysterious fungal growth. Between 1990 and 1992, more than 1,000 Mediterranean striped dolphins succumbed to an infection resembling the one that devastated the harbor seals in 1988. High concentrations of PCBs turned up in many of the dolphin carcasses examined – both in the Gulf and the Mediterranean.

Finally, in 1991, 39 harbor seals were found stranded or dead off the New York coast – 62 percent more than a year earlier. Investigators found not only high concentrations of PCBs in the few seals tested, but also low counts of the white blood cells that fight off infection, MERI reports.

Though apparently unrelated, all these episodes involved populations that carried high burdens of toxic organochlorines – such as PCBs and dioxins – in their bodies or that lived in waters polluted with high quantities of toxic industrial chemicals. Because studies have demonstrated that PCBs and certain other organochlorines can suppress the infection-fighting immune system of laboratory animals, Shaw says that a circumstantial case began to build linking pollution to the viral epidemics devastating animals at the top of the marine food chain.

Indeed, such concerns prompted Osterhaus and his colleagues at the National Institute of Public Health and Environmental Protection in Bilthoven, the Netherlands, to launch a unique study involving harbor seals (*Phoca vitulina*). Now, three years later, they're able to report data strongly supporting their initial suspicions – that pollution may affect vulnerability to disease.

The Dutch team captured pups living along the northeast coast of Scotland. For a year, the researchers fed the animals a normal diet consisting of herring. Over the subsequent 2-year feeding trial, the 14 females and 8 males continued to eat herring. Half received fish from the relatively clean North Atlantic; the rest dined on fare harvested from the industrially polluted Baltic.

Chemical analyses showed that the Baltic fish carried 10 times as much toxic organochlorine contamination as did the North Atlantic herring. Osterhaus points out, however, that all the fish came from catches that had been destined for human consumption.

Every 6 to 9 weeks, the researchers took blood samples and made various measurements of the animals' immune functioning. Though both groups exhibited identical baseline values for the different factors, those fed Baltic herring quickly distinguished themselves once the study began.

For instance, the concentrations of

vitamin A in their blood dropped by 20 to 40 percent almost immediately and remained low throughout the 2 years. While the importance of this change is open to interpretation, Shaw notes that in other animal models, "vitamin A has been linked to disease resistance, with lower [blood] concentrations corresponding to increased vulnerability."

As vitamin A was falling, the concentrations of granulocytes in the blood of the Baltic-fed seals climbed and remained consistently elevated – at about 10 to 15 percent above those in the other group. Because this subpopulation of white blood cells is important in combating pathogenic bacteria, Osterhaus now speculates that seals eating the more polluted herring may have suffered from higher levels of chronic infections.

Even more impressive was an almost immediate plummeting in the activity of another population of white blood cells, known as natural killers (NK). These can attack foreign bodies without first having to recognize specific antigens – molecular "flags" that help trigger an immune response. NK activity in seals fed Baltic herring remained 20 to 50 percent below normal throughout the study, reflecting an apparent drop in the responsiveness of this important facet of the animals' immune system.

Another crucial component of the immune system also appeared somewhat depressed in seals eating the more polluted fish. B cells, which produce antibodies, and T cells, which orchestrate or directly participate in an animal's immune defense, will ordinarily proliferate when challenged with various types of antigens.

During their study, the scientists report in the March Ambio, the proliferative response of the T cells to a set of standard antigens dropped 25 to 60 percent in the seals fed Baltic herring, compared to those fed fish from the Atlantic. Additional, unpublished data suggest that the antibody responses of B cells also were impaired.

"I was surprised to see significant immune changes in animals that were fed on a normal diet using fish fit for human consumption," Osterhaus says. Extrapolating these findings back to the 1988 Baltic and North Sea distemper die-off, he adds, "I think we can now say that it's not unlikely that the seals' natural immune response to infection may have been impaired."

Molecular immunologist David A. Ferrick of the University of California School of Veterinary Medicine in Davis describes the feeding trial as a landmark study. "This is the first time anybody's done a good, controlled study looking at the direct effect of contaminant exposures on immune function or ability to resist disease," says Ferrick, codirector of the International Program for Marine Mammal Health at the Marine Mammal Center in Sausalito, Calif.

Peter Ralston

Maine harbor seal basks on ledge of rock in outer Penobscot Bay. Such blubbery marine mammals accumulate fat-seeking pollutants, some of which may affect an animal's vulnerability to disease.

Indeed, he notes, owing to the heavy regulatory protection afforded marine mammals by many nations, such a feeding trial – involving food known to be heavily contaminated – probably could not win approval in most countries, the United States included.

However, while describing the study's findings as interesting and important, Ferrick cautions against overinterpreting them. "The immunological differences that [the Dutch team] measured are not what you would call astounding" or even substantial enough to guarantee that they would compromise an animal's ability to fight infection.

But Ferrick also points out that the study animals were well maintained in captivity, with adequate stores of food. The immune responses observed in the seals fed Baltic herring may simply reflect the steady, chronic – and *subtoxic* – ingestion of potentially dangerous pollutants.

Many of the toxic chemicals detected in these fish accumulate in fat. "And their large blubber stores mean that marine mammals can accumulate far greater quantities of these contaminants than can most species," Ferrick notes.

In the wild, marine mammals periodically draw down that fat. It provides much of the energy that a mother delivers to her suckling young; it can also ensure an individual's survival during the periods of famine that inevitably seem to follow periods of abundance.

Remobilizing fat at such times may dump dangerous quantities of stored contaminants into the blood. What the Dutch team observed may therefore understate the potentially acute threat that chronic ingestion of pollutants might pose to animals under stress, Ferrick says.

Moreover, he points out, Osterhaus' group assayed immune function in these pinnipeds using tests developed for humans and laboratory animals. While they are the existing gold standard, those assays may be relatively insensitive for gauging the health of marine mammals, Ferrick believes.

It's for this reason that Ferrick's team has been working to develop diagnostics for individual components of marine animals' immune systems. Eight to 10 such tests – not only for harbor seals but also for dolphins – are undergoing final refinements. Other assays, for killer whales and sea otters, are in the works.

"If another *Exxon Valdez* accident happened tomorrow, we could already do a much better job [of characterizing its effects on the immune health of marine mammals]" than was possible in 1989, Ferrick maintains. "But what we still wouldn't be able to do is run diagnostics on the blood and do triage – say this animal should be sent for rehabilitation while that one needs to be euthanized." Acquiring this capability may take another 3 to 7 years, he estimates.

For now, Osterhaus and his colleagues continue to closely monitor the immune function of their captive seals. Those once fed Baltic herring now get the same diet of North Atlantic fish as the others. The goal: to see when – if ever – their immune parameters return to normal.

"While this is a very important study in establishing the link between environmental pollution in seals and illness, it's only one study," Shaw says. She emphasizes that there will have to be additional research to prove a causal association. And she'd like to see such studies begin soon, because there may be more than seals at risk.

Because of their large fat deposits and position at the top of the marine food chain, Shaw points out, "all marine mammals are bioaccumulating large quantities of environmental pollutants." Indeed, it's because of these properties, Osterhaus says, that "if any kind of adverse effects were to be expected, then [marine mammals] are the first you'd expect to see them in."

Shaw and Osterhaus note that because humans share that position at the top of the food chain, they also may be at risk. Such concerns haven't stopped Osterhaus from eating fish. However, he says, because the Wadden Sea – the source of fish in his area – is at least as polluted as the Baltic, "we make sure that [its fish] is not a substantial part of our diet."

But limiting fish intake should not be seen as the important take-home message, Osterhaus adds. "If a complete ecosystem is being polluted such that we see effects in its top predators, we have to realize that something is fundamentally wrong here and that we must do something to combat that pollution as it now exists." □