NATURAL TOXINS 2:236–251 (1994)

Accumulation of Paralytic Shellfish Toxins by Surfclams, *Spisula solidissima* (Dillwyn, 1897) in the Gulf of Maine: Seasonal Changes, Distribution Between Tissues, and Notes on Feeding Habits

Sandra E. Shumway, Sally A. Sherman, Allan D. Cembella, and Rhonda Selvin
Bigelow Laboratory for Ocean Sciences (S.E.S.), Department of Marine Resources (S.E.S., S.A.S.), Phytoplankton Analyses (R.S.), West Boothbay Harbor, Maine; National Research Council, Institute for Marine Bioscience, Halifax, Nova Scotia, Canada (A.D.C.)

ABSTRACT Accumulation of paralytic shellfish poisoning (PSP) toxins by surfclams, *Spisula solidissima,* was studied over a period of two years at two inshore locations in southern Maine and at six stations on Georges Bank in the Gulf of Maine. Whole animals as well as individual tissues (siphon, mantle, digestive gland, foot, adductor muscle, gill) were analyzed for PSP-toxicity levels using the standard AOAC mouse bioassay. Analyses of gut contents were carried out on surfclams from both inshore and offshore locations to identify the type of particles ingested. Surfclams feed primarily on phytoplankton and detrital material characteristic of the overlying seawater and surface sediment. No evidence was found for any selection based on particle size or type. Elevated levels of PSP toxins were noted in surfclams from Georges Bank more than two years after initial toxification. Toxins were not evenly distributed among the various tissues of surfclams. Initially, maximum toxicity among surfclam tissues was found in digestive glands; however, subsequent analyses of samples collected later in the year indicated that toxicity in gill and mantle tissues had increased relative to initial values. No toxicity was detected in adductor muscles. Surfclams are characterized by a high variation in total toxin load among individual animals, with a tendency for decreasing variation as toxin levels increase. Archived data from the Main Department of Marine Resources revealed annual and seasonal patterns of toxin accumulation by surfclams, i.e., toxin accumulation is an annual event, with initial increases in toxicity usually occurring in early spring.

Key Words: Paralytic shellfish toxins, Bioaccumulation, Biotransformation, Shellfish, Gut contents, Public health, Fisheries management

INTRODUCTION

The surfclam (= bar clam, hen clam), *Spisula solidissima,* occurs from Cape Hatteras, North Carolina to the Gulf of St. Lawrence, Canada [Merrill and Ropes, 1969]. Greatest concentrations generally occur nearshore in the turbulent waters of oceanic beaches, just beyond the breaker zone, at depths <18 m [Ropes 1979, 1980]. Surfclams account for approximately 25% of the total molluscan catch in the United States and rank second in total landings behind the sea scallop, *Placopecten magellanicus* [Stamatopoulos, 1993]. In the Gulf of Maine, populations are limited to the north shore of Massachusetts, the southeastern portion of Maine, and Georges Bank.

Surfclams are of great commercial importance along the mid-Atlantic seaboard and on Georges Bank, with annual commercial landings in the United States of approximately 20,000 metric tons of meat [Anonymous, 1992]. Commercial landings in 1991 were 66.2 million pounds ($\approx 3.1 \times 10^7$ kg of shellfish meat) valued at $US 29.2 million [O'Bannon, 1992]. While most surfclams are fished from New Jersey, New York, and Maryland, commercially important beds are also exploited on Georges Bank and, where they are harvested from coastal zones, e.g., the bottom dredge fishery in southern Maine, they are of significant commercial importance to the local economy. Furthermore, recent studies have indicated that young, cultured surfclams are a promis-

Received October 15, 1993; accepted for publication December 15, 1993.

Address reprint requests to Dr. Sandra E. Shumway, Natural Science Division, Southampton College, LIU, Southampton, NY 11968.

All work was performed at the Bigelow Laboratory for Ocean Sciences and the Maine Department of Marine Resources.

ing product [Goldberg, 1980; Krzynowek et al., 1980; Krzynowek and Wiggin,1982] and at least one aquaculturist in Maine is currently producing and marketing juvenile surfclams [W. Mook, personal communication].

Previous reports on the noxious effects of algal blooms on *Spisula* have generally been limited to mass mortalities associated with blooms of the dinoflagellates *Ceratium tripos* [Mahoney and Steimle, 1979; Steimle and Sindermann, 1978] and *Gymnodinium breve (Ptychodiscus brevis)* [Tiffany and Heyl, 1979]. Surfclams have, however, been implicated in human illnesses associated with toxic aglae. Two cases of neurotoxic shellfish poisoning (NSP), a syndrome typically associated with *Gymnodinium breve* blooms, resulted from consumption of *Spisula solidissima raveneli* from the Gulf of Mexico [Hemmert, 1975]. Only a single account of diarrhetic shellfish poisoning (DSP) has been related directly to contamination in surfclams *(Mactra chinensis)* [Anraku, 1984].

Surfclams, similar to other filter-feeding bivalve molluscs, readily accumulate paralytic shellfish poisoning (PSP) toxins when they are exposed to toxigenic phytoplankton. In the Gulf of Maine, the toxic dinoflagellate, *Alexandrium* spp., is considered to be the primary (and perhaps exclusive) causative organism responsible for PSP [Yentsch and Hurst, 1979].

Although not usually considered a serious PSP hazard, surfclams have previously been linked to toxic incidences in humans. Medcof et al. [1947] included data on toxic *Mactra* (=*Spisula*) *solidissima,* primarily from Canadian waters, and Bond and Medcof [1957] reported one case of PSP in humans involving this species. Prakash et al. [1971] noted that although surfclams are sometimes very toxic in the Bay of Fundy, they accounted for <1% of reported shellfish poisonings and no deaths.

Prior to the summer of 1988, PSP toxicity in surfclams was considered to be strictly a near-coastal problem in the Gulf of Maine. Such toxicity has been a recurrent problem for decades, and regular testing of surfclams for PSP toxicity in coastal Maine waters began in 1975 [see Shumway et al., 1988, for detailed summary]. Paralytic shellfish toxins were detected for the first time in surfclams from offshore waters (Georges Bank) in 1988 [see White et al., 1993a,b, for a complete description of these events]. Briefly, during the summer of 1988, elevated toxicity levels (2,000 μg saxitoxin equivalents [STX equiv] 100 g tissue^{-1}) were detected in digestive glands of sea scallops *(Placopecten magellanicus)* from the Canadian sector of Georges Bank [W. Watson-Wright, pers. comm.]. Gillis et al. [1991] have reported levels as high as 1,440 μg STX eqiv 100 g tissue^{-1} in the viscera of *P. magellanicus*. Unprecedented high toxicity (up to 100 μg STX equiv 100 g tissue^{-1}) was also recorded in blue mussels *(Mytilus edulis)* from Nantucket Shoals [J. Hurst, pers. comm.]. In mid-August 1989, PSP toxicity (300–500 μg STX equiv 100 g tissue^{-1}) was detected in whole surfclams from southern Georges Bank, resulting in a 90-day emergency closure of the American sector to the harvesting of surfclams, ocean quahogs, and whole scallops [Sharifzadeh et al., 1991; White et al., 1993a]. The surfclam fishery was reopened briefly in the spring of 1990 but was closed again in May when toxic surfclams (600–1,530 μg STX equiv 100 g tissue^{-1}) were found. White et al. [1993a] suggested that, since there are no reports of PSP from consumption of offshore shellfish prior to 1990, PSP toxins were not present at high levels prior to these recent events. The area was closed indefinitely as per Amendment 8 of the Surfclam/Ocean Quahog Fisheries Management Plan and remains closed as of this writing (January 1994), imposing a significant economic loss upon both the fishing industry and the supporting processing industry.

Data are presented here for seasonal changes in PSP toxicity of individual tissues and whole surfclams from inshore and offshore localities in the Gulf of Maine. Where available, data on the blue mussel *(Mytilus edulis),* a species which detoxifies quickly, are also presented. This interspecific comparison allows for a determination of whether the toxins present in *Spisula* were recently acquired or whether they are remnants of previous toxic events. Since prior studies of the feeding habits of *Spisula* are lacking, data were collected to establish: 1) the overall composition of the surfclam diet, and 2) the presence of known toxic algal species.

MATERIALS AND METHODS

Inshore

Surfclams, *Spisula solidissima* (shell length 80–110 mm) were collected monthly from two inshore stations at Head Beach and Goose Rocks Beach in southern Maine (see Fig. 1) from March 1990 through August 1991. Clams were dug by hand and transported whole within 2 hours to the laboratory at Boothbay Harbor, where individual tissues were dissected for the determination of PSP toxicity. Tissues were separated as digestive gland, mantle, siphon, gill, adductor muscle, and foot (see Fig. 2) and rinsed briefly in clean seawater. Individual tissues from six clams were pooled for each analysis. Tissues from whole clams (n = 6) were also pooled and tested immediately for PSP toxicity using the standard three-mouse bioassay [AOAC, 1984; see Halstead and Schantz, 1984, for complete description]. Data are presented as μg STX equiv 100 g tissue^{-1}. Samples for gut content analysis were taken in the field, immediately upon capture. After careful removal of the dorsal shell valve, the animal was washed with filtered (0.45 μm) seawater to be sure no debris remained to be confused with possible food items/gut contents. Entire digestive glands (diverticula) including stomachs were removed and placed in Lugol's iodine preservative. Tissues were macerated but not homogenized. Simultaneous samples of the "fluff layer" and of the overlying seawater were taken for comparison with gut contents. These were also preserved in Lugol's solution. Samples were analyzed later using a Sedgewick-Rafter chamber

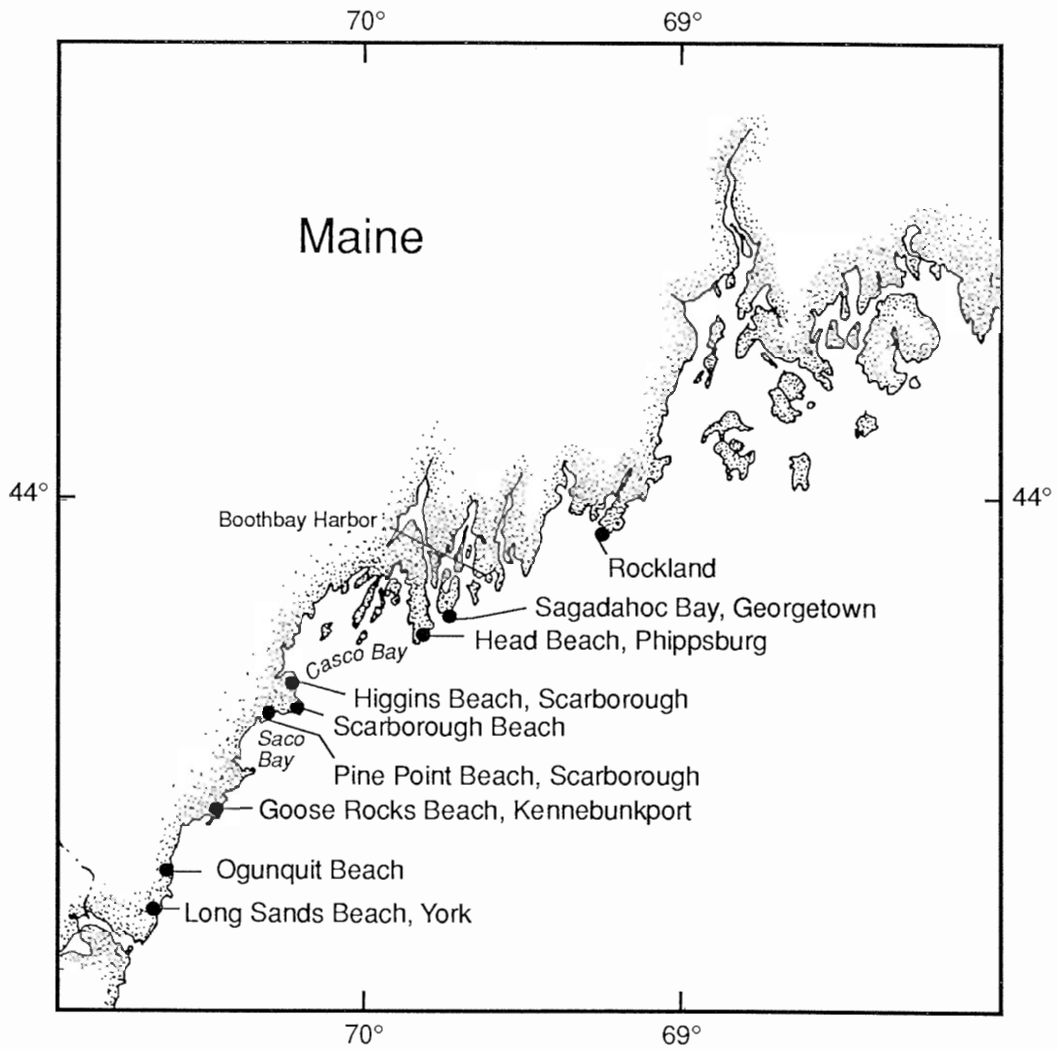

Fig. 1. Map showing location of inshore sampling locations, Head Beach and Goose Rocks Beach. Other locations noted are archived data from Maine Department of Marine Resources.

(1 ml) after a 1:1 dilution with filtered seawater. Initially, some samples were also preserved in formalin to compare preservation methods. Lugol's solution was chosen as the superior preservative for subsequent samples.

Samples of preserved gut contents (0.1–0.2 ml) were drawn by pipet for analysis. In initial sampling trials, increasing the sample size to 1 ml did not yield additional species representation. Initial trials using Davidson's fixative or buffered formalin were not as successful as Lugol's iodine solution. Gut contents (n = 41) were identified using standard bright-field and phase-contrast microscopic techniques as described previously [Shumway et al., 1987; Newell et al., 1989].

Offshore

After the initial discovery of toxic shellfish on Georges Banks in 1989, our attention focused on the determination of

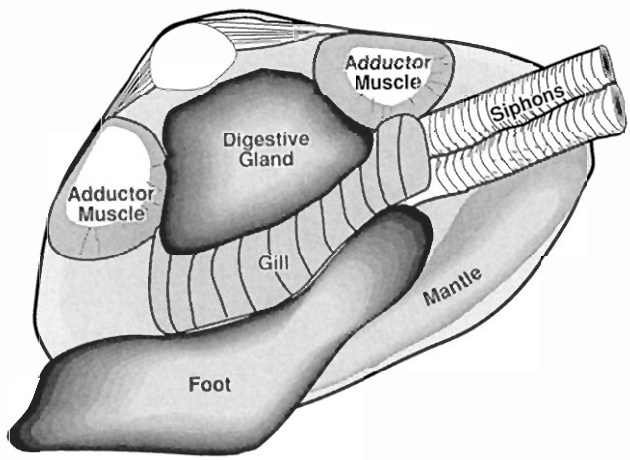

Fig. 2. Diagrammatic representation of surfclam tissues sampled for toxin analysis.

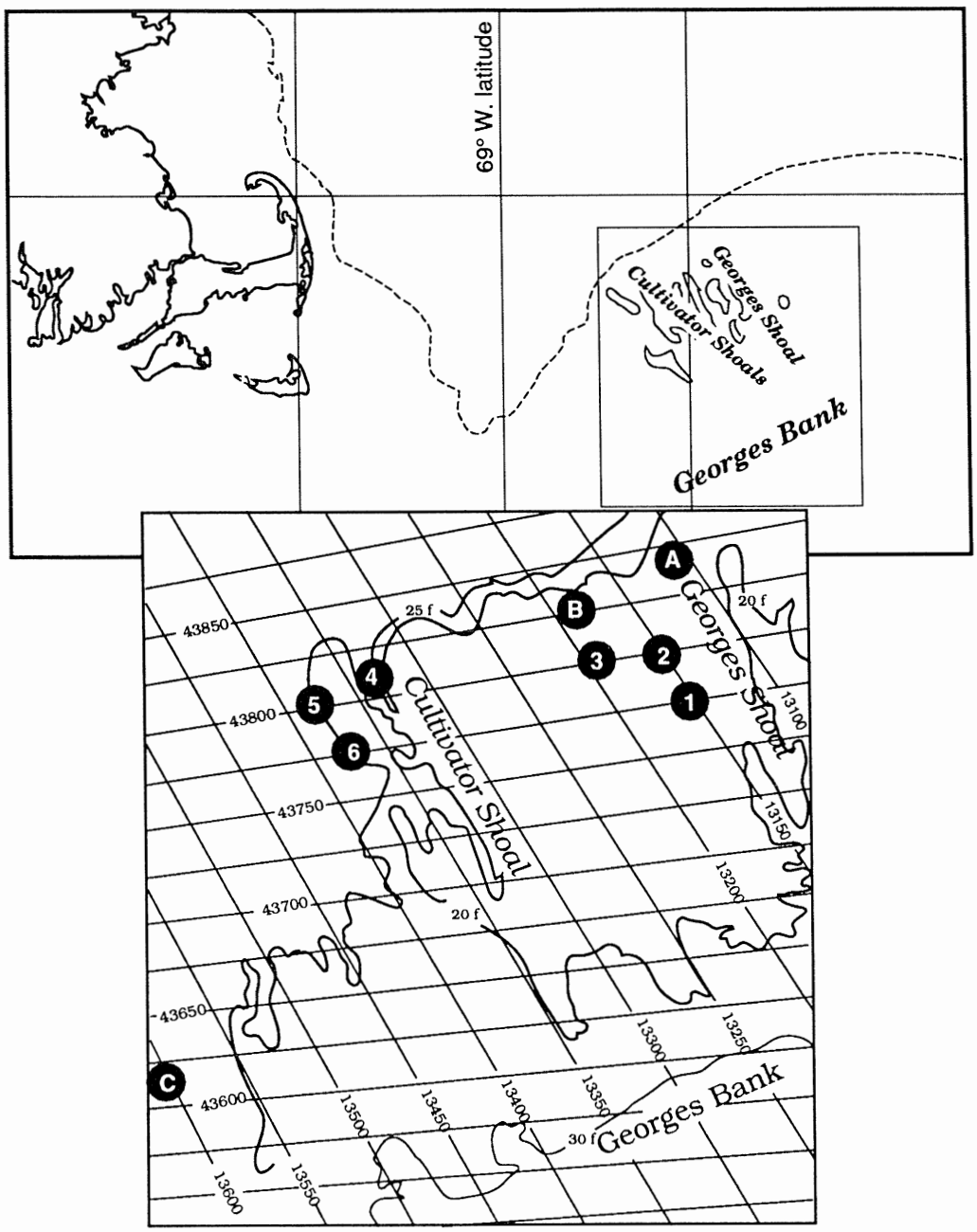

Fig. 3. Location of sampling stations on Georges Bank off the northeastern coast of the United States. Grid lines in bottom part of figure are Loran C lines [after White et al., 1993b].

the level and distribution of PSP toxicity among tissues of commercially important populations of surfclams in this region. Adult surfclams (shell length 110–170 mm) were collected with a hydraulic clam dredge by a commercial fishing vessel. Clams were collected during 13 cruises between June 28, 1990, and May 5, 1992, from six stations on Georges Banks (Fig. 3). Bottom tows were of approximately 5–10 minutes duration at approximately 2–2.5 knots. As with clams from inshore localities, tissues (see Fig. 2) were dissected from six individual surfclams and pooled for analysis of total toxicity by the AOAC [1984] mouse bioassay. A second set of tissues was frozen and stored at −80°C for subsequent analyses of PSP-toxin composition by high performance liiquid chromatography with fluorescence detection (HPLC-FD) [Cembella et al., 1993a,b; Cembella and Shumway, 1993].

In an initial attempt to monitor loss of PSP toxins, several hundred surfclams were collected during the cruise of September 13, 1990, and returned to the Boothbay Harbor facility where they were immediately placed in running seawater

TABLE I. Changes in PSP Toxicity (μg STX Equiv 100 g Tissue^{-1}) of *Spisula solidissima* After Extended Periods of Depuration in Laboratory Tanks*

		Date		
Station	Tissue	11/19/90 (t = 67 d)	1/15/91 (t = 124 d)	3/14/91 (t = 182 d)
13193-43799	Whole	1,705	2,579	678
(Station 3)	Siphon	2,232	1,971	2,120
	Gill	8,240	8,600	3,750
	Dig. gland	4,759	3,398	4,240
	Mantle	3,393	2,015	2,261
	Foot	349	404	344
	Add. muscle	<40	81	42
13358-43804	Whole	1,078	1,479	—
(Station 5)	Siphon	1,113	1,260	—
	Gill	4,557	4,294	—
	Dig. gland	4,252	6,684	—
	Mantle	2,359	2,194	—
	Foot	227	185	—
	Add. muscle	54	58	—

*Each score represents a pooled sample of six clams.

at 15°C. Elastic bands were placed around the clams to enhance survival. This does allow for filtration but compensates for lack of pressure normally provided by sediments. Clams were maintained in the laboratory in running seawater at ambient temperature for 6 months. Incoming seawater was prefiltered through a sand filter to remove all particles >20 μm, which would include PSP toxin producing dinoflagellates. Clams were subsampled (see Table I) for the determination of total PSP toxicity.

Due to extremely hazardous weather conditions on Georges Bank, samples for gut analyses could not be collected on a regular basis. Samples of phytoplankton were collected using a Niskin bottle from two offshore stations (#3 [Loran 13193-43799] and #8 [Loran 13876-43838]; Tables IV and V) to confirm the presence of dinoflagellates (i.e., *Alexandrium* spp.) likely to be responsible for measured PSP toxicity of surfclams from these sites. Offshore sampling was usually carried out under adverse conditions and often with a short-handed crew. Clams were sacrificed immediately upon landing and the guts were removed and preserved as for inshore samples.

The presence of large quantities of toxic shellfish, coupled with the large size of individual surfclams, provided a unique opportunity to examine variability in PSP toxin accumulation among individual animals. A total of 42 sets of 10 randomly selected individual surfclams were assayed for PSP toxicity using the mouse bioassay. These toxicity measurements were undertaken to establish a database for development of statistically sound testing protocols for fisheries management and public health protection.

Archived Data

In addition to experimental data, results from archival records of the Maine Department of Marine Resources were compiled for two locations on the Maine coast, Scarborough Beach and Head Beach (see Figs. 1, 8, 9). Comparative data on mussels, *Mytilus edulis,* from the same or nearby localities (in the case of Scarborough, the nearest primary sampling station is the Spurwink River, approximately one mile away) are also presented.

RESULTS

Inshore

Figures 4 and 5 summarize toxicity data for *Spisula* collected from Head Beach and Goose Rocks Beach from March 1990 through September 1990. Measurable PSP toxicity was present during the entire 18 months of monitoring in all tissues except for foot and adductor muscle. While maximum PSP-toxicity levels were higher at Goose Rocks Beach than at Head Beach, the seasonal pattern was similar, with maximum toxicity occurring during the summer (June to August). Furthermore, the distribution of total toxicity among individual tissues is similar at these stations. Total toxicity levels in whole surfclams were low at both locations with maximum recorded values of 210 μg STX equiv·100 g tissue^{-1} at Head Beach and 147 μg STX equiv·100 g tissue^{-1} at Goose Rocks Beach. Although analyses of whole clams were not carried out until May 25, 1990, samples of digestive glands and other tissues contained low toxicity levels at this time; the toxicity present during the early spring was probably carried over from previous toxic events. Lack of measurable toxicity in mussels collected in close proximity to the surfclams is further evidence for such a carry over of toxicity. The sudden increase in toxicity of the digestive glands at both locations (June 26, 1990, at Head Beach and July 24, 1990, at Goose Rocks Beach) indicates that a toxic event occurred during the early summer. Subsequent sampling showed a gradual decrease in toxicity of digestive glands through December and a repeated rise in toxicity during the spring and summer of 1991. Adductor muscle and foot tissue never exhibited toxicities exceeding the regulatory limit of 80 μg STX equiv 100 g tissue^{-1} during the study period and whole animal toxicity only exceeded the regulatory limit during June of 1990 at Head Beach, and from June through August of the same year at Goose Rocks Beach. No statistically significant correlations (Pearson product-moment correlation, $P \leq 0.05$) could be established in pairwise comparisons of toxicity levels in any of the individual tissues or between whole animal toxicity and toxicity of any individual tissue(s).

Offshore

Levels of PSP toxicity (μg STX equiv 100g tissue^{-1}) in whole surfclams from the six stations on Georges Bank are shown in Figure 6. It was our initial hope that, as in some other species of bivalve molluscs including mussels and sea scallops, PSP toxins would be concentrated primarily in the digestive gland, leaving commonly processed tissues (foot, siphon, and mantle) harvestable. While total toxicity varied considerably among individual animals and among stations,

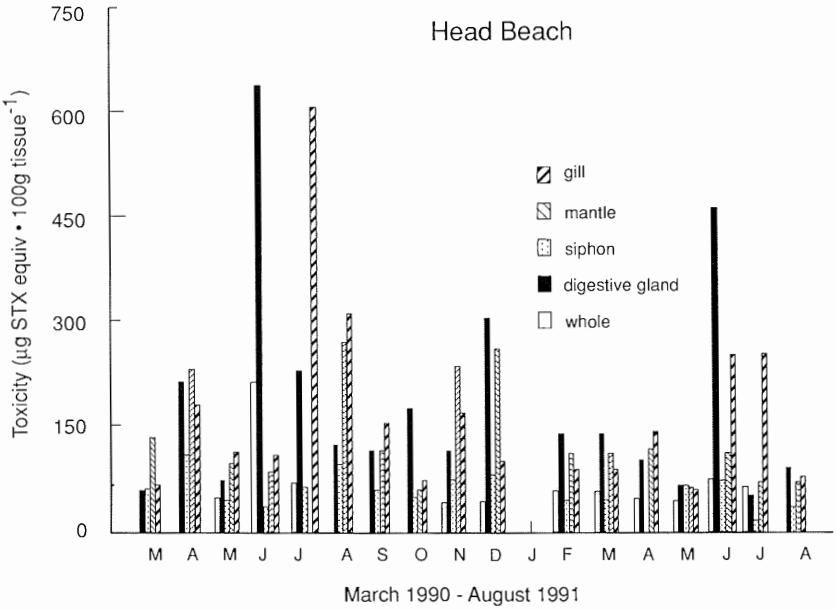

Fig. 4. Toxicity data for *Spisula solidissima* collected from Head Beach, Maine. Each histogram represents a pooled sample of 6 individual clams or tissues.

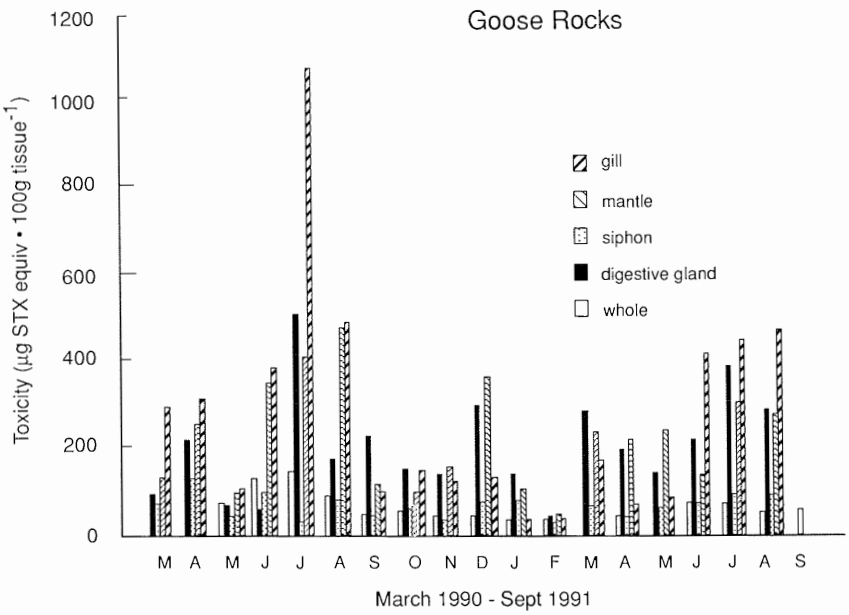

Fig. 5. Toxicity data for *Spisula solidissima* collected from Goose Rocks Beach, Maine. Each histogram represents a pooled sample of 6 individual clams or tissues.

the trend was the same at all locations. Toxicity in whole surfclams was initially high (>500 µg STX equiv 100 g tissue^{-1}) and increased rapidly to levels in excess of 6,000 µg STX equiv 100 g tissue^{-1}) in the late spring and early summer. There was a rapid decline in total toxicity by August 1990, followed by a prolonged period of gradually declining toxicity to persistent levels which remained above the mouse bioassay detection limit (40 µg STX equiv 100 g tissue^{-1}). As with clams from inshore locations, no statistically significant correlations could be made among toxin levels in any of the individual tissues or between whole animal toxicity and toxicity of any individual tissue.

Toxicity of individual tissues for all of the cruises with all stations combined, is given in Figure 7. Each point represents mean toxicity (n = 6) of pooled tissue samples (n = 6) from each cruise. High levels of toxicity were sustained throughout the winter months at both inshore and offshore stations. Toxicity was not evenly distributed among the tis-

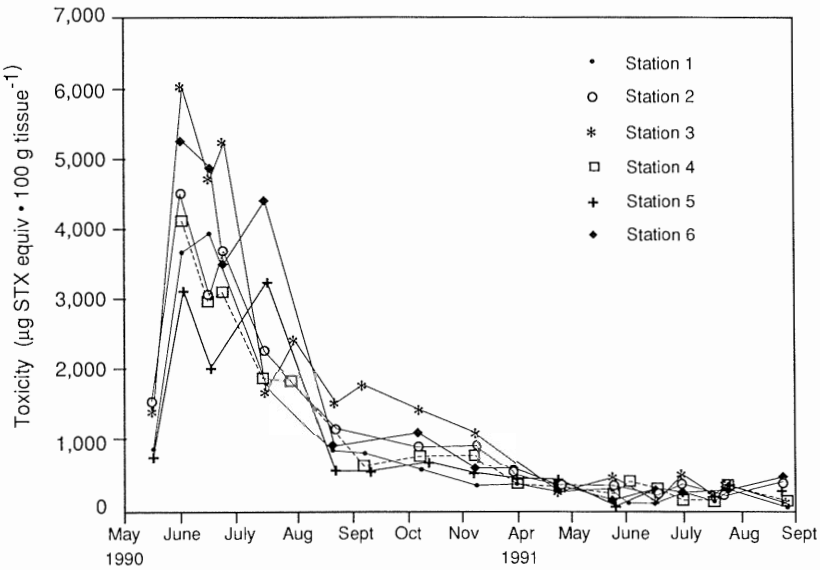

Fig. 6. Levels of PSP toxicity in whole surfclams *(Spisula solidissima)* at the six stations on Georges bank shown in Figure 3. Results are based on composite samples of 12 animals for each sample.

sues and substantial differences in relative amounts were noted. Initially, toxicity was primarily concentrated in the digestive glands; however, the gill, siphon, and mantle subsequently served as primary toxin storage organs. Total toxicity in digestive glands followed the pattern for whole animals; however, maximum levels in other tissues (gill, mantle, siphon, foot) were not attained until approximately three months after the presumed bloom event. A general toxicity pattern was noted: digestive gland > mantle = gill > siphon = foot > adductor muscle. Toxicity levels in adductor muscles ranged from 40–97 µg STX equiv 100 g tissue^{-1}. There were occasions, especially during the summer months (see Figs. 4, 5) when PSP toxicity of gill tissues exceeded that of any other tissue. Presumably, this net increase represents the equilibrium of toxin sequestration and biotransformation processes in surfclams. This general trend was also reflected in the corresponding analyses of individual toxins in various tissues by HPLC and will be presented in detail in forthcoming publications [Cembella et al., 1993a,b].

Toxin Elimination

Initial depuration studies in which highly toxic surfclams were collected from offshore localities (Stations 4 and 5; Table I; Fig. 3) and returned to the laboratory showed elevated toxicity levels even after 4 and 6 months maintenance in 20 µm-filtered seawater. Little, if any, toxicity was lost during the 4-month period (Table I). Our subsequent field studies corroborated these findings, since PSP toxicity in excess of quarantine levels was measured 2 years after the initial determinations.

Gut Content Analyses

Inshore

In the particle size range from 10 to 240 µm, a total of 26 distinct microalgal taxa, primarily diatoms and dinoflagellates, were identified to the specific or generic level in surfclam gut samples (Table IV). In addition, unidentified flagellates, detritus, and zooplankton were also found in gut samples. Most gut content samples consisted predominantly or exclusively of detrital material. Evidence of spawning (spent gonads) was seen in August 1990 and June 1991.

Both planktonic and benthic species were observed in the guts. Diatoms and dinoflagellates in concurrent water and sediment samples were generally also those represented in the gut contents (Tables II, III). The hypnozygotic cyst form in the PSP toxin producing dinoflagellate, *Alexandrium tamarense,* was observed at both sites in September 1991; vegetative cells were identified in the water samples during April, June, and August 1991. This delay in the appearance of hypnozygotes is typical of *Alexandrium* bloom dynamics. A lag phase was also observed between the first occurrence of *Alexandrium* vegetative cells in the gut contents and consequent toxicity peaks in shellfish tissues.

Several species of the planktonic dinoflagellate *Dinophysis* have been directly implicated in incidents of diarrhetic shellfish poisoning (DSP) in various areas of the world (although not yet in the Gulf of Maine). In any case, four dinophysoid species (*D. acuminata, D. caudata, D. norvegica,* and an unidentified morphotype) were found in surfclam guts, sediment samples, or in the water column during spring, summer, and fall.

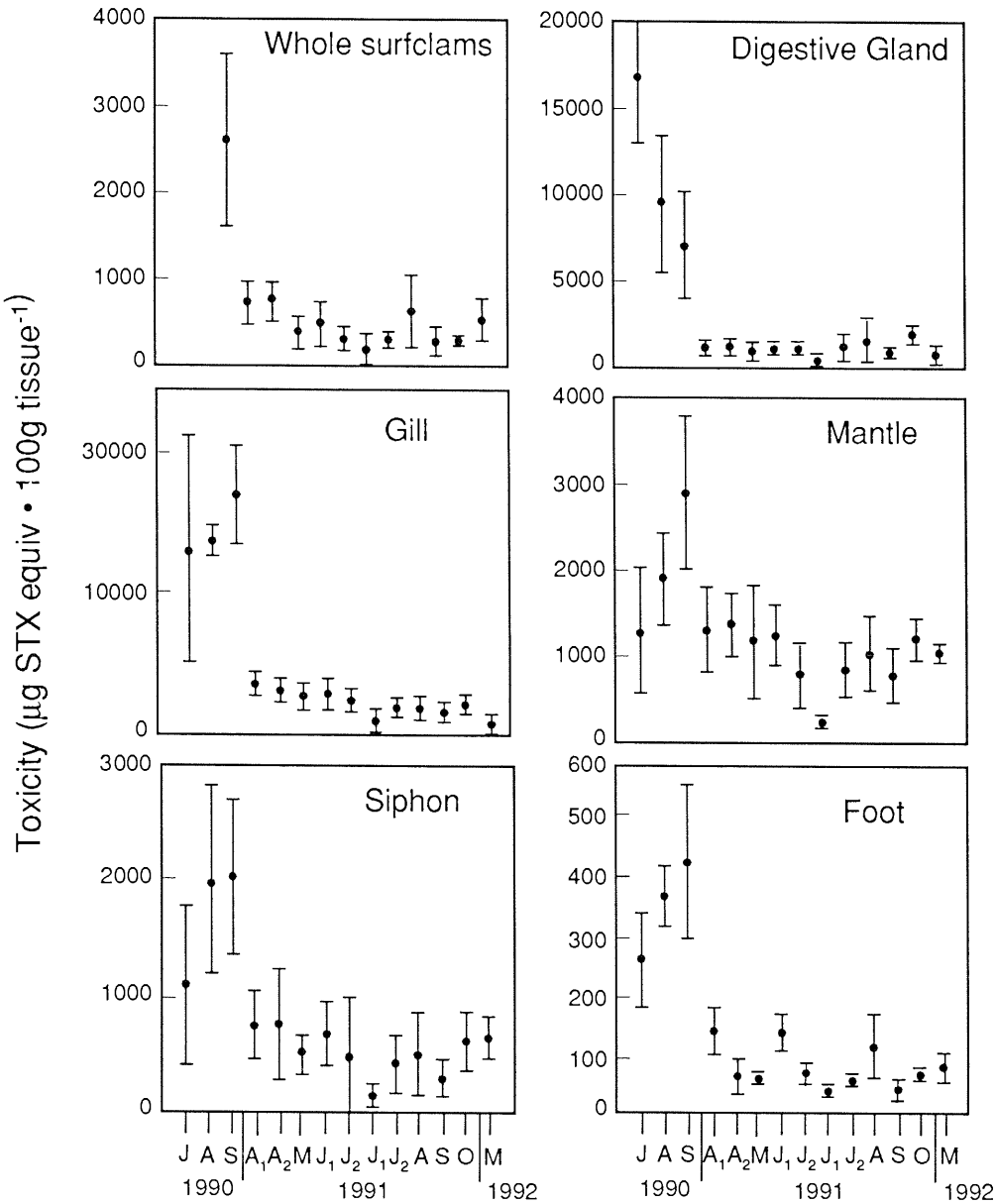

Fig. 7. Levels of PSP toxicity in whole surfclams *(Spisula solidissima)* and in individual tissues. Each point represents the mean toxicity of 6 individual clams from all six stations on any particular cruise date. Error bars represent one standard deviation.

Offshore

The gut samples collected from offshore sites contained primarily detrital material with few identifiable phytoplankton (Tables IV, V). The offshore plankton sampling was not comprehensive and was only carried out as time and weather permitted; however, it is significant that one of the samples yielded cells of *Alexandrium tamarense,* the organism suspected to be responsible for the observed PSP toxicity. D.M. Anderson [pers. comm.] has also identified *A. tamarense* in low numbers from these waters. These observations represent the first documentation of this species from offshore waters of the Gulf of Maine. It is perhaps significant that no vegetative cells or cysts of *A. tamarense* were identified in the gut samples of offshore surfclams (Table VI).

Archived Data: Maine Departmet of Marine Resources

Surfclams have been sampled sporadically by the Maine Department of Marine Resources for determination of PSP toxicity for the past 20 years. Sampling stations were established at eight locations in southern Maine (see Fig. 1). While some surfclams exhibited PSP toxicity (maximum recorded from inshore areas: 7,934 µg STX equiv 100g

TABLE IV. Phytoplankton Species Present in Water Samples From Station 8 (Loran 13193-43799)

	1991			
Species	5/23	7/2	7/16	9/18
Bacillariophyceae				
Amphiphora sp.		x		
Chaetoceros spp.				x
Ditylum brightwellii			x	
Melosira sulcata		x	x	
Navicula spp.		x	x	
Nitzschia spp.			x	
Nitschia cf seriata				x
Rhizosolenia sp.		x	x	
Skeletonema costatum		x		
Thalassionema nitzschiodies		x		
Thalassiosira sp.		x	x	
Unidentified centric species	x			x
Unidentified pennate species	x			x
Dinophyceae				
Alexandrium tamarense				x
Ceratium spp.	x	x	x	x
Dinophysis acuminata	x	x	x	x
Dinophysis acuta	x	x	x	
Dinophysis caudata				x
Dinophysis norvegia	x	x	x	
Dinophysis rotundata		x	x	
Sinophysis sp.	x		x	
Peridinum spp.	x	x	x	
Prorocentru micans		x	x	x
Scrippsiella spp.		x		x
Unidentified dinoflagellate	x		x	

TABLE V. Phytoplankton Species Present in Water Samples From Station 3 (Loran 13879-43838)

	1991				
Species	5/23	6/4	7/16	8/28	9/18
Bacillariophyceae					
Chaetoceros spp.			x		
Corethron hystrix			x		
Coscinodiscus spp.				x	x
Eucampia zoodiacus			x		
Guinardia flaccida	x	x	x		
Melosira sulcata			x		
Navicula spp.		x	x		
Nitzschia spp.			x		
Rhizosolenia sp.	x	x	x		
Thalassionema nitzschiodies			x		
Thalassiosira sp.	x	x	x	x	x
Unidentified centric species	x				x
Unidentified pennate spp.			x		x
Dinophyceae					
Ceratium fusus	x	x	x		
Ceratium lineatum	x	x	x		
Ceratium longipes		x	x		
Ceratium tripos	x	x	x		
Dinophysis acuminata			x		x
Dinophysis acuta	x		x		
Dinophysis caudata				x	
Dinophysis norvegica	x	x	x	x	x
Dinophysis rotundata			x		
Dinophysis sp.			x		
Peridinium spp.			x		x
Prorocentrium micans	x		x		
Scrippsiella trochoidea		x			
Unidentified dinoflagellate			x		x

levels than neighboring surfclams. Not only do these data demonstrate the seasonal accumulation of PSP toxicity by both species, they also clearly show prolonged toxin retention by surfclams. Toxins accumulated by mussels during the same time periods were quickly eliminated, typically within a few weeks.

Toxin Variability Among Individual Surfclams

There was a large variation in PSP-toxicity levels among individual clams. Results of these studies are summarized in Figure 10 [see also White et al., 1993b]. The coefficient of variation (CV) of each data set ranged from 19 to 99%. Examples of the degree of variation are depicted graphically in Figure 10, showing actual toxicity of individuals clams in selected sets (complete data set is available); the mean CV for the 42 surfclam data sets was 48.6%. There was a tendency for the variation among individual surfclams to decrease as toxicity increased (Fig. 10). Analysis of PSP-toxicity level versus shell length of surfclams showed no significant correlation over the size range tested [White et al., 1993b].

DISCUSSION

Surfclams are highly efficient filter-feeders and are not apparently subject to major adverse effects of exposure to PSP toxins even at high levels for extended periods. Previous studies of surfclams indicated no effect of PSP toxins on heart rate [Gainey and Shumway 1988a], rates of filtration [Shumway et al., 1985], clearance and irrigation [Shumway and Cucci, 1987], or shell-valve activity [Shumway and Cucci, 1987]; however, a decreased rate of oxygen consumption and an increased rate of pseudofeces production [Gainey and Shumway, 1988b] have been shown in this species upon exposure to a highly toxic isolate of *Alexan-*

TABLE VI. Algal Species Identified in Guts of *Spisula solidissima* From Stations 3 and 5A (Loran 13876-43838; 13358-43804) on September 13, 1990

Species	Size (μm)	Habitat[a]
Bacillariophyceae		
Cocconeis scutellum	20–30	B
Melosira sulcata	35–38	B
Thalassiosira sp.	20–35	P
Unidentified pennate diatom		
Other		
Prorocentrum micans	55	P

[a] B = benthic; P = pelagic.

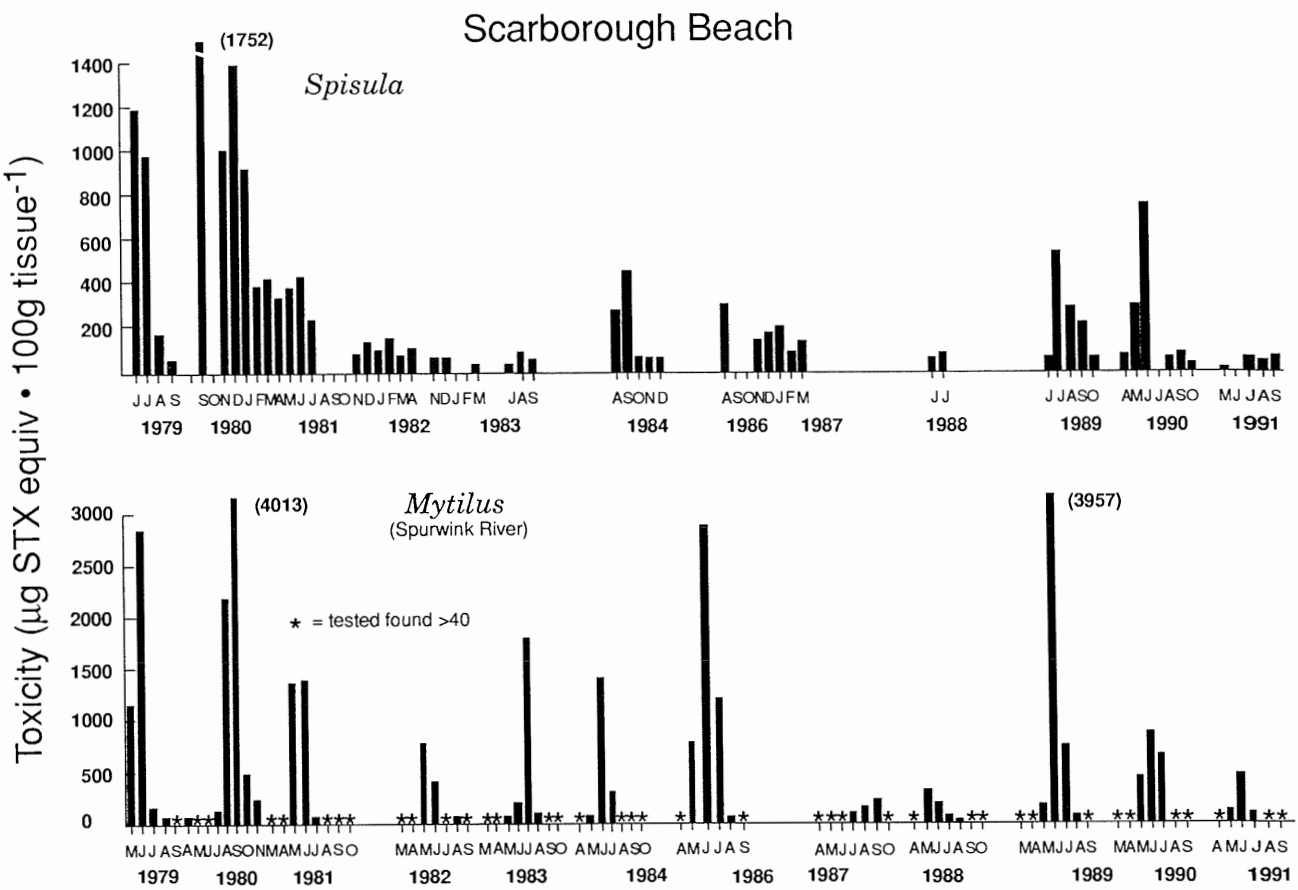

Fig. 8. Toxicity data for *Spisula solidissima* collected from Scarborough Beach between June of 1979 and September 1991. Each histogram represents a composite sample of 6 individual clams. Data for *Mytilus* represent a composite sample of 15–20 individual mussels at each sampling date.

drium tamarense in culture. Studies by Twarog et al. [1972] demonstrated that *Spisula* is highly sensitive to tetrodotoxin (TTX), the neurotoxin produced by puffer fish; no data are available for the sensitivity of surfclams to PSP toxins, purine neurotoxins with similar sodium-channel blocking activity in nerve fibers.

Gut Contents

Few data are available on the feeding habits of *Spisula solidissima*. Leidy [1878] reported an abundance of diatoms in the digestive tract of surfclams from a New Jersey Beach, but specifically mentioned only the pennate species *Amphipora constricta* and the ciliated protozoan, *Tintinnus*. In the only other report, Belding [1910] also listed diatoms as the primary food of surfclams.

We found that the diet of surfclams is composed of a variety of phytoplankton species typically present in the overlying seawater and on the sediment surface. There is no evidence for selection of any particular size fraction of particles or species of phytoplankton. The presence of large amounts of detrital material may be an artifact of the sampling strategy or may be due to rapid digestion of some algal species; however, it is not unlikely that the diet of a filter-feeding species such as *Spisula*, which lives in highly energetic environments where surface particulates are likely to be resuspended, would be dominated by detrital particles.

The PSP toxicity found in surfclams and other shellfish on Georges Banks is assumed to result from ingestion of the toxic dinoflagellate, *Alexandrium tamarense*, yet the evidence remains circumstantial. Motile cells of this species were identified from water samples at Station 3 in September 1991 (Table IV), although no cells or cysts were found in the guts. Further evidence for *A. tamarense* as the causative organism of PSP toxicity is the similarity between PSP toxin profiles, as determined by HPLC analysis, of surfclam tissues during the initial rise in summer PSP toxin levels at inshore and offshore sites and those of *Alexandrium* isolates obtained from adjacent areas in the Gulf of Maine [Cembella et al., 1993a].

Seasonal Variation

Seasonal variation in toxicity is more prominent than geographical differences among inshore and offshore surfclam populations. Increased toxicity levels are usually observed

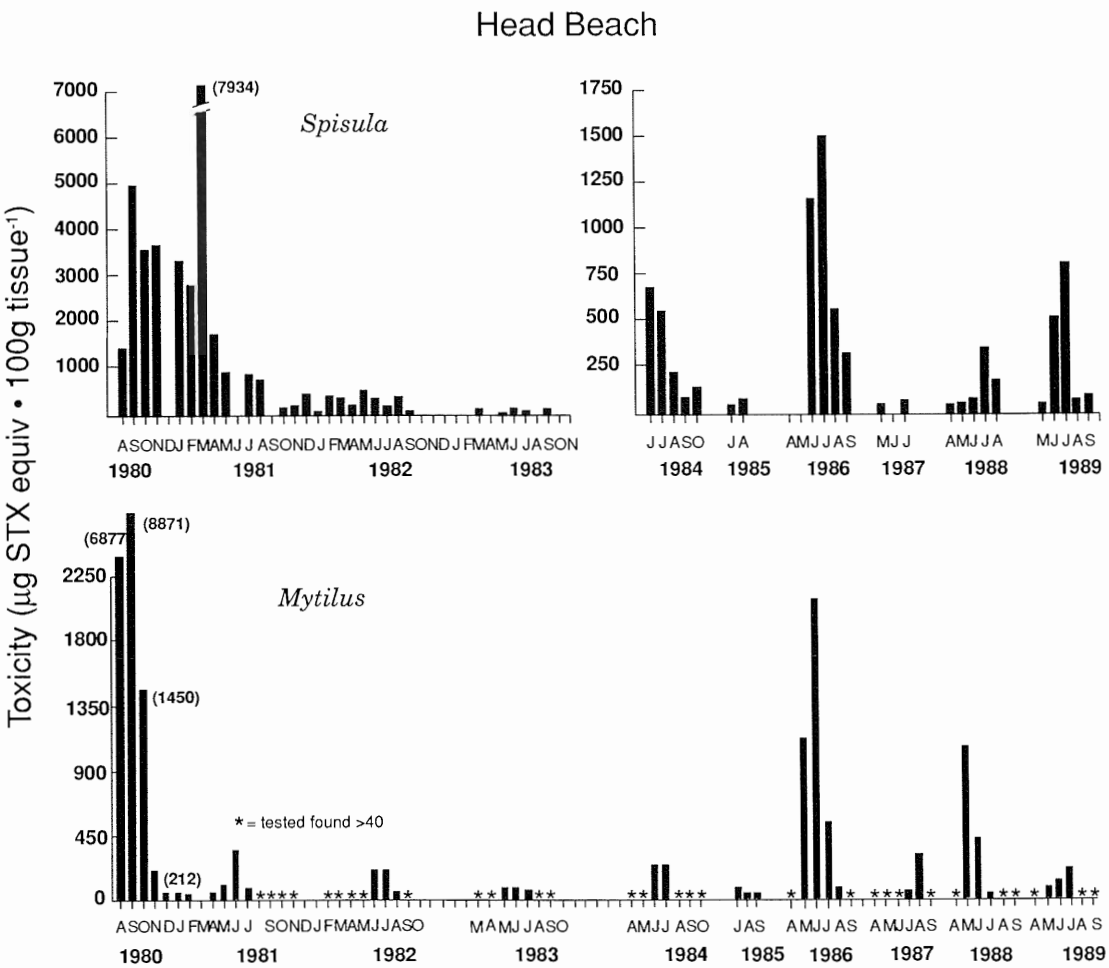

Fig. 9. Toxicity data for *Spisula solidissima* collected from Head Beach between August 1980 and September 1989. Each histogram represents a composite sample of 6 individual clams. Data for *Mytilus* represent a composite sample of 15–20 individual mussels at each sampling date.

during early spring and fall at inshore locations. Surfclams from offshore regions exhibited a large initial peak of PSP toxicity followed by a gradual decline over the ensuing months. There was no definitive evidence of further major blooms of toxic phytoplankton during the study period: the bloom responsible for the initial high toxicity recorded in surfclams from offshore regions may have been a rather anomalous incident.

Some of the variability in PSP toxicity found among individual surfclams can certainly be attributed to imprecision in the AOAC mouse bioassay [±20% CV; Adams and Furfari, 1984]. Furthermore, the exceedingly wide range in PSP toxicity among surfclams collected within several hundred meters suggests substantial small-scale differences in exposure to toxic dinoflagellates, in feeding behavior, or in the kinetics of toxin uptake and elimination. In any case, the high individual variation (beyond that accounted for by the mouse bioassay) poses a major impediment to the establishment of any routine monitoring program.

Anatomical Distribution of Toxins

Surfclams readily accumulate PSP toxins and distribute the toxins differentially among their tissues. Prolonged retention of PSP toxins by *Spisula* has been noted since early work on the anatomical distributon of toxins in this species by Medcof et al. [1947] and Prakash et al. [1971]. The data from a later detailed study by Blogoslawski and Stewart [1978] must be regarded as preliminary, since they were obtained at only two time points, for clams which had already experienced several months of detoxicification in the field. Medcof et al. [1947] reported PSP toxicities in mouse units (1 MU = 0.18–0.23 µg STX equiv depending upon the strain of mouse) of 29,400 in digestive glands, 59,000 in gills, 460 in adductor muscles, and 1,000 in the remaining

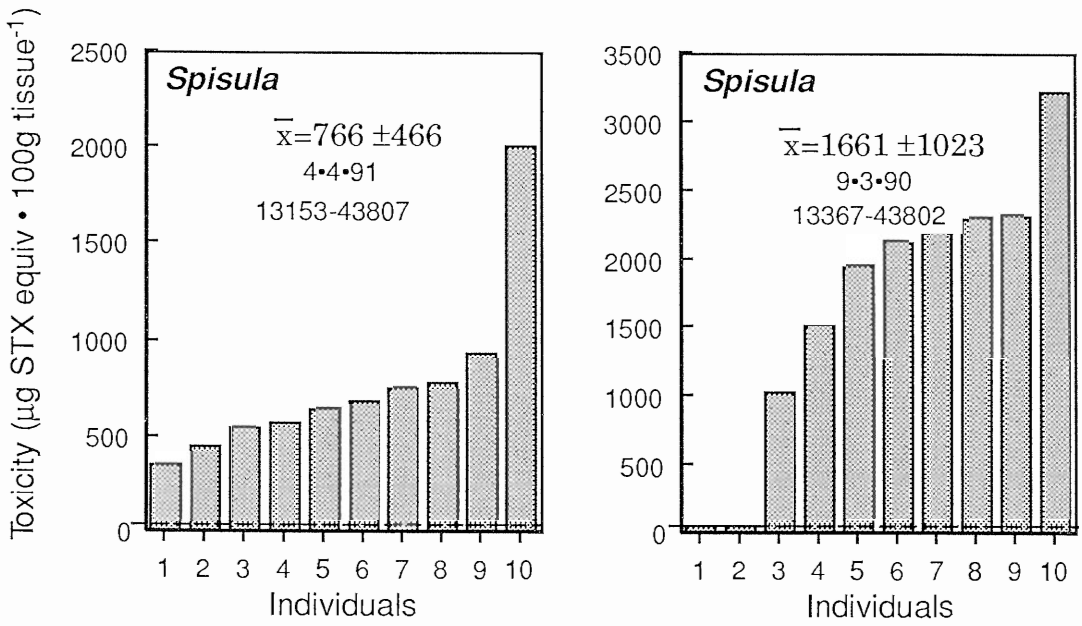

Fig. 10. Toxin levels of individual surfclams, *Spisula solidissima*. Only 2 of 42 data sets are shown. Each graph gives the mean toxin level, standard deviation, date of collection, and Loran C coordinates of the collection site [after White et al., 1993b].

parts. Prakash et al. [1971] reported a similar pattern and measured highest toxicity (μg STX equiv 100 g tissue^{-1}) in gills (9,450) and digestive glands (4,700), compared to lower levels in adductor muscles (74), siphons (32), and other parts (160) of the clam. No whole clams were analyzed. Both studies showed that the surfclams remained toxic from year to year, retaining toxins longer than any other mollusc tested.

We have reaffirmed the long-term retention of PSP toxins by surfclams for a period that may exceed 2 years. Moreover, we have extended our knowledge of the effect of toxic blooms on the shellfisheries of the Gulf of Maine, including the offshore fishing grounds of Georges Banks. As found in previous studies, PSP toxicity was not evenly distributed among the various tissues. Highest levels were recorded initially from the digestive glands and subsequently in gill and mantle tissues.

We note here a major difference in storage sites of PSP toxins between sea scallops *(Placopecten magellanicus)* and surfclams. While extremely high levels of toxicity (>20,000 μg STX equiv 10 g tissue^{-1}) were measured in gill tissue from surfclams, PSP-toxin levels as determined by HPLC-FD rarely exceeded the mouse bioassay detection limit (40 μg STX equiv 100 g tissue^{-1}) in gills of sea scallops from adjacent locations in the Gulf of Maine [Cembella et al., 1993b]. Clearly, there are differences in the metabolic fate of these toxins and, as with toxin accumulation in other species, this appears to be a species-specific phenomenon. The storage of PSP toxins among several tissue compartments, including the foot, mantle, and siphon, make harvesting such tissues during toxic events highly risky.

Toxin Elimination

Most shellfish species examined to date appear to eliminate PSP toxins associated with toxic phytoplankton blooms to below regulatory levels within a matter of weeks [see Shumway, 1990]. Three notable exceptions include the Alaska butter clam, *Saxidomus giganteus;* the sea scallop, *Placopecten magellanicus;* and the surfclam, *Spisula solidissima,* all of which are characterized by prolonged toxin retention (months to years) [Shumway and Cembella, 1993; Cembella et al., 1993a,b; Shumway, 1990; Shumway et al., 1988; Quayle, 1969; Beitler and Liston, 1990). In the butter clam, *Saxidomus giganteus,* the siphons are the main site of long-term accumulation [Pugsley, 1939; Beitler and Liston, 1990]. These clams are known to be toxic year-round in some regions, with toxicity levels in siphons which may exceed by several times the toxicity of the total remaining tissues. Most fluctuations in toxicity throughout the year occur in the siphons rather than in the whole bodies [Chambers and Magnusson, 1950].

For surfclams, no significant differences in the rate of PSP toxin elimination were found between specimens collected at regular intervals from stations on Georges Bank and those collected from the same stations and held in running seawater in the laboratory. Blogoslawski and Stewart [1978] suggested that ozone treatment might be useful as a means of depuration; however, these results were never

duplicated and were later contradicted in similar studies on *Mya arenaria,* the softshell clam. Depuration of surfclams is not considered to be a viable or economically feasible proposition.

Heating/Cooking

Only the foot, mantle, and siphon of large adult *Spisula* are usually consumed; however, with smaller individuals, the whole animal may be eaten. Although traditionally this seafood product is rarely eaten raw, the foot tissue is now being used for sushi in some markets. Adult surfclams are sold primarily to food processing companies, where they are used in chowders and stews, or where they are breaded and frozen as clam strips. Wilhelm and Martin [1991] reported that heat processing can substantially reduce the levels of PSP toxicity in *Spisula*. The reduction in toxicity through such processing is the combined result of removal of highly toxic tissues from the surfclams (e.g., digestive gland and gills) and of heat-induced loss of much of the remaining toxicity from the tissues. Promising results were obtained with clams containing PSP toxicity levels as much as five times the regulatory limit of 80 μg STX equiv 100 g tissue^{-1}. It must be stressed, however, that these results are preliminary and that the technique may only be effective when initial toxicity is relative low (<500 μg STX equiv 100 g^{-1}).

Management and Product Quality Problems

Spisula solidissima is regularly sampled from near-shore locations as part of the PSP toxin monitoring program in the State of Maine designed to provide optimum utilization and management of the resource during outbreaks of toxic algae. Recent aquaculture ventures with *Spisula* grown subtidally in inshore areas of the Gulf of Maine warrant continued monitoring of this species for the presence of PSP toxins. The prolonged retention of toxins by surfclams represents not only a loss of several million dollars in landed value, but also presents an acute management problem for both fisheries managers and public health officials. This problem is further exacerbated in the offshore populations by the difficulty in sampling these regions. While nearshore populations can be readily sampled and harvesting may be restricted during periods of toxicity, offshore clams cannot be collected as easily. The cost and logistical problems associated with disposal of contaminatd shellfish make dockside monitoring a tenuous proposition at best. Furthermore, because of the high variability among individual animals [White et al., 1993b; Cembella et al., 1993a] a large sample size is required for representative PSP toxin analysis. Finally, a lot-by-lot testing of product would still be needed if inspection were to be carried out dockside.

The possible effects of cooking or other heat treatment on the PSP-toxicity levels of surfclam tissues should be explored further, although it is already clear that this will only be useful when initial toxicity levels are low. Moreover, cooking usually reduces the value of the product and may not be an acceptable alternative for the consumer.

Aquaculturists should be made acutely aware of the potential problems associated with growing juvenile surfclams in areas prone to outbreaks of toxic algae. Given the prolonged PSP toxin retention time of surfclams, it is not inconceivable that an entire crop could be rendered useless by the transient occurrence of a single bloom.

ACKNOWLEDGMENTS

We are indebted to George Nardi, New England Fisheries Development Association; Jerry Levine, Blount Seafood; and the crew of the fishing vessel Wando River for making this project possible. Dave Whittaker and Kristin Geib braved the elements above and beyond the call of duty to collect offshore samples. J. Hurst, J. Barter, D. Clifford, and K. Geib provided laboratory assistance. Jim Rollins prepared the figures. We also acknowledge the cooperation of the FDA/NMFS Marine Biotoxin Survey Contract Committee. This study was funded in part by a U.S. Food and Drug Administration contract (223-89-4064) to the Massachusetts Health Research Institute and in part by grant/cooperative agreement (NA-90-AA-H-SK030) from the National Oceanic and Atmospheric Administration awarded to the New England Fisheries Development Association. The views expressed herein are those of the authors and do not necessarily reflect the views of NOAA or any of its subagencies. This is Bigelow Laboratory for Ocean Sciences Contribution 93008 and National Research Council Contribution 38034.

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