Response of saltmarsh fungi to the presence of mercury and polychlorinated biphenyls at a Superfund site

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Abstract: Ascomycetous fungi are the major decomposers of standing-decaying smooth cordgrass (Spartina alterniflora), the major grass of saltmarshes of the southeastern United States. In Brunswick, Georgia, smooth-cordgrass marshes have received a potentially severe chemical insult at the USEPA LCP Superfund Site [dumping of mercury and polychlorinated biphenyls (PCB)]. We have examined levels of living-fungal standing crop (as ergosterol) and fungal sexual productivity (rate of ascospore expulsion) in naturally decaying leaf blades of smooth cordgrass at the LCP site, at a nearby, moderately polluted site, and at a pristine site in Georgia. Although toxicant levels in sediments at the LCP site are very high (total Hg, to 71 µg g⁻¹ dry sediment; methylmercury, to 190 ng g⁻¹; PCB, to 156 µg g⁻¹), living-fungal biomass was higher at the LCP site (about 890 µg ergosterol g⁻¹ organic mass of decaying-leaf system, for dead blades on wholly dead shoots) than at the nearby moderately polluted site (about 630 µg g⁻¹) or the pristine site (about 590 µg g⁻¹). Ascospore release was also higher at LCP than at the pristine site. Only methylmercury at tens of ng g⁻¹ sediment gave any evidence of negative impact upon living-fungal crop. We speculate that urban/industrial nitrogen input was responsible for the higher biomass of fungi at the Brunswick sites, and that either the toxicants and/or the hypothesized N input were responsible for the major difference in cordgrass-fungal species composition found (replacement of Phaeosphaeria spartinicola by Phaeosphaeria halima) by sexual production rate, fungal biomass, LCP Chemical, methylmercury, Phaeosphaeria spartinicola, Spartina alterniflora

Key Words: ascospore-expulsion rate, fungal biomass, LCP Chemical, methylmercury, Phaeosphaeria spartinicola, Spartina alterniflora

INTRODUCTION

The highly productive saltmarshes of the southeastern United States are being exposed to degradative pressure due to human residential and commercial development (e.g., Vernberg et al., 1992). The degree of saltmarsh-environmental insult ranges from subtle residential/commercial influence (Porter et al., 1997) to direct input of highly toxic waste [mercury and polychlorinated biphenyls (PCB): Winger et al., 1993; Kannan et al., 1997; Sprenger et al., 1997; see also Abramowicz, 1995; Porcella et al., 1995; Lewis, 1997; Wu et al., 1998]. To gauge the impact of this development pressure, the influence of known saltmarsh pollutants upon key segments of the saltmarsh ecosystem must be determined. One important component of this ecosystem is the fungal decomposers of marshgrass (Newell, 1993a, 1996; Newell and Porter, 1999). From smooth cordgrass (Spartina alterniflora), the predominant grass of southeastern U.S. saltmarshes (Dardeau et al., 1992), ascomycetous decomposers have been provisionally estimated to produce about 730 g organic fungal mass m⁻² yr⁻¹, with a yield-efficiency from standing-dead cordgrass of approximately 50% (Newell and Porter, 1999). Marshgrass-ascomycetous production is known to flow to detritivorous marsh invertebrates (Newell and Bärlocher, 1993; Newell and Porter, 1999), which probably thereby makes this fungal production a major feeder into the trophic relay of marsh-animal yield to nearby waters (Kneib, 1997). Marine-restricted fungi are also known to be capable of forming complexes with heavy metals (e.g., Sunda and Gessner, 1989) and of producing polyphenol-oxidizing enzymes (Raghukumar et al., 1996).

We report herein our examination of the levels of marshgrass-fungal living standing crops (as ergosterol; Gessner and Newell, 1997; Fell and Newell, 1998) and rates of fungal sexual activity in severely contaminated, moderately contaminated, and uncontaminated saltmarshes on the southeastern coast of Georgia, USA. We also determined in parallel the densities of saltmarsh periwinkles, that could naturally maintain lower levels of fungal standing crop (Newell and Bärlocher, 1993).

MATERIALS AND METHODS

Sites.—The principal sampling transect was laid out at the United States Environmental Protection Agency (USEPA) LCP Chemical Superfund Site on the
were branching marshes. The wetland was sampled for its salt marsh plants, including smooth cordgrass, *Spartina alterniflora*. Elemental mercury and PCB (primarily Aroclor 1268), resulting from waste discharged into the salt marsh at the LCP Site during the period 1966–1971 and 1979–1995 (Winger et al., 1993; Sprenger et al., 1997). We arranged our transect including 9 stations in the smooth-cordgrass marsh, so that it crossed through a zone of high contamination (to 71 µg total Hg and 156 µg PCB per g dry sediment), with lower concentrations at either end. A second, 9-station transect was laid out in the saltmarsh directly across the Turtle River (1.9 km west of the LCP transect), outside the boundaries of the LCP Superfund Site. This “Cross-River” site was arranged so that marsh topography of the transect was similar to that of the LCP transect, but the contaminant concentrations at the Cross-River transect ranged only to 0.7 µg Hg and 0.4 µg PCB per g dry sediment. Elevations of the transect stations were surveyed within the transects with respect to the lowest station in each transect (range: LCP = 0.4 m; Cross-River = 0.2 m).

Industrial entities within 7 km of the LCP site along the east bank of the Turtle River and its eastern branching tributaries include a major pulp and paper mill, a large sewage-treatment plant, a major freight port, three seafood-processing facilities, and a major power plant. Lying along the east of the Turtle River are downtown Brunswick and its suburbs (population, 17000). At the head of a Turtle River creek (Burnett Creek) about 6 km upstream along the river is a second Superfund site (Brunswick-Escambia; pollutants: arsenic, creosote and pentachlorophenol).

Our third data set was collected within the marshes of Sapelo Island (31°23’N, 81°17’W), an area not directly impacted by industrial effluents (Chalmers, 1997). Maximum toxicant values found for 10 stations at Sapelo Island approximately corresponding to the marsh topography at the Brunswick stations were 0.05 µg Hg and 0.01 µg PCB per g dry sediment. The values for fungal variables from Sapelo marshes were collected separately as part of a multi-year analysis of dynamics of fungal living crop and fungal rates of production in standing-decaying smooth cordgrass (Newell and Porter, 1999). Samples were taken from three height-form plots (short, intermediate, tall) at each of three marsh watersheds (Doboy Sound, Upper Southend Creek, and Upper Duplin River), and data were pooled for comparison to the Brunswick samples.

**Sampling.**—In June 1997, at each station of the Brunswick transects, smooth-cordgrass canopy height (natural vertical extension) was measured, and the concentration of visible saltmarsh periwinkles (*Littoraria irrorata*) was determined by counting in three nonselectively placed ½-m² quadrats. Two partially-lived shoots bearing standing, decaying leaves were sheared from the rhizome at the sediment level (leaves of smooth cordgrass die in sequence from bottom of shoot toward top; Newell et al., 1998). Two wholly dead (brown) shoots were also severed at the sediment, and the four shoots were placed in labeled plastic bags for return to the laboratory (within 6 h). From the Sapelo stations, only wholly dead shoots were collected in May 1997. Temperature and salinity of creekwater at the Brunswick transects were 24.3°C and 25.3–26.9‰. Temperature and salinity of creekwater at the Sapelo sites were 20–24°C and 17–28‰.

After measuring the height above the sediment of the ligule (blade/sheath interface) of the lowest intact dead blade on each shoot, this blade was severed at the ligule. The severed blades were cut to 11.5-cm length from the ligule, rinsed (to remove clay films) for 10 sec under running tapwater (Newell and Palm, 1998), drained, measured for width at the 10-cm point from the ligule, and allowed to air dry at about 23°C and 58% relative humidity. Air drying is an effective means of preserving fungal ergosterol of smooth-cordgrass ascomycetes (Newell, 1995). Prior to air drying, each blade was examined under the stereomicroscope at × 12–50 for recording of the degree to which invertebrate shredders had grazed the blades. Grazing categories were: 1 = no grazing seen; 2 = slightly grazed (±20% of abaxial surface removed); 3 = grazed (≥20% of abaxial surface removed); 4 = shredded (≥2 splits at least 5 cm long).

**Ergosterol analysis.**—Ergosterol analysis of the samples was conducted as an index of living fungal mass (Gessner and Newell, 1997; Fell and Newell, 1998). Extraction and measurement by liquid chromatography were as described by Newell (1993b). Two-cm lengths were cut from the nonligule end of each air-dried blade sample and used for ergosterol extractions.

**Organic densities.**—One-cm lengths were cut from the nonligule ends of each air-dried blade. Dry mass was determined after microwave drying (6 min at full power in a 1000 W oven) (Newell, 1993b). Organic content was determined after ashing at 450°C for 4 h (Newell, 1993b).

**Ascospore expulsion.**—Rates of ejection of ascospores by the predominant ascospore-expelling ascomycetes in our sampled smooth-cordgrass blades were mea-
ured by the ascospore-capture technique of Newell and Wasowski (1995; after Aylor and Anagnostakis, 1991). Modifications of the Newell and Wasowski procedure were: blade pieces were wetted in tapwater for 10 sec rather than 10 min; target cover slips were located 7 mm beneath blades, rather than 1.5 cm; incubations were under 12/12 h on/off, 30 μE m⁻² s⁻¹ photosynthetically available radiation, rather than 10 μE; incubations were for 72 h rather than 168 h. Only blades from wholly dead shoots at six stations in each of the Brunswick transects were used in ascospore-capture incubations. Spores counted were for the following species: Phaeosphaeria spartinicola, Phaeosphaeria halima, and Mycosphaerella sp. 2 (Kohlmeier and Kohlmeier, 1979).

**Pollutant measurements.**—Five sediment cores (5 cm diam, 10 cm depth) were taken from each individual station along our sampling transects (center and 1 m north, south, east and west) and divided lengthwise into thirds. Thirds from each core at each station were combined and homogenized for 5 min using a paint shaker and the resultant composite sample was used for chemical analysis. All manipulation of core sections was done under a N₂ atmosphere.

For PCB analyses (after Kannan et al., 1997), approximately 10 g freeze-dried whole sediment was homogenized with kiln-fired Na₂SO₄ and extracted with 400 mL CH₂Cl₂ in a glass Soxhlet apparatus. Dibromo-2-octfluorobiphenyl (DBOFB) was added as a recovery surrogate prior to extraction. The CH₂Cl₂ extract was reduced and exchanged to hexane using a Kuderna-Danish type concentrator and treated with acid-activated copper powder to remove elemental sulfur. This extract was applied to a glass column slurry packed with 2 g silica gel activated at 130 C overnight. PCB were eluted with 100 mL hexane. This extract was reduced to approximately 1 mL in a TurboVap (Zymark Corp.).

Extracts were analyzed on Varian 3400CX gas chromatographs with electron capture (GC-ECD) and ion trap mass spectrometric (GC-ITMS) detectors. The columns used were 30 m × 0.25 mm coated with 0.25 μm of DB-5 and DB-XLB (J&W Scientific) for the ECD and ITMS, respectively. The GC-ECD oven was programmed from 120 C to 260 C at 2 C min⁻¹ and the GC-ITMS oven was programmed from 60 C to 280 C at 4 C min⁻¹. Both instruments were calibrated using serial dilutions of a 28-component PCB standard mixture [SRM2262; National Institute of Standards and Technology (NIST), Gaithersburg, Maryland, USA] and individual hepta- through nonachlorobiphenyl congeners (UltraScientific). The GC-ECD was used for primary quantitation and the GC-ITMS, operating in the scanning electron ionization mode, was used for congener confirmation.

Procedural blanks, PCB-spiked beach sand and a standard reference marine sediment (SRM1941a; NIST) were analyzed in conjunction with each batch of 15 marsh sediments. Blanks did not contain any target PCB above the nominal congener-specific detection limit (ca 1 ng g⁻¹ dry weight). Surrogate (DBOFB) and spiked PCB recoveries exceeded 80% in all samples. The mean recovery of PCB in SRM1941a with certified concentrations exceeded 90%.

Total mercury in sediment and biological tissue samples was determined using the procedures described in Smith (1993). The method involves spiking the sample with an enriched ²⁰³Hg isotope and microwave digestion with 5 mL of HNO₃. The samples are subsequently analyzed using cold vapor reduction into an ICP/MS (inductively coupled plasma/mass spectrometer). The isotopic ratio of ²⁰²Hg/²⁰³Hg is accurately determined and the concentration of total mercury is calculated from this ratio. The detection limit for the method has been determined to be approximately 5 ng g⁻¹.

The determination of methylmercury in sediment samples was accomplished using a combination of two published techniques (Horvat et al., 1993; Liang et al., 1994). In this method, methylmercury is determined by aqueous phase ethylation, cryogenic trapping and chromatographic desorption followed by detection with cold-vapor atomic fluorescence, with separation from interfering species by distillation.

**Statistical analyses.**—Correlation, regression, and analyses of variance (ANOVA) were performed using SPSS/PC+ Version 5.0 (Norusis, 1992). Significance level was set at ²⁰²Hg/²⁰³Hg is accurately determined and the concentration of total mercury is calculated from this ratio. The detection limit for the method has been determined to be approximately 5 ng g⁻¹.

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**RESULTS**

**Natural environmental variables.**—Average living-canopy height was about 20 cm greater at LCP than at Cross-River (x̄ = 97 ± 23 cm, versus 79 ± 20 cm), but neither canopy height nor snail concentration (x̄total = 22 ± 28 snails m⁻²) differed significantly be-
between the LCP and Cross-River sites ($P = 0.07$ and 0.33 respectively). Leaf blades were larger at LCP than at Cross-River ($x$, leaf width, LCP vs Cross-River, 1.17 ± 0.26 cm vs 0.96 ± 0.23 cm, $P = 0.01$). As expected (Newell, 1993a), height of ligules above the sediment was greater for dead blades on wholly-dead shoots than for partially-living shoots ($x$ = 19.6 ± 8.7 cm vs 9.6 ± 4.5 cm, $P < 0.001$), but it was also greater for LCP than Cross-River (17.7 ± 9.9 cm vs 10.7 ± 4.6 cm, $P = 0.001$). There was no significant difference between LCP and Cross-River sites with respect to shredder impact upon decaying blades ($x$, impact index, LCP, 1.9 ± 1.0; Cross-River, 1.8 ± 1.0; $P = 0.52$). As expected (Newell, 1993a), snail density at the Brunswick sites was correlated to canopy height ($r = -0.76$, $P < 0.001$, n = 33).

Average natural-variable values for the Sapelo marshes were close to those for the Brunswick sites ($x$, canopy height = 115 ± 41 cm; leaf width = 1.03 ± 0.34 cm; snail density = 24 ± 25 m$^{-2}$).

**Toxicants.**—Average sediment toxicant concentrations are given in Table I.

**Fungal mass versus natural variables.**—Ergosterol contents of dead blades on wholly dead shoots were not significantly correlated to leaf widths, canopy height, ligule height, snail density, or shredder impact. Ergosterol contents of dead blades on living shoots were correlated to leaf widths ($r = 0.77$, $P < 0.001$) and snail densities ($r = -0.47$, $P = 0.05$), but not canopy height, ligule height, or shredder impact. When both leaf width and snail density were entered into stepwise multiple regression against ergosterol of dead blades on living shoots, significance of snail density was reduced ($P = 0.08$).

**Fungal variables versus sites and pollutants.**—Ergosterol content of standing-decaying blades was higher for the LCP transect, and for dead blades on wholly-dead shoots (Fig. 1) (2-way ANOVA, $P_{site} = 0.008$; $P_{blade-type} < 0.001$; $P_{interaction} = 0.66$, n = 66 blades total). Ergosterol content of Sapelo blades (dead-on-dead, n = 27 blades) was the lowest of the three sites (Fig. 1).

Correlation analysis revealed that there was no statistically significant relationship ($n = 17$; $r = -0.10$; $P = 0.72$) between ergosterol content of decaying blades and total mercury content of sediment at the LCP site, where the range of mercury contents was much greater (to 71 µg g$^{-1}$ dry sediment) than at the Cross-River site (to 0.7 µg g$^{-1}$). The same was true for methylmercury ($r = -0.18$, $P = 0.50$) and PCB ($r = -0.16$, $P = 0.54$) (range of methylmercury contents: LCP, to 84 ng g$^{-1}$ dry sediment, Cross-River, to 15 ng g$^{-1}$) (range of total PCB contents: LCP, to 156 µg g$^{-1}$ dry sediment; Cross-River, to 0.4 µg g$^{-1}$). There was one subset of data for ergosterol content that suggests toxic impact upon fungi; it was the data for dead blades on living shoots at LCP, correlated to methylmercury content of sediments (with logarithmic transformation, which gave a closer fit): n = 9; $r = -0.79$; $P = 0.01$. Leaf width for dead-on-live blades was also significantly correlated to methylmercury level at the LCP site: $r = -0.75$, $P = 0.02$. When leaf width and log$_{10}$ methylmercury level were brought into a stepwise regression, with log$_{10}$ ergosterol of dead-on-live as the dependent variable, only methylmercury was a significant variable ($P$ reduced to 0.48 for leaf width). Neither leaf width nor ergosterol content were correlated to survey elevation for dead-on-live blades at LCP (leaf width: $r = 0.05$, $P = 0.90$; ergosterol: $r = 0.41$, $P = 0.27$). At the Cross-River site, there was no significant relationship between er-

**Table I.** Concentrations (x ± SD) of toxicants per g dry sediment at the three saltmarsh sites sampled (LCP = LCP Chemical Superfund site; CR = Cross-River, 2 km west of LCP; Sapelo = Sapelo Island, nonindustrial site)

<table>
<thead>
<tr>
<th>Site</th>
<th>Total mercury</th>
<th>Methylmercury</th>
<th>PCB</th>
</tr>
</thead>
<tbody>
<tr>
<td>LCP</td>
<td>21 ± 22 µg</td>
<td>69 ± 50 ng</td>
<td>53 ± 53 µg</td>
</tr>
<tr>
<td>CR</td>
<td>0.5 ± 0.1 µg</td>
<td>10 ± 4 ng</td>
<td>0.3 ± 0.1 µg</td>
</tr>
<tr>
<td>Sapelo</td>
<td>36 ± 12 ng</td>
<td>0.6 ± 0.5 ng</td>
<td>5 ± 2 ng</td>
</tr>
</tbody>
</table>

**Fig. 1.** Ergosterol content (x ± SD) of decaying blades of smooth cordgrass (Spartina alterniflora) in highly polluted (LCP Chemical Superfund Site), moderately polluted (CR, Cross-River, 2 km west of LCP), and unpolluted (Sapelo Island) saltmarsh sites. Open bars, decaying leaves on partially-living shoots (no data for Sapelo); lined bars, decaying leaves on wholly-dead shoots.
gosterol content of dead blades on living shoots with sediment methylmercury \((n = 9; r = -0.06; P = 0.88)\).

Rates of ascospore expulsion were not significantly different between the LCP and Cross-River sites \((\bar{x}_{\text{total}} = 5494 \pm 10309\) spores \(\text{cm}^{-2} \text{72h}^{-1}; n = 12; P = 0.80)\). There were no significant correlations found between rates of ascospore expulsion and toxicant variables. Average rate of ascospore release for Sapelo samples was 2593 ± 2346 spores \(\text{cm}^{-2} \text{72h}^{-1}\). At the Sapelo stations, \(P. spartincola\), Mycosphaerella sp. 2, and \(P. halima\) accounted for 52, 48, and 0% of the spores counted, respectively. At the Brunswick stations, \(P. spartincola\), Mycosphaerella sp. 2, and \(P. halima\) accounted for <1, 68, and 32% of the spores counted, respectively.

**DISCUSSION**

Of the three toxicants (PCB, total mercury, and methylmercury) at the LCP Superfund Site, only methylmercury gave evidence of negative influence upon standing crop of living-fungal mass. The closer fit of log-transformed data suggests that the toxic impact of methylmercury may begin to rise nonlinearly at levels of tens of ng g\(^{-1}\) sediment. Since relationship of leaf width of smooth cordgrass was also negative with methylmercury at tens of ng g\(^{-1}\) sediment, and declining leaf width is associated with increased levels of stress upon the grass (e.g., Dai and Wiegert, 1997), it may be that when methylmercury concentrations in the substratum are very high, both the living grass and its decomposers are inhibited. Since ergosterol levels in decaying leaves that had resided at an average of twice as high above the sediment (dead blades on dead shoots versus dead blades on living shoots) were not apparently impacted by sediment methylmercury, it may be that only the fungi in the lowest decaying leaves on shoots, close to sediments, are affected.

It should be noted that the reduction seen in ergosterol content of dead-on-live blades at LCP was from levels higher than found at the Cross-River site, so the potential methylmercury effect is inhibition of the extra fungal accumulation at the LCP site. We must remind readers here that what we measured was standing crop of living fungi (as ergosterol). It is possible that lower fungal biomass at stations with very high methylmercury concentrations was a result of higher rates of fungal output (e.g., mycophagy, autolysis), rather than toxically-inhibited accretion, as we are assuming for the discussion in this paragraph. Also, it is possible that some of the methylmercury present was a fungal product (Gadd, 1993).

We found no impact of mercuric or PCB toxicants on living-fungal standing crop when comparing the transects of the LCP and Cross-River sites, even though the toxicant concentrations in the sediments of the LCP site were 7 to 177-fold greater. There are several possible hypothetical explanations of this finding, including the following: (i) the toxicants could be so tightly bound to sediment particles, that they have no influence upon grass shoots or shoot-decomposers (Winger et al., 1993; Kannan et al., 1997; Wainwright and Gadd, 1997). Since marsh-clay sediments are regularly resuspended and deposited upon smooth-cordgrass shoots (Newell et al., 1992), this explanation seems unlikely. Furthermore, both grass tissue and tissue of marsh periwinkles (which eat decaying cordgrass: Newell and Bärlocher, 1993) at the LCP sites contain elevated mercury and PCB contents that are correlated to sediment toxicant concentrations (living cordgrass, Hg to 10 \(\mu\)g g\(^{-1}\) dry tissue, PCB to 19 \(\mu\)g g\(^{-1}\); snails, Hg to 40 \(\mu\)g g\(^{-1}\), PCB to 59 \(\mu\)g g\(^{-1}\): Sprenger et al., 1997). Mercury and PCB contents of smooth-cordgrass blades are ≤70 ng g\(^{-1}\) and 25 ng g\(^{-1}\) dry mass, respectively, at unpolluted sites (Newell et al., 1982; Sprenger et al., 1997). Mercury and PCB contents of saltmarsh periwinkles at low-pollution sites are ≤600 ng g\(^{-1}\) and 50 ng g\(^{-1}\) dry tissue, respectively (Sprenger et al., 1997).

(ii) Saltmarsh ascomycetes could be resilient to mercuric and PCB poisoning. Since lignocellulolytic fungi can degrade PCB (e.g. Dietrich et al., 1995; Novotny et al., 1997), and cordgrass ascomycetes are lignocellulolytic (Newell et al., 1996), it perhaps is sensible that PCB would not be saltmarsh-fungitoxic. Hicks and Newell (1984) discovered that Hg at a concentration of approximately 90 \(\mu\)M g\(^{-1}\) dry leaf (740 \(\mu\)M Hg L\(^{-1}\) seawater) of smooth cordgrass in seawater/leaf microcosms did not inhibit growth or respiration of a saltmarsh ascomycete (*Phaeosphaeria spartincola*, as *P. typhina*; Leuchtmann and Newell, 1991), and may have caused a positive shift in the ratio of fungal:bacterial respiration [see also Purkayastha and Mitra, 1992: production of 1.4 kg of basidiomata by *Volvariella volvacea* in straw-compost frames (1 m\(^2\)) sprayed with 2 L of a solution of 2 mg L\(^{-1}\) mercuric chloride (control production, 2.1 kg)]; Høslund, 1995: median highest in vitro concentration of Hg permitting growth for 15 litter-decomposing basidiomycetes, 200 mg L\(^{-1}\); Krantz-Rülcker et al., 1996: no in vitro toxic effect of Hg below 200 to 2000 \(\mu\)g L\(^{-1}\) in nonchelating glucose medium for three species of fungi; review: Wainwright and Gadd (1997)]. Insensitivity of cordgrass ascomycetes at the LCP site (total Hg concentration 4 to 71 \(\mu\)g g\(^{-1}\) dry sediment) is consistent with these findings.

(iii) Nitrogen subsidy in the urban/industrial environment could boost fungal ability to resist toxicant...
poisoning ("dual enrichment/destructive disturbance": Wainwright and Gadd, 1997). Nutritional sufficiency can lead to greater resistance of fungi to metal toxicity (Wainwright and Gadd, 1997), and productivity of saltmarsh ascomycetes is known to be nitrogen limited (Newell, 1996). The high nitrogen-release potential of the urban/industrial facilities (see McClelland et al., 1997), including a sewage-treatment plant, on the same tidal riverbank as the LCP site, may well explain the high levels of living fungal standing crops at the LCP site (Fig. 1), and the hypothetical nitrogen sufficiency could have added to the toxicant resilience of the cordgrass ascomycetes. Another potential indicator of nitrogen subsidy at the LCP versus the Cross-River site was the 20% wider leaves and 65% higher ligule heights of the cordgrass shoots (Dai and Wiegert, 1997).

(iv) Rates of mycophagy could be reduced by toxic-pollutant impact on fungus-eating invertebrates. For the major known saltmarsh mycophagous invertebrate (saltmarsh periwinkles), we did not find any significant reduction in densities at the Brunswick sites. Kneib and Pattavina (unpubl. data) found no obvious reductions in densities or sizes of a mycophagous amphipod (Uhlrotcheta spartinopha: Kneib et al., 1997) at the Brunswick sites, and Sprenger et al. (1997) report that a test amphipod (Leptocheirus plumulosus) showed no acute poisoning effects when exposed to sediments from the LCP site. However, since there may be other saltmarsh mycophagous invertebrates than those currently identified (Newell, 1996; Newell and Porter, 1999), the possibility that some portion of the mycophagous assemblage of the LCP marsh has been killed or suppressed cannot be ruled out. Note that Sprenger et al. (1997) report the development of lesions and mortalities to standard fish (Oryzias latipes, Japanese medaka) embryos exposed to sediments from the LCP site, that were not found for embryos exposed to control-marsh sediments, and Winger et al. (1993) found feeding inhibition of a freshwater amphipod (Hyalella azteca) by LCP sediments.

(v) Environmental conditions of industrial/urban marsh sites could induce changes in species composition of saltmarsh-fungal assemblages. We found very different compositions among the species involved in ascospore expulsion at the industrial, Brunswick sites versus the undeveloped, Sapelo sites (namely, nearly complete replacement of P. spartinicola by P. halima at the Brunswick sites). Since both toxicant (Hg species and PCB) and probably dissolved nitrogen concentrations were much higher at the industrial sites, it may be that human perturbations underlay this species alteration. It could be that P. halima is better adapted to eutrophy and/or toxic pollution, and that under urban/industrial conditions, a P. halima-dominated mycoflora is capable of accumulating higher living crops than the P. spartinicola-dominated mycoflora can (pollution-driven shift to more active species: Wainwright and Gadd, 1997). More sampling of urban/industrial versus pristine cordgrass marshes is needed to test this species-replacement hypothesis.

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LITERATURE CITED


Gessner, M. O., and S. Y. Newell. 1997. Bulk quantitative methods for the examination of eukaryotic organoo-


