Final Report

A Survey of the Shallow Water and Intertidal Benthic Invertebrates at Three Sites in the Vicinity of The Chalk Point Steam Electric Station

A Study Conducted for The Patuxent River Damage Assessment

of the Chalk Point Oil Spill

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In April 2000, oil was released into Swanson Creek at the site of the Chalk Point Steam Electric Station (Figure 1). The oil spread into the Patuxent River and was carried down the river more than 25 km, potentially impacting coastal habitats within this area. This study was designed as a simple survey to measure the abundance of infaunal invertebrates in sediments from three low intertidal - shallow subtidal sites in the vicinity of the release. The three sites that were sampled included: 1) a reference or control site in Hunting Creek that was up-river from the release point and not impacted by oil, 2) a moderately-oiled site in Trent Hall Creek down-river from the release point, and 3) a heavily-oiled site in Swanson Creek, the location of the release. Samples were collected in late September 2000, approximately 5 months after the oil was released.

In order to compare the fauna among the three sites, 10 replicate core samples were taken at each of the three sites. To minimize the natural variation among sites every effort was made to select sampling locations within each creek that were at similar tidal heights, were located adjacent to marsh habitat, and had similar sediment types. All 3 sites were sampled on the same day within 3 h of each other.

Methods

Field Collections

All samples were collected at the three sites on 20 September 2000. To prevent any possible contamination of samples, the sites were sampled in order of their exposure to oil. Samples were first taken at the control site in Hunting Creek (76° 39' 26" Long, 38° 33' 26" Lat), then in Trent Hall Creek (76° 40' 52" Long, 38° 28' 45" Lat), and finally in Swanson Creek (76° 41' 34" Long, 38° 32' 39" Lat). At each site we measured water temperature and salinity using a YSI 85 meter. Next a 20-m transect parallel to the shore was marked at water depths of 15 - 37 cm. Because the samples were collected on a receding tide the mean water depth also was decreased from a mean of 35.5 $\pm$ 1.0 (SE) cm in Hunting Creek to 25.5 $\pm$ 0.6 cm in Trent Hall Creek to 18.6 $\pm$ 0.8 cm in Swanson Creek in an effort to sample at the same depth relative to Mean Low Water. Ten sites along each transect were chosen randomly and marked. At each marked sampling site a 15-cm diameter core sample was taken at a distance of 50 cm perpendicular to the transect. The shoreward or seaward direction of the 50-cm displacement was chosen randomly.

For each sample we pushed the corer approximately 20 cm into the sediment. Before the corer and sediment core were extracted, we measured the water depth and then took a 200 g sediment sample adjacent to the core tube. This sample was given to a representative of the Trustee Council for potential chemical analysis. After this sample was taken, the corer was extracted. Each core sampled the sediment to a depth of approximately 20 cm resulting in our sampling 3.5 l of sediment. The sediment collected from each replicate core was placed in a separate plastic lidded container, returned to the laboratory and refrigerated.

Laboratory Analyses

Initial processing of the cores, consisting of sub-sample collection and sieving of the bulk of the sediment for macroinvertebrates, was done within 24 h of collection. Sub-samples were taken from the mixed core sediments in each container. Sub-sampling included the collection of 100 g of the sediment for grain size analysis and 20 ml that was frozen for subsequent analysis of organic matter content. These three types of samples were collected as follows:

1. Macroinvertebrate Samples. After the sub-samples were taken, the remaining bulk of the sediment was sieved using a 500 µm screen. All material retained on the screen was fixed in formalin and stained with Rose Bengal. After approximately 1 wk samples were transferred into alcohol. Subsequently, samples were examined using a dissecting microscope and all macroinvertebrate organisms were identified and counted.

2. Sediment Grain Size Analysis. The 100 g samples were sieved using a series of stacked screens with mesh sizes of 2000, 1000, 500, 250, 125 and 62 µm (Butler and McManus 1979, Buchanan 1984). The sediment was sieved wet to avoid aggregation of the finer particles as a consequence of drying. All water and sediment passing through the 62 µm sieve were retained and filtered to retrieve the fine silt - clay sediments (Butler and McManus 1979). The proportion of the sample in each of the 7 size classes was determined by volume and dry weight and used to characterize the sediment and any differences within and among the three field sites.

3. Organic Matter Content. The 20-ml frozen samples were analyzed using the ignition method (McCave 1979, Rosa et al. 1991) for organic matter content. Each sample was dried at 60°C, weighed, combusted at 450° C, and then re-weighed. The difference before and after combustion was used to estimate the proportion of organic matter in each sample. Combustion was at 450° C to avoid the combustion of any shell (carbonate) material in the samples (Rosa et al. 1991).

Data Analyses

The objective of all statistical analyses was to compare the three sites and determine whether they differed from one another. All analyses were conducted using JMP 4.0 statistical software (SAS Institute 2000).

The main type of analysis conducted was a standard one-way analysis of variance (ANOVA). Differences among the three stations for individual taxa, trophic groups, and species richness were all analyzed using one-way ANOVA. For all analyses in which a significant difference was found ($p<0.05$), a Tukey multiple comparison test (Tukey 1991, Kramer 1956, SAS Institute 2000) was used to contrast the individual stations.

The similarity in community composition among the samples collected at the three sites was also analyzed using hierarchical clustering using Ward's minimum variance method for linking clusters (SAS Institute 2000). Three types of analyses were conducted: 1) similarity among individual samples based on the abundances of all taxa in the samples, 2) similarity among individual samples based on the abundances of the 8 most abundant or dominant species, and 3) similarity among the dominant species based on their abundances in the samples.

Results

Environmental Differences

A limited number of environmental parameters were measured for each site. These included the water temperature and salinity at each site and the sediment grain size distribution and organic matter content of each sample. Water temperature varied slightly from 21.3°C at Hunting Creek to 23.8°C at Trent Hall to 25.5°C at Swanson Creek. These differences probably resulted from the order in which the sites were sampled with Hunting Creek being sampled first in the morning and Swanson Creek being sampled in the mid-afternoon when air temperatures had increased. Also the sampling day was sunny and solar radiation could be expected to warm the shallow water at the sites. Unlike temperature there was a substantial difference in salinity among the sites with the salinity at Trent Hall (9.6 ppt) being almost double that at Hunting Creek (5.3 ppt) and Swanson Creek (5.6 ppt).

The analysis of sediment grain size indicated that there was little difference among the sites in the types of sediments that were present. When analyzed by dry weight (Figure 2), Swanson Creek had a significantly lower mean percent of it's sediment in the 125 μm class than the other two sites and Trent Hall had a significantly lower percentage in the >2000 μm class. Likewise when analyzed by volume (Figure 3), Swanson Creek had a higher percentage of silt-clay (<63 μm) than the other sites, but this was balanced by lower percentages in the 63 μm and 125 μm classes. Overall, these differences indicate little, if any, meaningful differences in the sediments at the three sampling sites.

The analyses of organic matter content in the sediments also indicated little difference among the sites (Figure 4). The mean percent organic matter was highest in the sediments from Swanson Creek (5.3% ± 1.4 SE) compared to those from Hunting Creek (3.2% ± 0.7) and Trent Hall Creek (2.6% ± 0.8). However, these differences were not statistically significant.

Distribution of Dominant Taxa

The fauna sampled at the three sites was fairly typical of shallow subtidal areas within the Patuxent River and adjacent Chesapeake Bay. Given the high spatial and temporal variability in the abundances of these species, the densities observed at the three sites were within the ranges seen in the region (Holland 1985, Holland et al. 1977a, 1977b, 1980), The principal macrofaunal invertebrate groups were polychaete annelids, nemerteans, molluscs, and crustaceans. Meiofaunal groups (e.g. copepods, nematodes) were also sampled. However, because we used a 500 μm screen in processing the samples, the much smaller organisms in these groups were under-represented. Below we compare the abundances of the dominant taxa among the three sites. All statistical inferences are based on one-way ANOVA's with $p<0.05$.

Polychaetes. Polychaetes were the most abundant taxa found at all three sites. The polychaete fauna consisted of two species, the sub-surface deposit feeding *Heteromastus filiformis* and *Nereis succinea*, an omnivore which also preys on other infauna. Surprisingly, no other polychaete species were found, including any surface deposit-feeding species.

Both species of polychaetes were significantly more abundant in Trent Hall Creek (Figure 5). Their densities were almost 3 times greater at this site than the other two sites. They were the two most abundant species at all three sites.

Nemerteans. The only nemertean found at the sites was the predator *Micrura leidyi*. This species was in fairly low abundance (Figure 5) with no significant difference found among the three sites.

Molluscs. Three species of molluscs were found in samples from the three sites. These included the bivalves *Macoma balthica* and *Rangia cuneata* and the gastropod *Hydrobia* sp. *Rangia* is a suspension-feeder typically most abundant in low salinity areas. Although *Macoma* can also suspension-feed it is principally a surface deposit-feeder, especially in muddy sediments (Olafsson 1986, 1989). *Hydrobia* is an omnivorous grazer on the surface sediments.

The distribution of *Macoma* was similar to the two species of polychaetes (Figure 6). It was significantly more abundant in Trent Hall Creek, with densities > 3 times higher there than in the other two sites. *Rangia* exhibited a different pattern with its density being significantly higher in Hunting Creek than in Trent Hall Creek (Figure 6). Densities in Swanson Creek were intermediate between the other two stations and not significantly different from either. Finally, *Hydrobia* was found only in Swanson Creek and its densities were significantly higher there than at the other two sites (Figure 6).

Arthropods. Arthropods formed the most diverse taxonomic group of macrofauna at the three sites, including the cumacean *Leucon americanus*, the amphipod *Leptocheirus plumulosus*, the isopods *Edotea triloba* and *Cyathura polita*, the grass shrimp *Palaemonetes pugio*, the mud crab *Rhithropanopeus harrisii*, chironomid insect larvae, and the barnacle *Balanus improvisus*. Except for the suspension-feeding barnacle, all of these species graze on the sediment surface with *Leucon* and *Leptocheirus* considered to be surface deposit-feeders and the remaining species considered omnivorous (Chesapeake Bay Program 2000).

Of the arthropods, the two isopods, *Edotea* and *Cyathura* were most abundant. *Cyathura* exhibited no significant difference among the three sites (Figure 7). *Edotea*, on the other hand, was significantly less abundant in Trent Hall Creek than at the other two sites (Figure 7).

The only other arthropod found to vary significantly among the sites was the cumacean *Leucon*. *Leucon* was significantly more abundant in Trent Hall Creek than in Hunting Creek, where it was absent (Figure 7). Its abundance in Swanson Creek was intermediate and not significantly different from either of the other two sites.

The remaining arthropod species were in fairly low densities and exhibited no significant differences among the three sites (Figure 8). The amphipod *Leptocheirus* was found only in Hunting Creek and the mud crab *Rhithropanopeus* was found only in Trent Hall Creek and Swanson Creek.

Meiofauna. Both meiofaunal groups, copepods and nematodes, were more abundant in Trent Hall Creek than the other two sites (Figures 9). However, these differences were not statistically significant.

Distribution of Larger Taxonomic Groups

The distribution among the three sites of each of the major taxonomic groups (polychaetes, molluscs, arthropods, and meiofauna) was also compared. Polychaetes were greater than 50% of the total fauna at all three sites with their proportional abundance being significantly greater in Trent Hall Creek than at the other two sites (Figure 10). Molluscs comprised 10-20% of the fauna at all three stations (Figure 10) and meiofauna were 5-10% of the sampled fauna (Figure 10). Neither of these groups differed significantly among the sites. Finally, arthropods were >20% of the fauna in Swanson and Hunting Creeks (Figure 10). These percentages were significantly greater than the <10% found for Trent Hall Creek.

Trophic Group Differences

The taxa in each sample were also pooled by trophic group or guild in order to examine whether any potential differences existed among the three sites in trophic dynamics. The feeding guild of each taxon was assigned using the Chesapeake Bay Program classification system (Chesapeake Bay Program 2000). Based on this system the sites had four major trophic groups; deep, sub-surface deposit feeders (*Heteromastus*), surface deposit feeders (*Leptocheirus, Leucon, Macoma*), omnivores/carnivores (chironomids, *Cyathura, Edotea, Hydrobia, Micrura, Nereis, Palaemonetes, Rhithropanopeus*), and suspension feeders (*Rangia, Balanus*).

Both the sub-surface deposit feeder (Figure 11) and surface deposit feeder (Figure 11) groups were significantly more abundant in Trent Hall Creek. Much of this results from the high densities of *Heteromastus* and *Macoma* in Trent Hall Creek. Omnivores (Figure 11) were significantly more abundant in Trent Hall Creek (largely from high *Nereis* densities) than in Hunting Creek with Swanson Creek being intermediate and not significantly different from either of the other two stations. Finally, suspension feeders (Figure 11) were significantly more abundant in Hunting Creek (as a consequence of high *Rangia* densities) than Trent Hall Creek with Swanson Creek being intermediate.

Community Patterns

Overall, total faunal densities in Trent Hall Creek were approximately double those found at the other two sites and significantly higher (Figure 12). This was largely a consequence of the high abundances of *Heteromastus, Nereis*, and *Macoma* in Trent Hall Creek. However, this difference in abundances was not reflected in the species richness of the sites. The mean number of species found at the three sites varied between 7 and 9 with no significant differences found among the sites (Figure 12).

Hierarchical cluster analyses generally showed the same types of patterns for communities as was seen for individual taxa. Cluster analyses were done using the abundance data for all taxa (Figure 13) and also with only the most abundant macrofaunal taxa (*Nereis, Heteromastus, Macoma, Rangia, Micrura, Leucon, Hydrobia, Edotea, Cyathura*) to weight the analysis towards dominant species (Figure 14). In general, the results for both analyses were similar and demonstrated the community differences among the three stations. When all taxa were included (Figure 13) 5 of the 10 Trent Hall samples cluster together as a group distinct from all other samples. Likewise, 7 of the 10 Swanson Creek samples along with 1 Hunting Creek sample formed a second distinct grouping. The remaining samples formed a larger, mixed group dominated by samples from Hunting Creek. However, within this group there are 4 distinct sub-groups of a) 5 Hunting Creek samples, b) 3 Trent Hall samples, c) a single Hunting Creek sample, and d) a mixed group of the remaining samples. When the analysis was repeated with only the dominant species (Figure 14) 7 of the Trent Hall samples formed a distinct cluster. The cluster of 7 Swanson Creek samples and 1 Hunting Creek sample did not change and there was a grouping of 2 Hunting Creek samples. The remaining grouping had similar but less distinct sub-groupings as in the previous analyses. Overall these analyses suggest distinct differences in the communities at the three sites but with a great deal of overlap.

Finally, the clustering of species based on their distributions among the 30 samples was also examined, one comparing all species (Figure 15) and the other (Figure 16) contrasting the most dominant taxa (*Nereis, Heteromastus, Macoma, Rangia, Hydrobia, Edotea, Cyathura*, and nematodes). Both analyses show much the same pattern. When all species were included (Figure 15), 5 major clusters were found including a) a distinct cluster with *Heteromastus* and *Nereis*, the two most abundant species that were also in greatest abundance in Trent Hall Creek, b) a cluster of three species that were dominant in Hunting Creek (*Rangia*, *Edotea*, and *Cyathura*), c) a cluster of lower density taxa that were also most abundant in Trent Hall Creek (*Macoma*, nematodes, and copepods), d) *Hydrobia* that was only found in Swanson Creek, and e) a large group of the remaining species that generally had low densities and a mixed distribution among the sites. When the 8 most dominant taxa were analyzed separately (Figure 16) the pattern was largely the same with a) the distinct Trent Hall dominant *Heteromastus, Nereis* cluster, b) the Hunting Creek dominant *Rangia, Edotea* cluster, and c) a mixed cluster species more prevalent in Trent Hall Creek (*Cyathura, Macoma,* and nematodes) or Swanson Creek (*Hydrobia*).

Discussion

This study compares the abundances of sediment-dwelling invertebrates at three sites on one sampling date. Five months prior to the sampling date, the three sites selected varied in their exposure to oil with Swanson Creek having heavy exposure, Trent Hall Creek moderate exposure, and Hunting Creek no exposure. Except for salinity, most other environmental variables measured appeared to be similar between the sites. Sediment grain size distributions differed slightly but sediments at all three sampling sites can be classified as muddy sands, similar to western shore sands and muds of Chesapeake Bay (Ryan 1953). Swanson Creek did have a slightly higher percentage of silt-clay. Organic matter content did not vary significantly among the three sites. Tidal heights of the sampling sites and measured water temperatures also varied little. The only measured environmental parameter that did differ among the sites was salinity with Trent Hall Creek having a salinity much higher than the other two sites. Given the location of Trent Hall Creek farther down the Patuxent River, it is likely that it has a higher mean salinity than the other two sites.

All three sites had taxa that are characteristic of shallow sub-tidal and intertidal habitats within the region (e.g. Holland 1985, Holland et al. 1977a, b, 1980, Marsh and Tenore 1990, McErlean 1964, Kemp et al. 2000) as well as in other estuarine systems (e.g. Sanders et al. 1965). Species composition at the sites was also similar to that found in earlier Academy of Natural Sciences surveys of shallow subtidal areas in the immediate vicinity of the present study (e.g. ANS 1971, 1978a, 1978b) as well as in intertidal habitats in some Patuxent River marshes (Osman and Whitlatch 1998). In a study of infaunal populations in the lower Patuxent River, Marsh and Tenore (1990) found that four species, *Nereis, Macoma, Leptocheirus*, and the spionid polychaete *Streblospio benedicti*, accounted for >90% of the community biomass and two of these, *Nereis* and *Macoma* were among the 3 most dominant species at the study sites. The other two species in the lower Patuxent study are opportunistic species that recruit in early summer. They often suffer high mortalities that result in low abundances by late August (Marsh and Tenore 1990), prior to the time of sampling in our study. *Nereis* and *Macoma* recruit in the late summer and fall (Holland et al. 1977b), making it likely for them to be in higher densities at the time we sampled. McErlean (1964) also found *Macoma* densities in the Patuxent River to be highly variable but he reported similar densities to those we found in the vicinity of the three sites.

Although many of the same species occurred at all three sites, the densities of many taxa at the three sites were clearly different with Trent Hall Creek having much higher overall densities (Figure 12) than the other two sites. All three sites were dominated by polychaetes, but this dominance was significantly greater in Trent Hall Creek (Figure 10). Both *Heteromastus* and *Nereis* had densities in Trent Hall Creek that were 3 times greater than at the other two sites (Figure 5). Mollusc densities did not differ among the sites (Figure 10), but the dominant species varied among the sites. *Macoma* was significantly more abundant in Trent Hall Creek, *Rangia* in Hunting Creek, and *Hydrobia* in Swanson Creek (Figure 6). Although arthropods had the greatest number of species, most species were in fairly low abundance at the sites. Overall, arthropods were a significantly greater part of the fauna in Hunting and Swanson Creeks than in Trent Hall Creek (Figure 10). The isopod *Edotea* was three times more abundant in Hunting and Swanson Creeks than in Trent Hall Creek and the cumacean *Leucon* was significantly more abundant in Swanson Creek (Figure 7).

These differences in faunal distributions also resulted in differences among the sites in trophic group composition. Because of the dominance of *Heteromastus* in Trent Hall Creek, deep sub-surface deposit feeders formed a significantly greater part of the fauna at this site (Figure 11). Omnivorous species formed the most abundant group at all three sites (Figure 11). However, in Trent Hall Creek this resulted mostly from the high densities of the sub-surface feeding *Nereis* while at the other sites omnivorous surface-feeding arthropods were more important. Surface deposit feeders were significantly more abundant in Trent Hall Creek, largely as a consequence of higher densities of *Macoma*, while the high abundances of *Rangia* in Hunting Creek made suspension feeders a significantly greater faunal component at this site. Site differences were also reflected in the community analyses. Although there were clear overlaps among the sites, the cluster analyses suggest that samples from the same site were more similar to each other than to samples from other sites. Using all of the taxa (Figure 13) or just dominants (Figure 14) made little difference.

Even though there were clear faunal differences among the three sites, it is not possible to determine the cause or causes of these differences. First, the sediment-dwelling shallow water and intertidal invertebrate fauna exhibits significant spatial (e.g. McErlean 1964) and temporal (Holland 1985, Marsh and Tenore 1990) variability. In addition, the sampling occurred at the end of the summer recruitment season and at a time when species such as *Nereis* and *Macoma* may be recruiting in high numbers (Holland et al. 1977b). Natural population variability coupled with the normally high spatial variability in recruitment could have contributed to the observed faunal differences among the sites.

Secondly, the large salinity difference observed between Trent Hall Creek and the other two sites may be indicative of natural environmental differences among the sites. Gradients in salinity have been shown to cause large differences in fauna along a river (e.g. Sanders et al. 1965). In addition, the significantly higher abundances of higher salinity species such as *Nereis*, *Heteromastus*, and *Macoma* (Figures 5, 6) in Trent Hall Creek and the significantly higher densities of lower salinity species such as *Rangia* (Figure 6) in Hunting Creek are consistent with a salinity effect.

Thirdly, it is possible that the differences in faunal distributions resulted from effects of the released oil. However, the evidence for this is quite mixed. The absence of *Leptocheirus* at the sites in both Swanson and Trent Hall Creeks is suggestive of oil effects. *Leptocheirus* is a species widely used in toxicity studies (e.g. McGee et al. 1998, Fuchsman et al. 1998, Norton et al. 1999, Horne et al. 1999) and might be expected to be a good indicator of oil effects. However, even though field populations seem to be more sensitive to toxic compounds than those reared in the laboratory (McGee et al. 1998), *Leptocheirus* has been found to be insensitive to some pollutants in field sampling studies (e.g. Horne et al. 1999). Other taxa used in toxicity testing such as *Hydrobia* (e.g. Forbes et al. 1995) and chironomids (e.g. Fuchsman et al. 1998) had distributions that did not correlate well with the known oil distribution. *Hydrobia* had significantly higher densities in Swanson Creek (Figure 6) and chironomids had lower densities (but not significant) in Swanson and Trent Hall Creeks (Figure 8). Finally, the distributions of broader taxonomic and trophic groups are also known to be affected by pollutants. Horne et al. (1999) in their study of salt marsh sediments found that highly contaminated areas were dominated by polychaetes and surface deposit feeders. Polychaetes and surface deposit feeders were significantly more abundant in Trent Hall Creek, but not in Swanson Creek (Figures 10, 11). Omnivores, which included many surface feeding taxa were more abundant at the two oiled sites than in Hunting Creek (Figure 11). Thus these results are also mixed in their support of an effect of oil on the distribution of the infauna at the three sites.

Fourthly, it is also possible that the process of cleaning oil from the intertidal sites could have had an effect on the faunal distributions. However, there is no way to distinguish this from the effects of the oil itself (or other environmental parameters) using the data collected.

Fifthly, the distributions of these sediment-dwelling invertebrates could also have been influenced by other toxic substances not associated with the oil spill. For example, Riedel et al. (1998, 2000) have shown that cadmium and other toxic trace elements vary spatially and temporally in the Patuxent River. In particular, cadmium was found in the highest concentration in oysters taken near Chalk Point and steadily declined towards the mouth of the river (Riedel et al. 1998). Given that species such as *Leptocheirus* have been shown to be affected by cadmium (e.g. McGee et al. 1998), it is possible that the decreases in many species from Hunting and Swanson Creeks to Trent Hall Creek could result from effects of contaminants, such as cadmium, not associated with the oil spill.

Finally, the differences in distributions could result from a combination of any or all of the factors described above. For example, if a significant acute mortality of infauna resulted from the oil spill or the process of cleaning intertidal areas, then the higher densities at Trent Hall could have resulted from recruitment in the 5 months after the spill. The absence of competitors and predators at this site could have enabled high recruitment of opportunistic species such as *Heteromastus* and *Nereis*. The lower numbers of these species at Swanson Creek could be attributed to the interacting effect of salinity on the distribution of these species.

Thus, it is impossible to know the causes of broad spatial differences in fauna from a set of one time samples taken at only three sites. This study has shown that significant, and sometimes large, differences do exist among the three sites, but it cannot make any inferences regarding the cause or causes of these differences. The design of the study was inadequate to determine such differences. Given the large spatial and temporal variability in environmental parameters (e.g. salinity) and biota inherent in estuarine habitats, a single, one-time set of samples cannot differentiate between causes.

The best and most direct approach to identifying and quantifying the effects of the oil spill on the biota would have been to have taken samples prior to the spill and compared these to those taken over time intervals after the spill. Given that the oil spread down-river over a period of many days, this could have been done at many sites and it is unfortunate that it was not.

Given the lack of sampling before sites were impacted, the best way to have identified oil damage would have been to sample many sites, both oiled and unoiled immediately and then at time intervals after. Samples taken immediately after the spill in April would have identified those species present. Individuals killed would not have been lost immediately. Comparing these samples to those taken later could have been used to distinguish differences resulting from the mortality of residents from those resulting from later recruitment. Differences or similarities seen in one-time sampling can be ephemeral (e.g. resulting from random recruitment differences that may be changed by subsequent recruitment or mortality) or they can be long-lasting. It is impossible to know the dynamic response of such a system based on a single set of samples.

Finally, the strong environmental gradients in an estuarine system such as the Patuxent River produce similar gradients in fauna. Natural spatial and temporal variability in populations can also result from random variation in recruitment, growth, and survival. These processes make it imperative that multiple sites in each class (e.g. control, moderately-oiled, heavily-oiled) be sampled so that oil effects can be isolated from natural differences. These sites should also be sampled repeatedly over a time range that can account for differences in recruitment as well as short- and long-term variations in mortality (including population structure changes, growth and reproductive output). By sampling only one site in each category it is not possible to determine whether a site is different because of the categorical difference (i.e. oil exposure) or just natural variability.

In summary, although this study can identify differences among the sites, it is inadequate to determine what caused those differences. It is also inadequate for evaluating whether any long-term damage to the intertidal and shallow subtidal fauna resulting from the oil spill has occurred.

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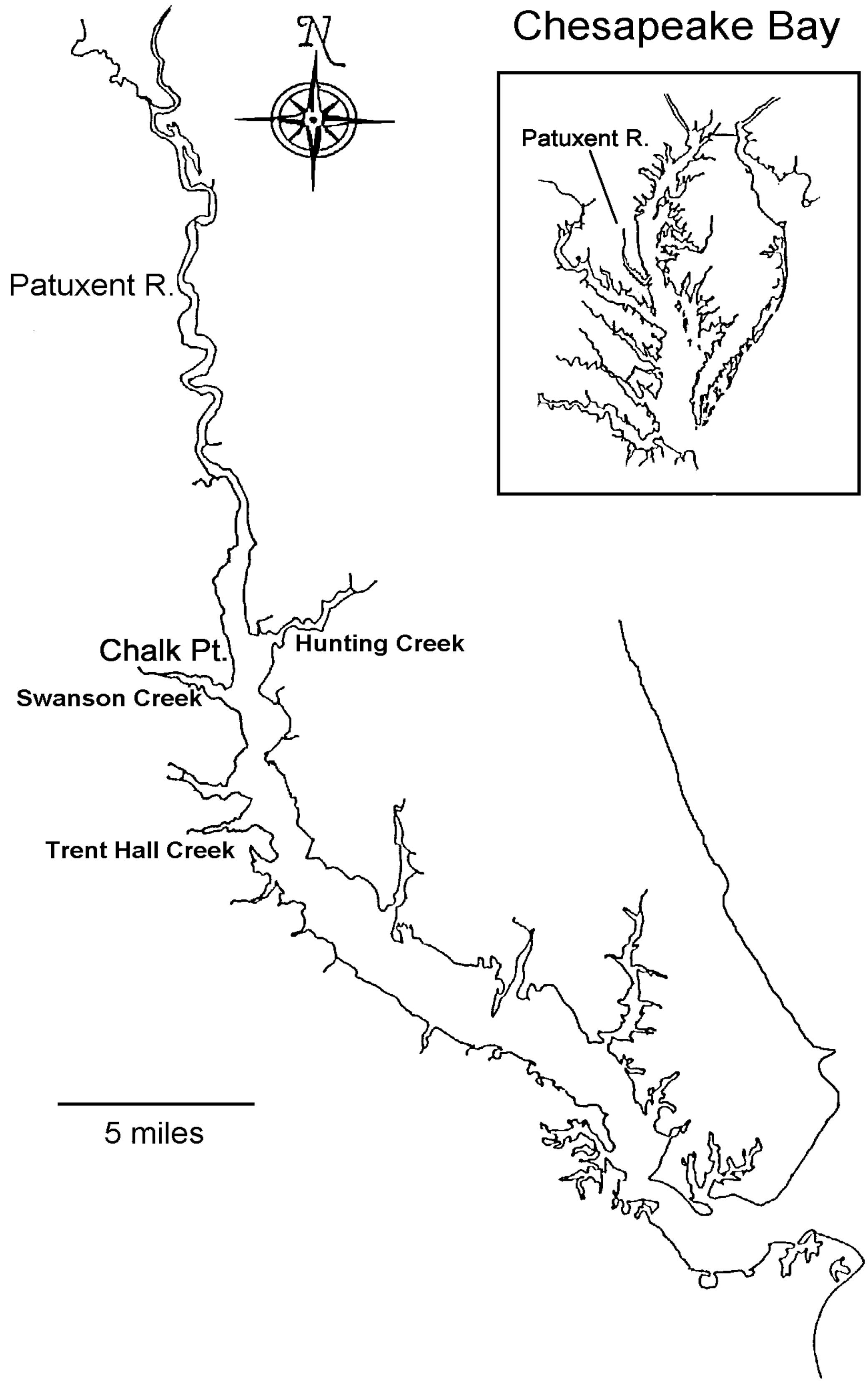

Figure 1. Map of Patuxent River showing the three sampling locations near the Chalk Point oil spill site.

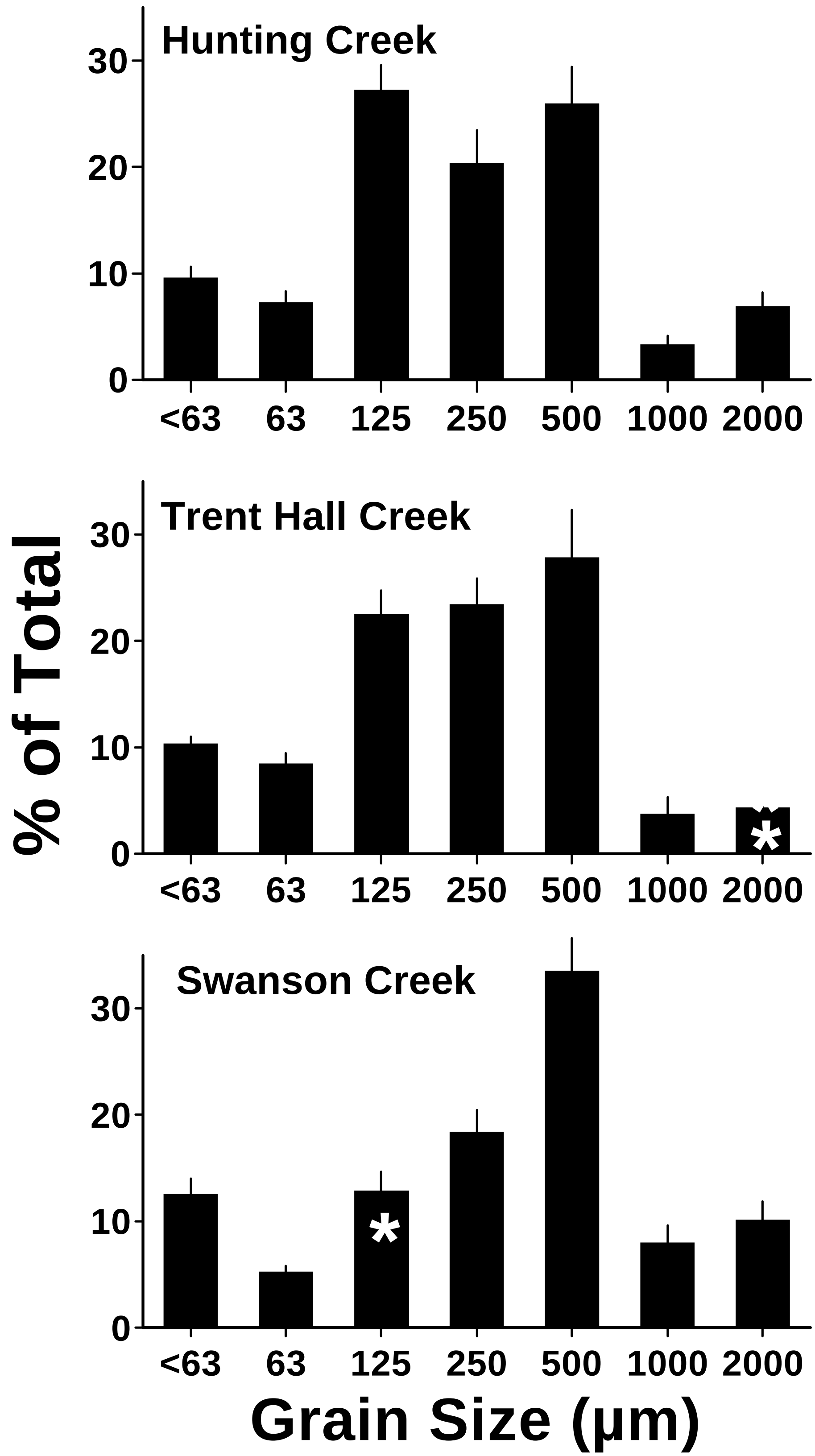

Figure 2. Comparison of grain size distributions at the three sampling sites. Means (±SE) are shown for each size class. * Indicates a significant difference from the other sites for the size class.

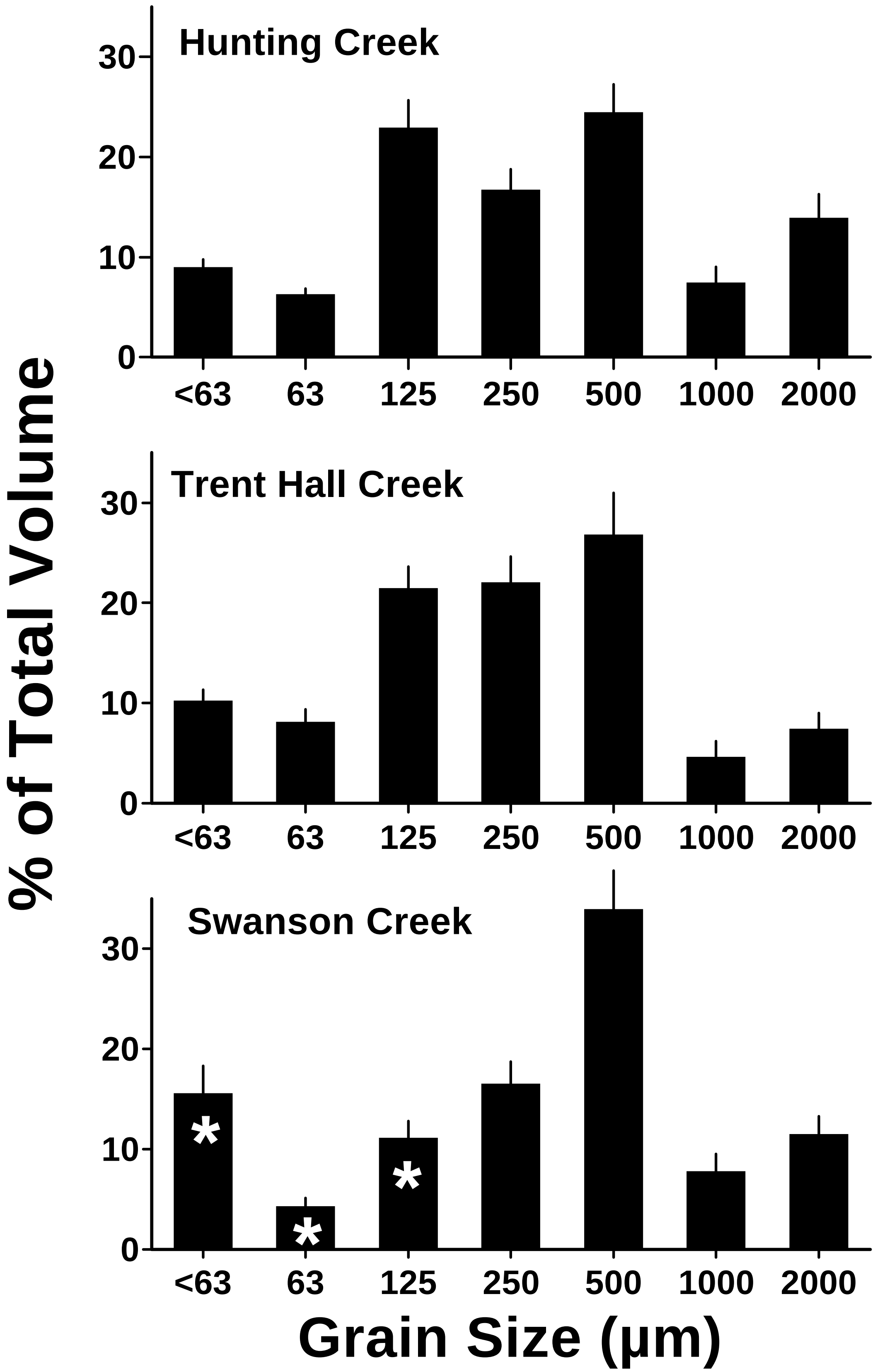

Figure 3. Comparison of grain size distributions at the three sampling sites based on volume. Means (±SE) are shown for each size class. * Indicates a significant difference from the other sites for the size class.

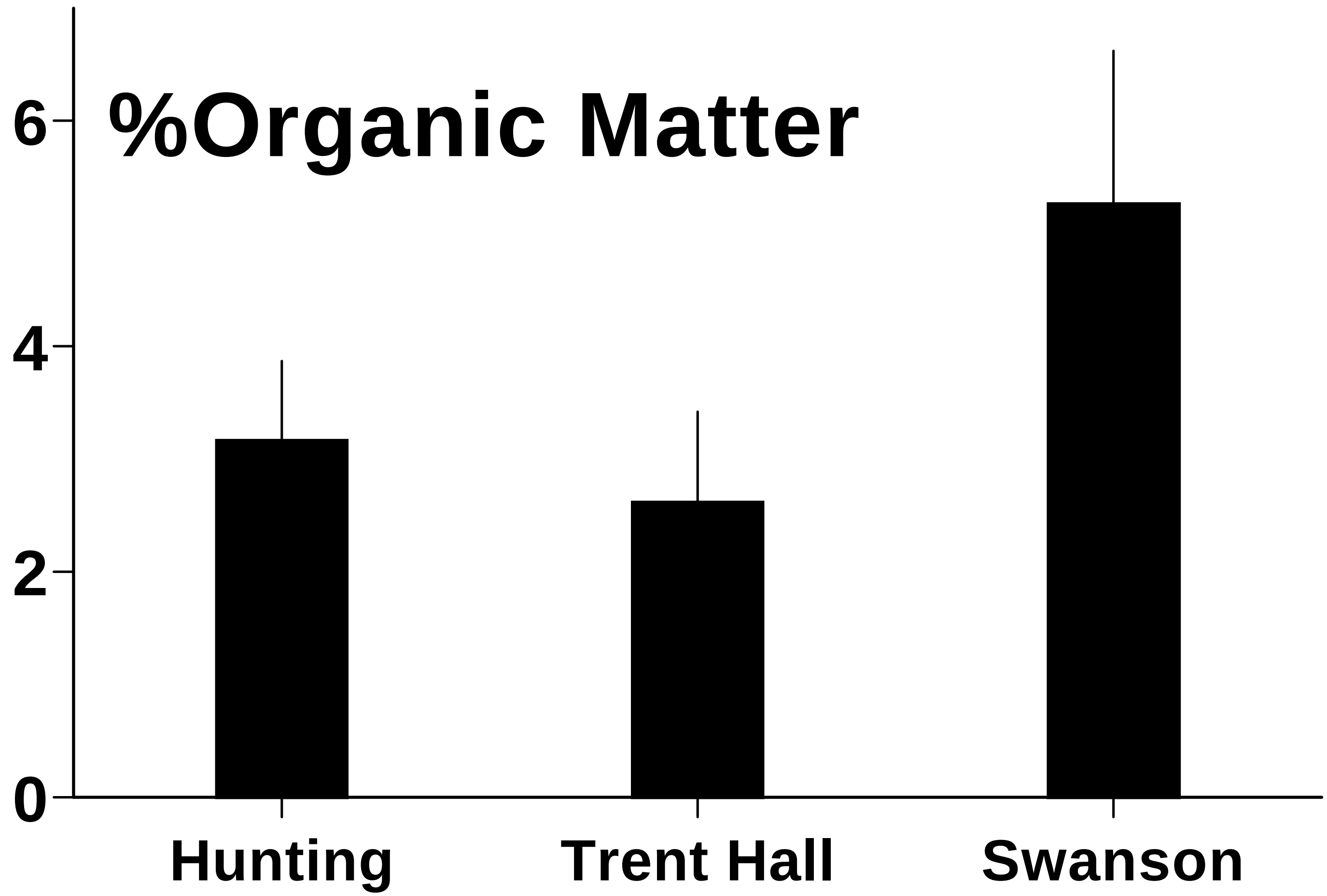

Figure 4. Comparison of the % Organic Matter in sediment samples from the three sampling sites. Means (±SE) are shown for each site.

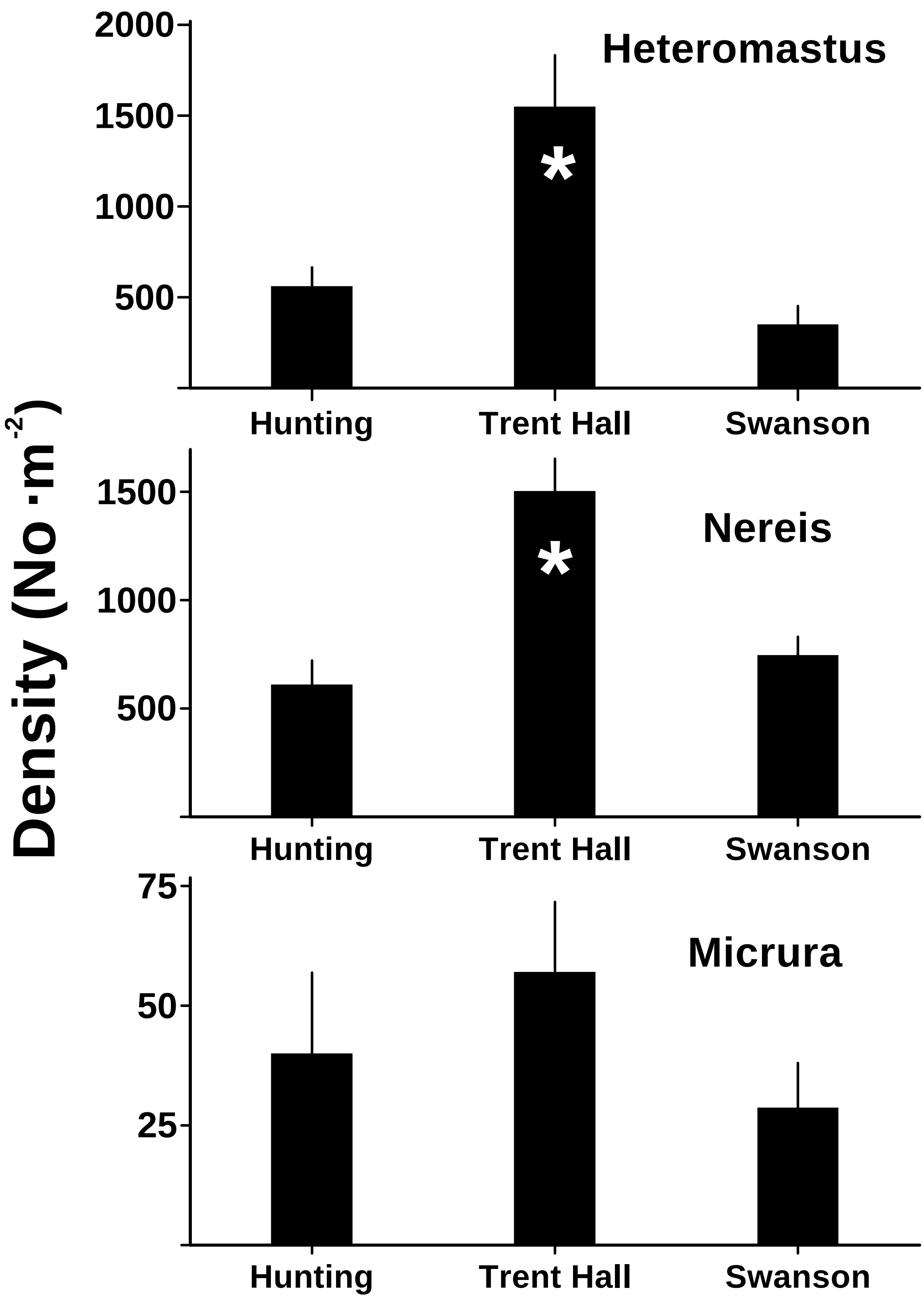

Figure 5. Comparison of the densities of *Heteromatus filiformis, Nereis succinea*, and *Micrura leidyi* among the three sampling sites. Means (±SE) are shown for each site. * Indicates a significant difference from the other sites.

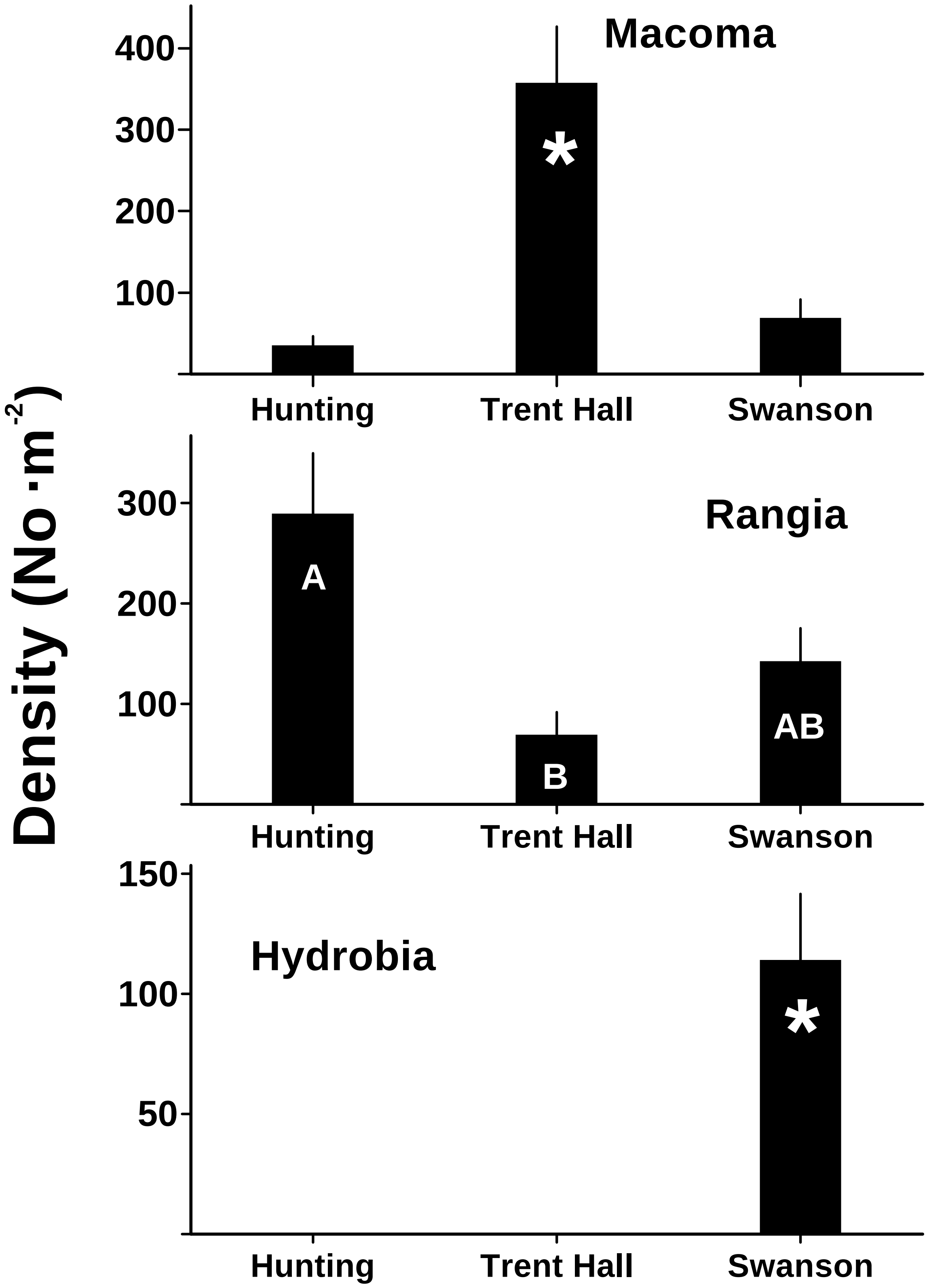

Figure 6. Comparison of the densities of *Macoma balthica*, *Rangia cuneata*, and *Hydrobia* sp. among the three sampling sites. Means (±SE) are shown for each site. * Indicates a significant difference from the other sites. Letters indicate sites with no significant difference.

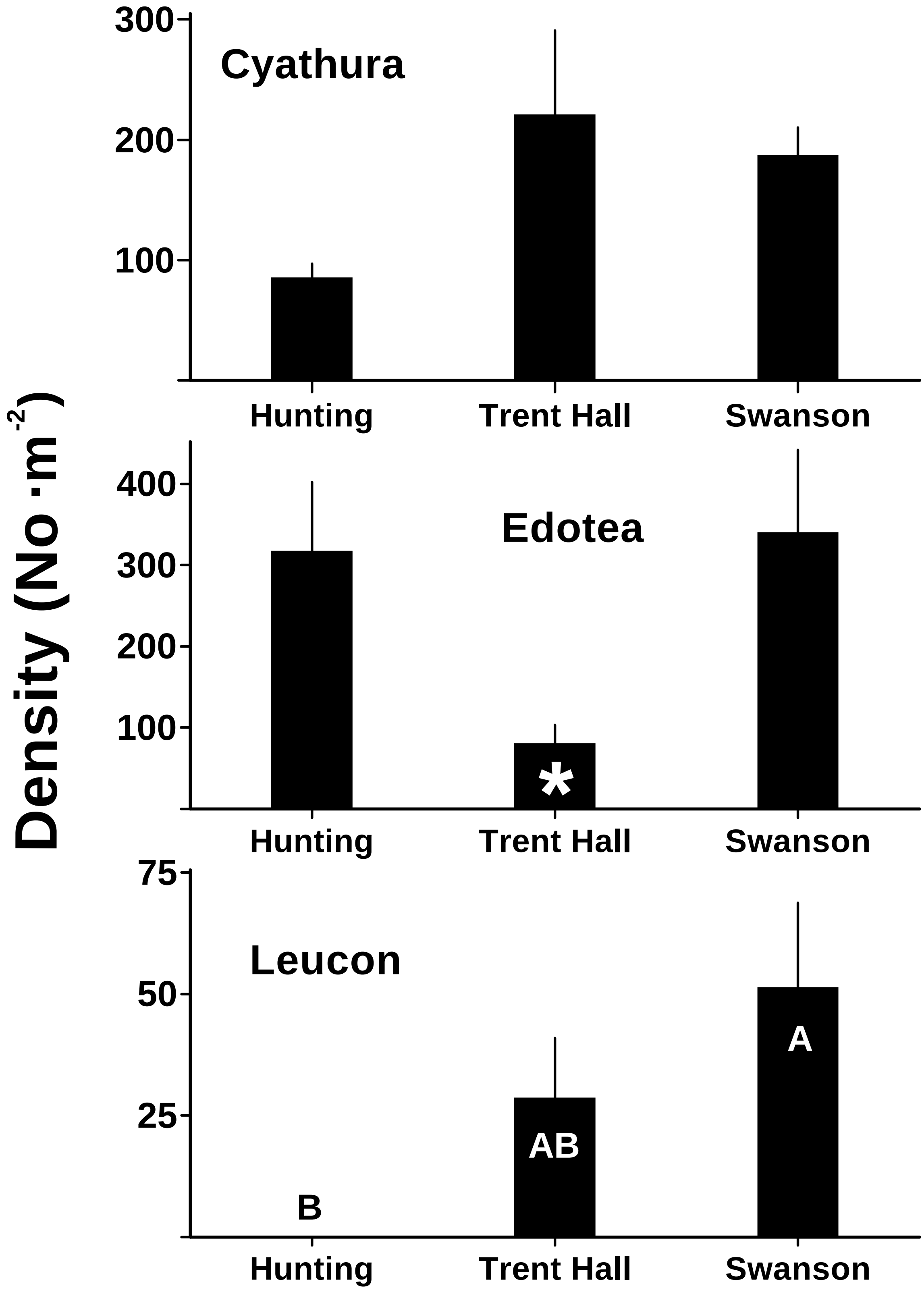

Figure 7. Comparison of the densities of *Cyathura polita*, *Edotea triloba*, and *Leucon americanus* among the three sampling sites. Means (±SE) are shown for each site. * Indicates a significant difference from the other sites. Letters indicate sites with no significant difference.

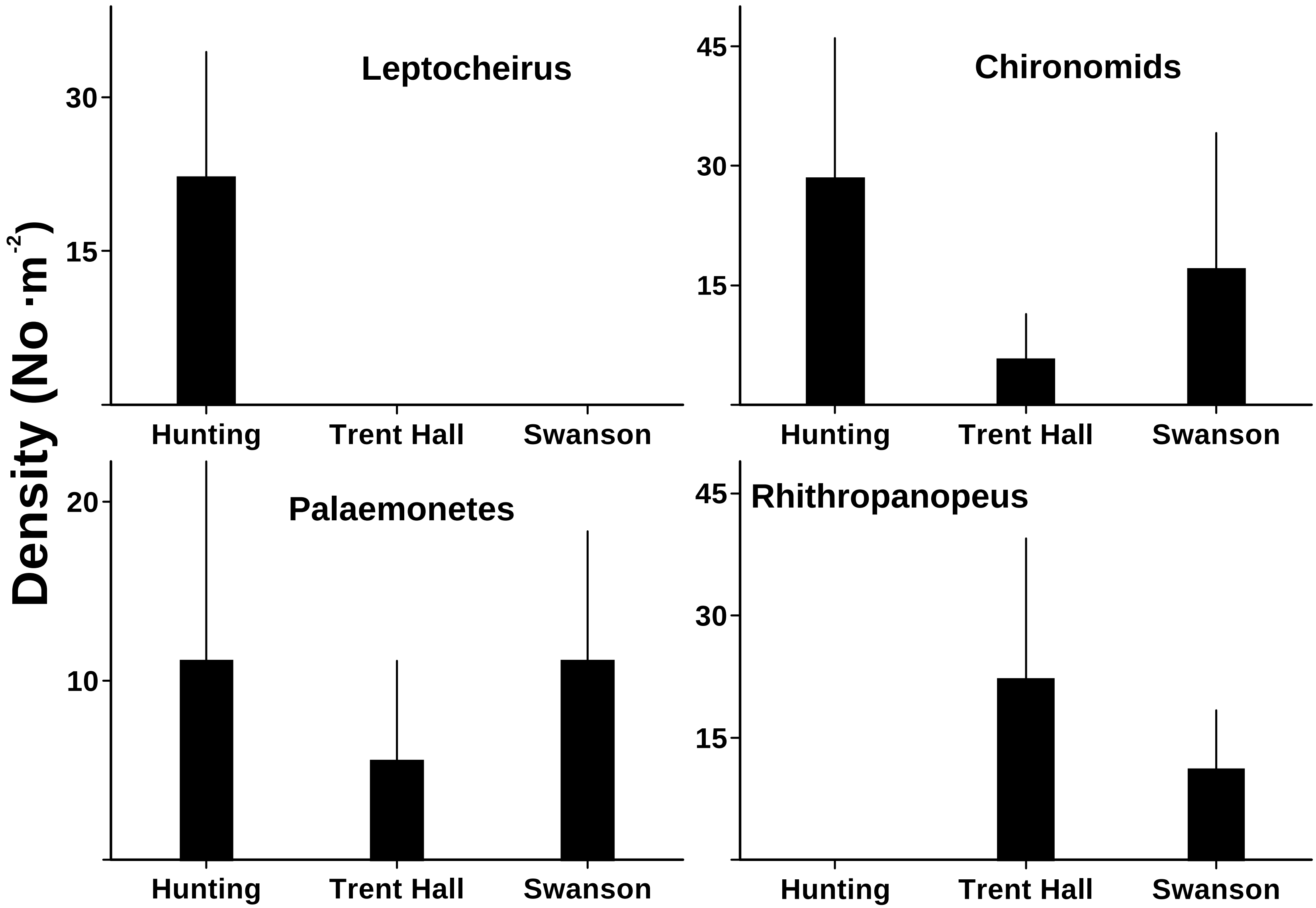

Figure 8. Comparison of the densities of *Leptocheirus plumulosus*, Chironomids, *Palaemonetes pugio*, and *Rhithropanopeus harrisii* among the three sampling sites. Means (±SE) are shown for each site.

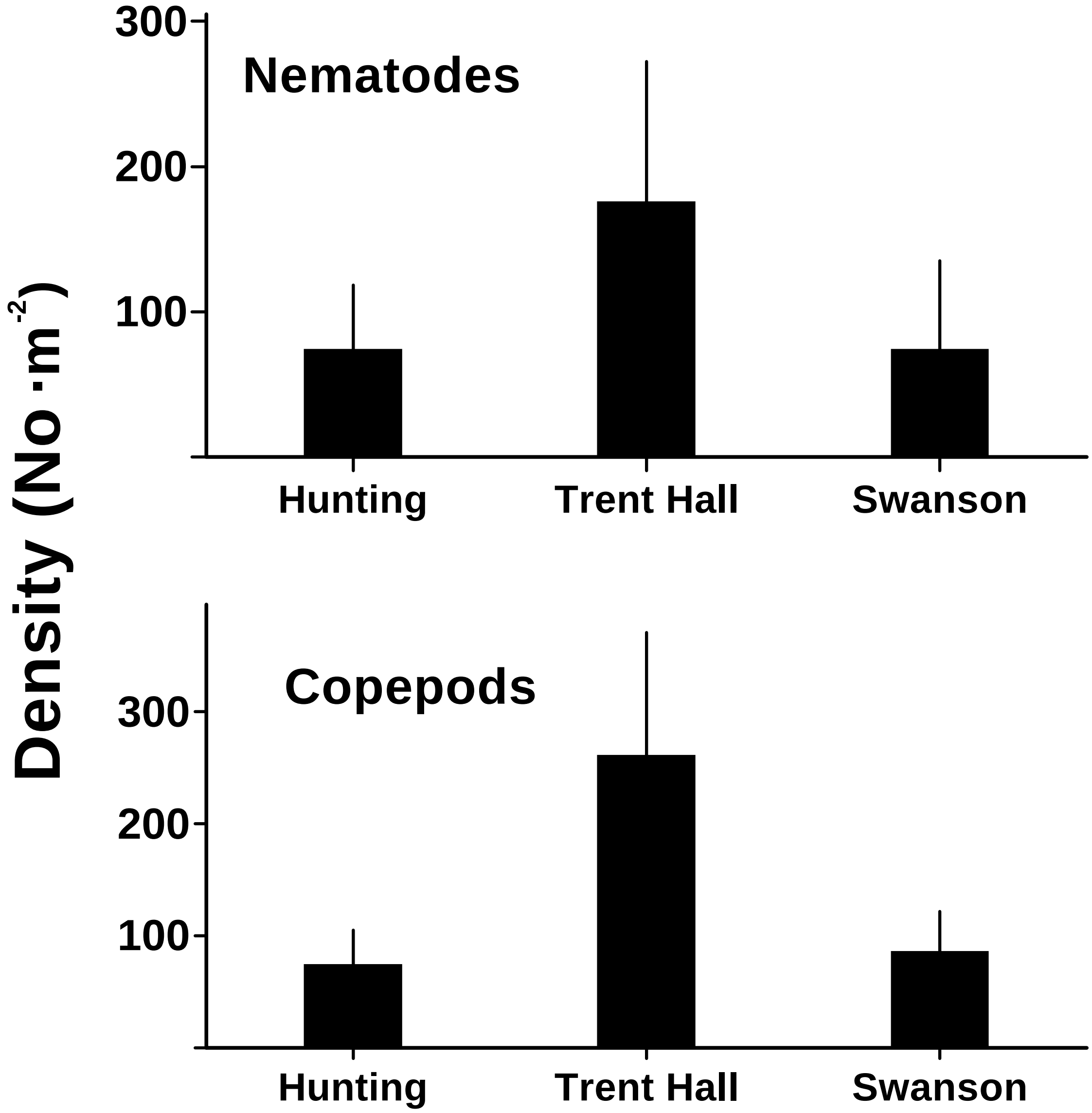

Figure 9. Comparison of the densities of Nematodes and Copepods among the three sampling sites. Means (±SE) are shown for each site.

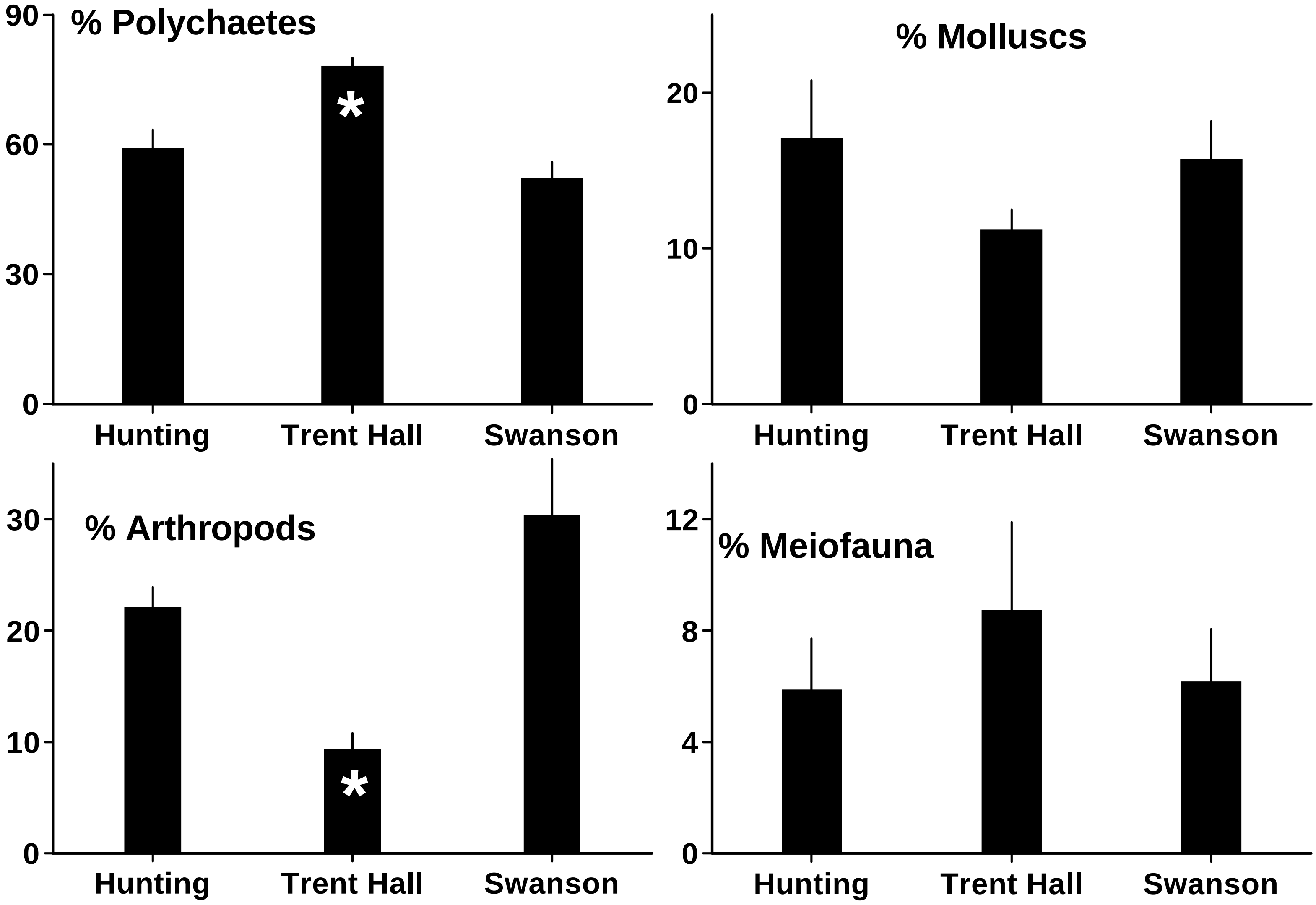

Figure 10. Comparison of the relative abundances of Polychaetes, Molluscs, Arthropods, and Meiofauna among the three sampling sites. Means (±SE) are shown for each site. * Indicates a significant difference between the site and others.

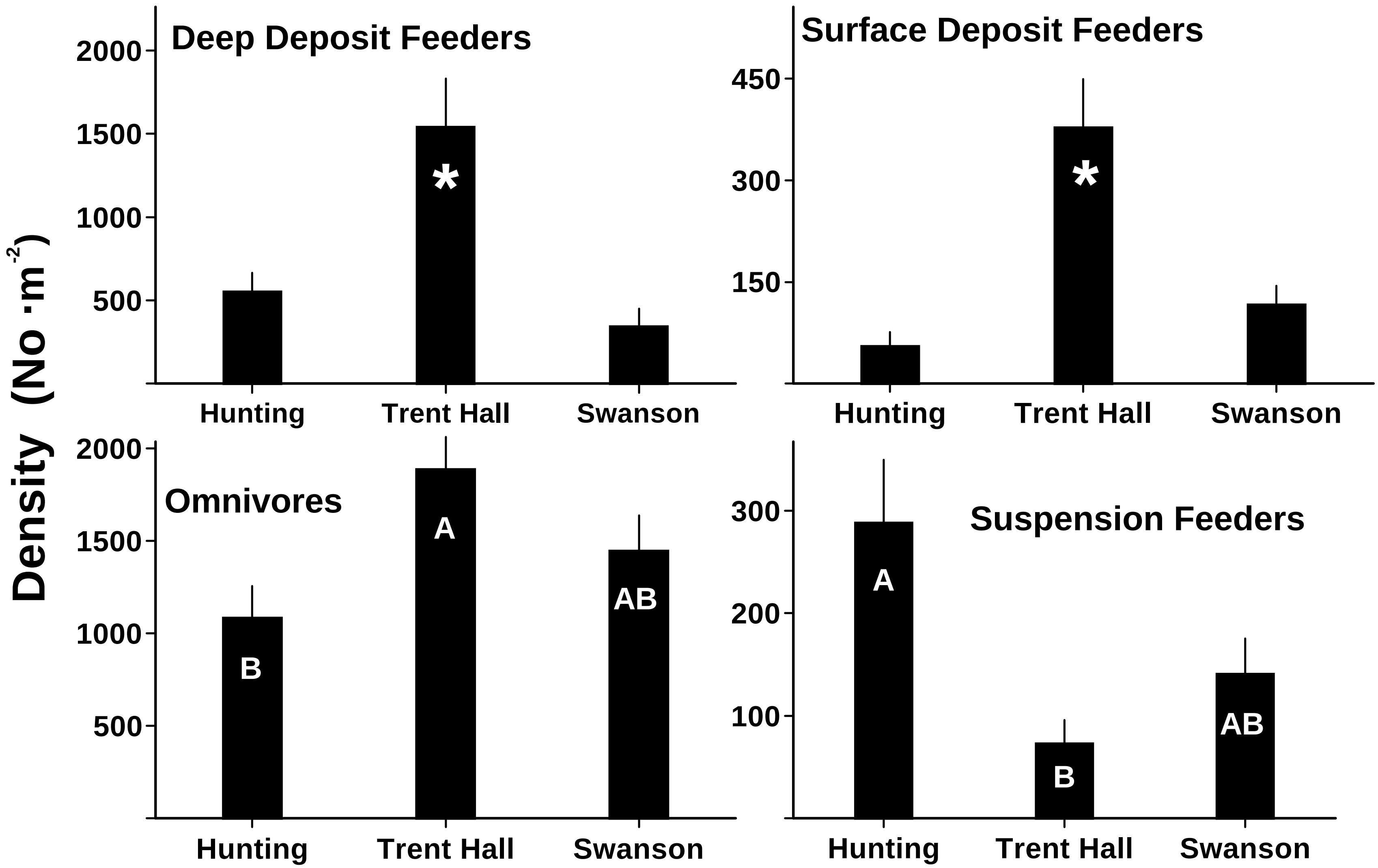

Figure 11. Comparison of the densities of four trophic groups among the three sampling sites. Means (±SE) are shown for each site. * Indicates a significant difference between the site and others. Letters indicate sites with no significant differences.

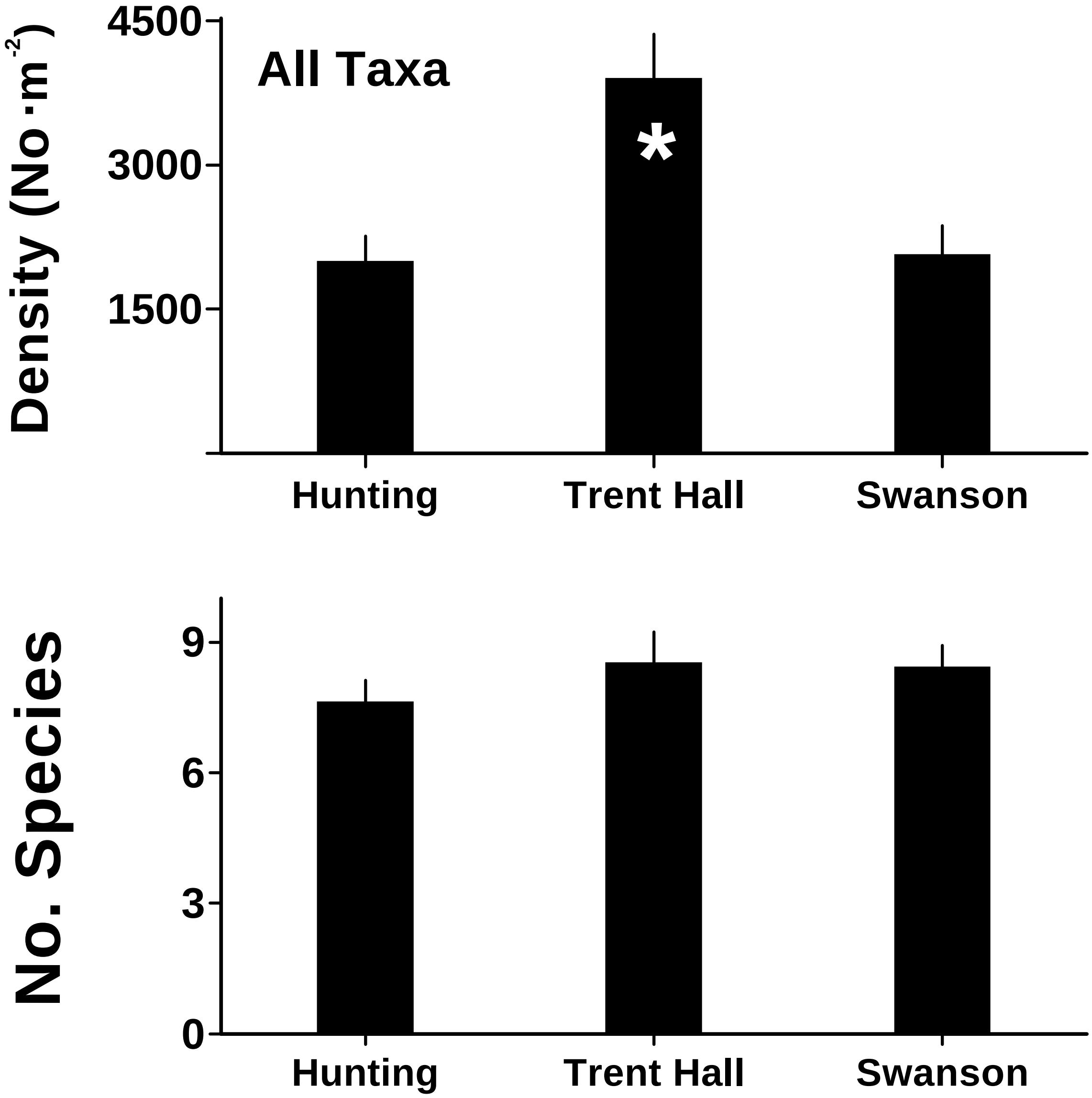

Figure 12. Comparison of the densities of All Taxa and the Number of Species among the three sampling sites. Means (±SE) are shown for each site.

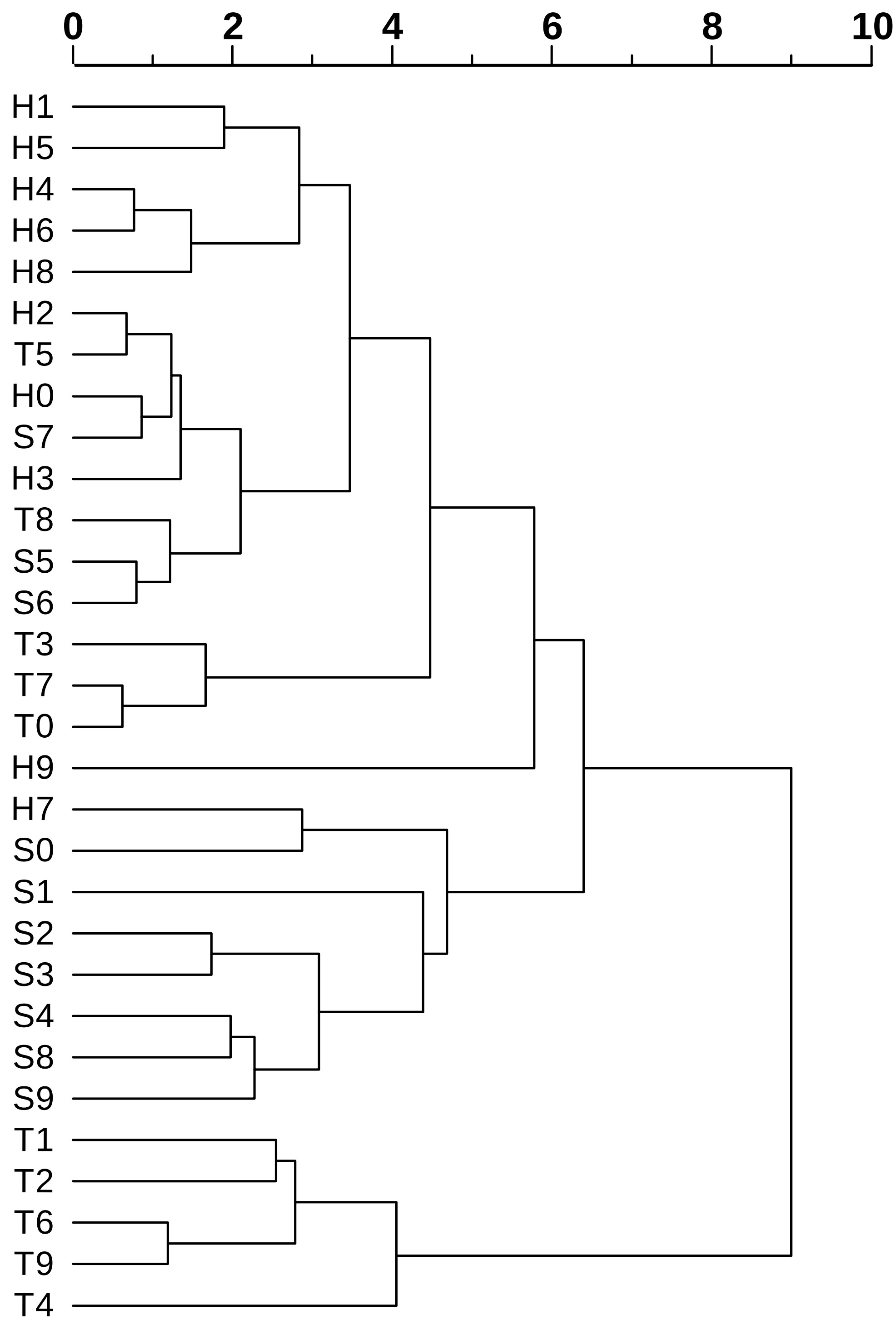

Figure 13. Dendrogram showing similarity among sites based on the densities of all taxa. Higher links indicate greater dissimilarity. H = Hunting Creek, S = Swanson Creek, T = Trent Hall Creek.

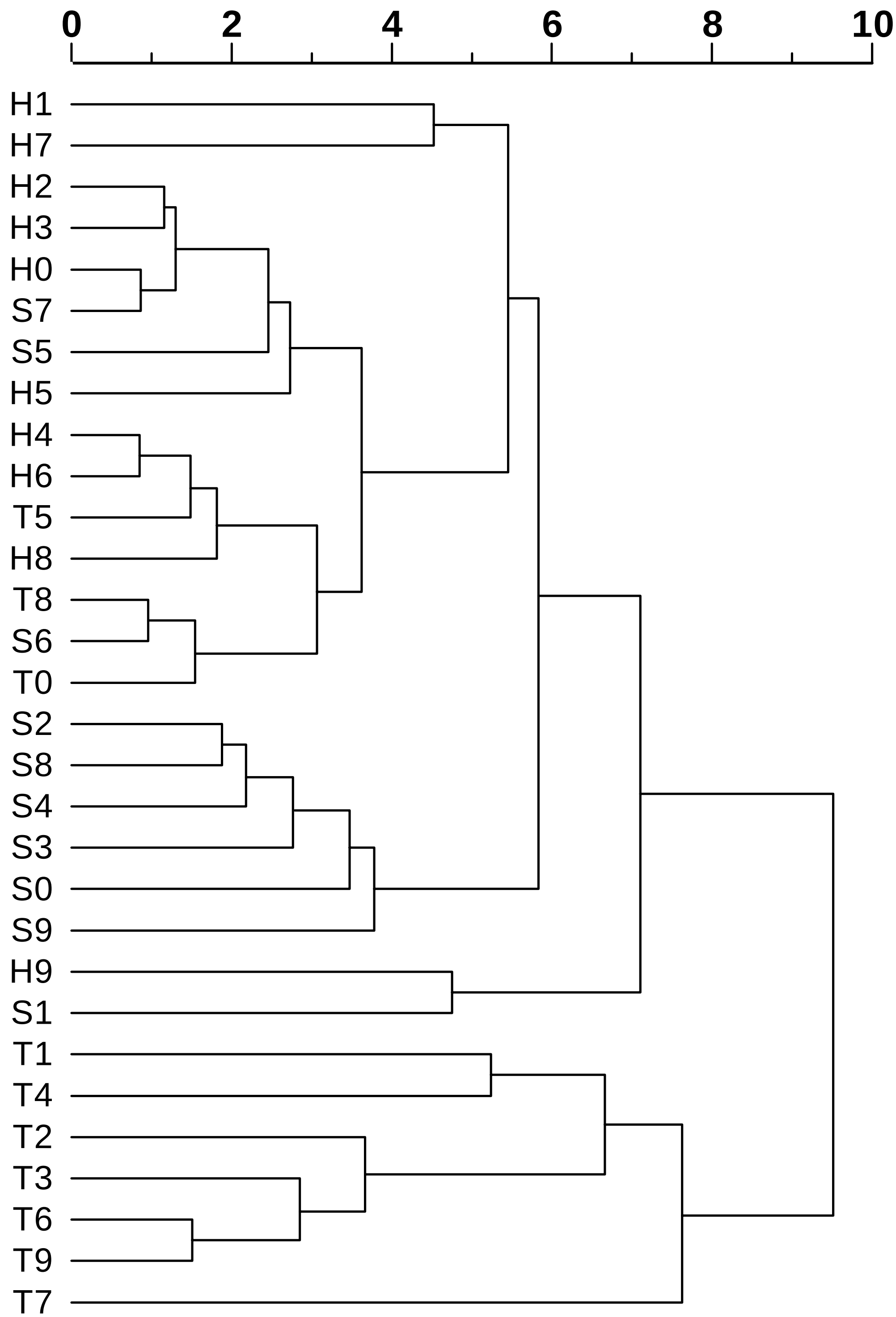

Figure 14. Dendrogram showing similarity among sites based on the densities of the dominant taxa. Higher links indicate greater dissimilarity H = Hunting Creek, S = Swanson Creek, T = Trent Hall Creek.

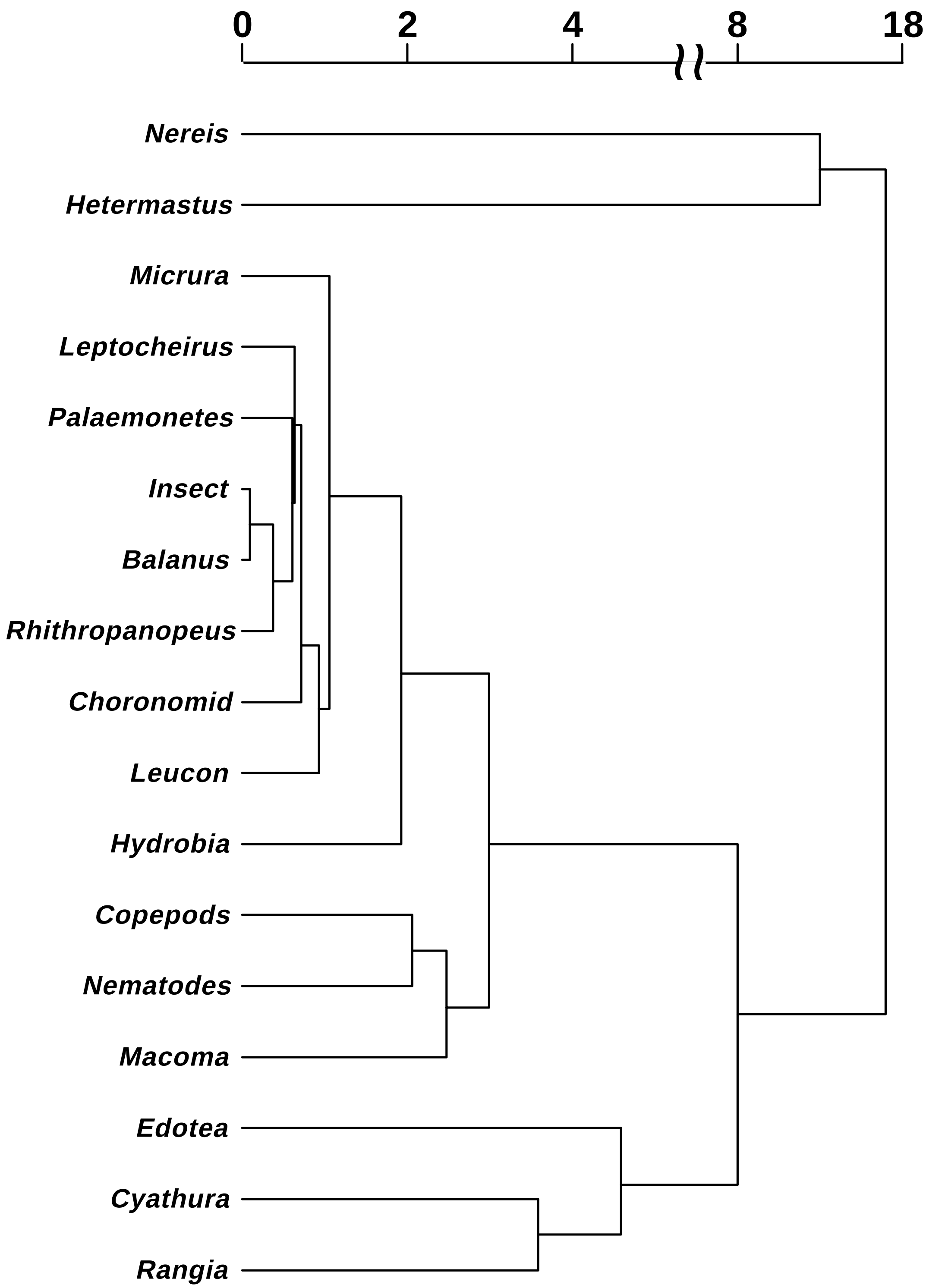

Figure 15. Dendrogram showing similarity among all species based on their distribution among sites. Higher links indicate greater dissimilarity.

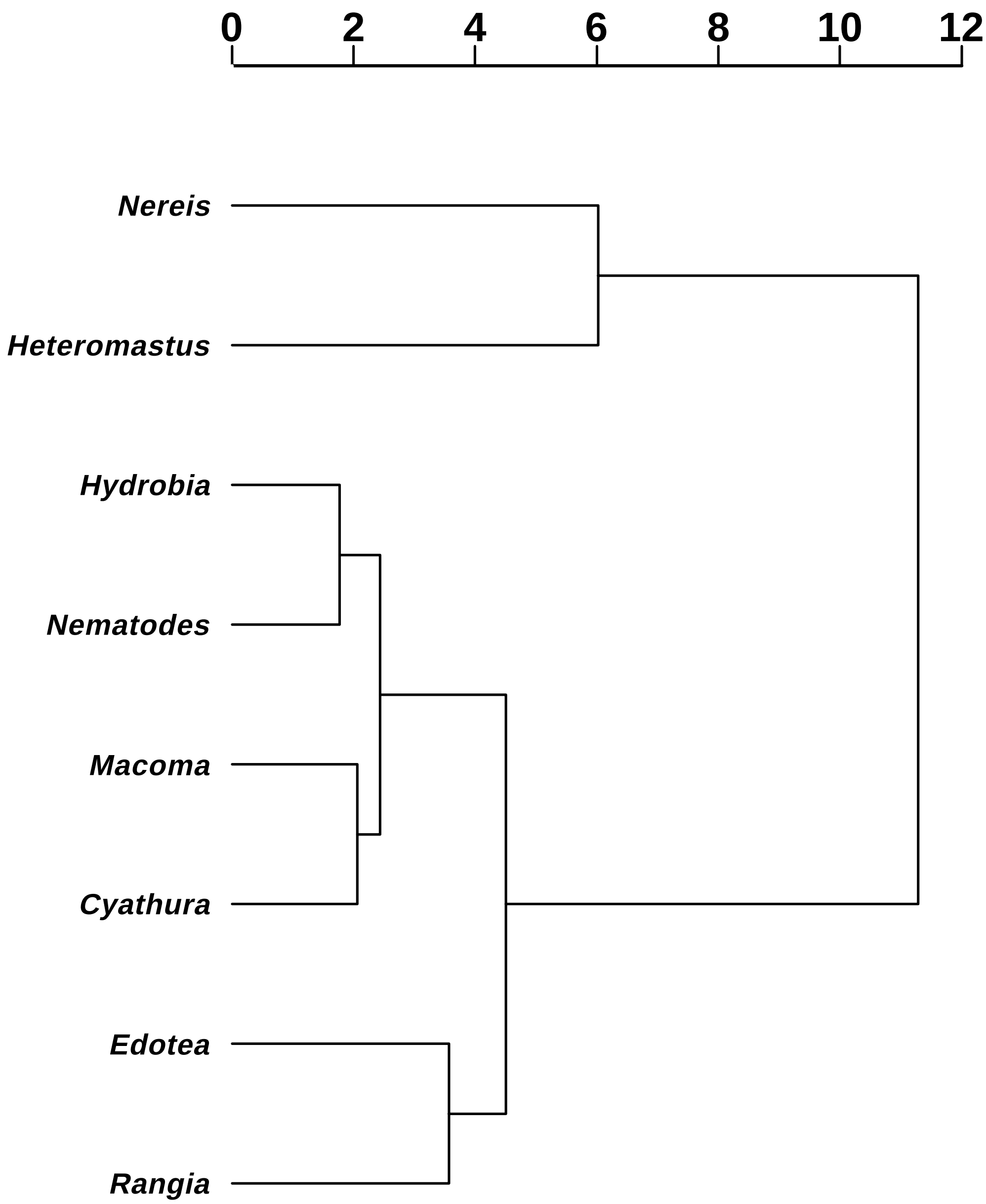

Figure 16. Dendrogram showing similarity among dominant species based on their distribution among sites. Higher links indicate greater dissimilarity.

APPENDIX I

FAUNAL AND SEDIMENT SAMPLING DATA FOR EACH STATION

	Hunting Creek										
Sample	**1**	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**	**10**	**Mean**
Water Depth (cm)	34	31	34	42	37	39	36	34	33	35	35.5
Time of collection	11:00	11:00	11:00	11:00	11:00	11:00	11:00	11:00	11:00	11:00	11:00
Salinity ppt	5.3	5.3	5.3	5.3	5.3	5.3	5.3	5.3	5.3	5.3	5.3
Temperature C	21.3	21.3	21.3	21.3	21.3	21.3	21.3	21.3	21.3	21.3	21.3
Nemerteans											
Micrura leidyi	1	0	0	1	0	1	3	1	0	0	0.7
Annelids											
Nereis succinea	11	6	4	10	3	12	10	15	26	10	10.7
Heteromastus filiformis	21	13	12	13	3	3	13	6	13	1	9.8
Bivalve Molluscs											
Rangia cuneata	4	2	0	6	6	6	11	9	6	1	5.1
Macoma balthica	0	1	0	0	2	0	1	0	1	1	0.6
Gastropods											
Hydrobia sp.	0	0	0	0	0	0	0	0	0	0	0.0
Cumaceans											
Leucon americanus	0	0	0	0	0	0	0	0	0	0	0.0
Barnacles											
Balanus improvisus	0	0	0	0	0	0	0	0	0	0	0.0
Amphipods											
Leptocheirus plumulosus	1	0	0	0	1	0	0	0	2	0	0.4
Isopods											
Edotea triloba	10	6	3	3	1	6	17	5	4	1	5.6
Cyathura polita	3	1	1	2	2	3	0	1	1	1	1.5
Decapods											
Palaemonetes pugio	2	0	0	0	0	0	0	0	0	0	0.2
Rhithropanopeus harrisii	0	0	0	0	0	0	0	0	0	0	0.0
Insects											
Chironomids	0	0	1	0	0	0	0	1	3	0	0.5
Adult Insects	0	0	0	0	0	0	0	0	0	0	0.0
Meiofauna											
Copepoda	3	0	0	0	1	1	5	0	3	0	1.3
Nematoda	0	1	0	3	0	1	0	0	8	0	1.3
% Organic Matter	3.2	1.9	1.6	2.3	4.8	3.2	1.7	2.5	1.6	8.8	3.2

Table A1. Abundances of macrofauna in ten core samples from the control site in Hunting Creek. Numbers in each core and the station means are shown. Incidental meiofauna numbers and the % organic matter are also shown.

	Trent Hall Creek										
	1	2	3	4	5	6	7	8	9	10	Mean
Water Depth (cm)	26	26	22	24	25	25	28	29	24	26	25.5
Time of collection	12:00	12:00	12:00	12:00	12:00	12:00	12:00	12:00	12:00	12:00	12:00
Salinity ppt	9.6	9.6	9.6	9.6	9.6	9.6	9.6	9.6	9.6	9.6	9.6
Temperature C	23.8	23.8	23.8	23.8	23.8	23.8	23.8	23.8	23.8	23.8	23.8
Nemerteans											
Micrura leidyi	1	2	0	2	2	0	0	1	1	1	1.0
Annelids											
Nereis succinea	36	23	23	25	10	31	34	17	37	29	26.5
Heteromastus filiformis	52	42	21	50	9	34	14	12	24	15	27.3
Bivalve Molluscs											
Rangia cuneata	1	0	0	4	3	1	0	1	1	1	1.2
Macoma balthica	3	6	8	13	0	10	3	5	10	5	6.3
Gastropods											
Hydrobia sp.	0	0	0	0	0	0	0	0	0	0	0.0
Cumaceans											
Leucon americanus	1	2	1	0	0	1	0	0	0	0	0.5
Barnacles											
Balanus improvisus	0	0	0	0	0	0	1	0	0	0	0.1
Amphipods											
Leptocheirus plumulosus	0	0	0	0	0	0	0	0	0	0	0.0
Isopods											
Edotea triloba	2	4	0	1	3	1	0	1	2	0	1.4
Cyathura polita	5	8	0	10	1	5	1	4	4	1	3.9
Decapods											
Palaemonetes pugio	0	0	1	0	0	0	0	0	0	0	0.1
Rhithropanopeus harrisii	0	1	0	0	0	0	3	0	0	0	0.4
Insects											
Chironomids	1	0	0	0	0	0	0	0	0	0	0.1
Adult Insects	1	0	0	0	0	0	0	0	0	0	0.1
Meiofauna											
Copepoda	12	3	0	19	1	5	1	1	4	0	4.6
Nematoda	16	1	1	10	1	1	0	0	0	1	3.1
% Organic Matter	1.6	1.9	2.3	2.5	3.5	1.2	9.4	1.4	0.9	1.4	2.6

Table A2. Abundances of macrofauna in ten core samples from the moderately-oiled site in Trent Hall Creek. Numbers in each core and the station means are shown. Incidental meiofauna numbers and the % organic matter are also shown.

	Swanson Creek										
	1	2	3	4	5	6	7	8	9	10	Mean
Water Depth (cm)	20	18	18	19	22	23	18	16	15	17	18.6
Time of collection	13:15	13:15	13:15	13:15	13:15	13:15	13:15	13:15	13:15	13:15	13:15
Salinity ppt	5.6	5.6	5.6	5.6	5.6	5.6	5.6	5.6	5.6	5.6	5.6
Temperature C	25.5	25.5	25.5	25.5	25.5	25.5	25.5	25.5	25.5	25.5	25.5
Nemerteans											
Micrura leidyi	1	0	1	1	0	1	0	0	0	1	0.5
Annelids											
Nereis succinea	21	16	8	15	9	14	4	15	11	18	13.1
Heteromastus filiformis	15	17	1	6	1	5	0	3	4	9	6.1
Bivalve Molluscs											
Rangia cuneata	5	1	1	3	3	3	0	2	1	6	2.5
Macoma balthica	0	0	3	2	2	2	0	0	0	3	1.2
Gastropods											
Hydrobia sp.	3	2	1	2	1	0	0	3	5	3	2.0
Cumaceans											
Leucon americanus	2	1	0	2	0	0	0	0	2	2	0.9
Barnacles											
Balanus improvisus	0	0	0	0	0	0	0	0	0	0	0.0
Amphipods											
Leptocheirus plumulosus	0	0	0	0	0	0	0	0	0	0	0.0
Isopods											
Edotea triloba	9	11	13	3	2	0	3	3	0	16	6.0
Cyathura polita	8	2	4	4	4	3	3	4	1	0	3.3
Decapods											
Palaemonetes pugio	0	0	0	0	1	0	0	0	1	0	0.2
Rhithropanopeus harrisii	0	0	1	0	0	0	0	0	0	1	0.2
Insects											
Chironomids	3	0	0	0	0	0	0	0	0	0	0.3
Adult Insects	0	0	0	0	0	0	0	0	0	0	0.0
Meiofauna											
Copepoda	1	2	0	2	0	0	1	1	1	7	1.5
Nematoda	11	0	0	0	0	0	0	1	0	1	1.3
% Organic Matter	2.9	3.0	6.8	2.5	4.6	2.5	16.4	2.1	6.4	5.5	5.3

Table A3. Abundances of macrofauna in ten core samples from the heavily-oiled site in Swanson Creek. Numbers in each core and the station means are shown. Incidental meiofauna numbers and the % organic matter are also shown.

STATION	Hunting Creek	Trent Hall Creek	Swanson Creek
Dry Weight (mg)			
<62.5 μm	9.0	10.1	11.6
62.5 μm	6.4	8.2	5.0
125 μm	27.3	22.5	12.4
250 μm	21.0	23.0	18.0
500 μm	27.6	28.2	34.3
1000 μm	2.7	3.6	8.4
2000 μm	6.0	4.3	10.3
Sand (>62.5 mm)	91.0	89.9	88.4
Silt-Clay (<62.5 mm)	9.0	10.1	11.6
Volume (ml)			
<62.5 μm	8.8	10.0	14.7
62.5 μm	6.3	7.9	4.2
125 μm	22.7	21.3	10.9
250 μm	17.2	21.7	16.0
500 μm	24.4	27.4	34.4
1000 μm	7.4	4.5	8.1
2000 μm	13.3	7.3	11.6
Sand (>62.5 μm)	91.2	90.0	85.4
Silt-Clay (<62.5 μm)	8.8	10.0	14.6

Table A4. Grain size analysis for sediments collected at the three sites. The mean percentage by size class is shown for each site. The means are based on 10 samples collected at each field site. Percentages by dry weight and by volume are shown.