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Free-living Nematodes from a Natural Oil Seep at La Brea, Trinidad and Tobago

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ABSTRACT

The La Brea oil seep in Trinidad is reportedly one of the largest natural oil seeps in the world. As part of a larger environmental survey of this seepage site, the free-living marine nematodes of the meiofauna were studied. Samples were collected using 60 mm corers at 10 stations at the seep site, between Point Courbarill and Point Rouge west of the Trinidad Pitch Lake. The nematode fauna was represented by 16 families and 32 species. Five families: Chromadoridae, Comesomatidae, Linhomoeidae, Monohysteridae and Ethmolaimidae, comprised approximately 75% of the total abundance. The species diversity (H') was 3.09 with a range of 1.39 to 2.83 between stations. Given the uniqueness of this ecosystem, the average taxonomic distinctness index of biodiversity (Δ^+) was applied to the nematode data and comparisons were made with other locations in the UK and Chile. Taxonomic distinctness (Δ^+) values for La Brea was determined to be 73.28, with the lowest value at station 6 (64.81). Although nematodes are relatively abundant in the sediment samples from La Brea, their extremely low taxonomic distinctness is indicative of a stressed environment.

Key words: free-living marine nematodes, Trinidad and Tobago, natural oil seep.

INTRODUCTION

The La Brea oil seep, located between Point Courbarill and Point Rouge west of the Trinidad Pitch Lake (Fig. 1), had been documented since the early 1800's (Mallet 1802). Seepage at this site still occurs, though it is not as prolific as described in early reports. A steady stream of oil globules can be seen emerging at the surface of the water over an area of approximately 70-100 m². The La Brea seep has one of the highest seepage rates in the world with an estimated volume of 100 barrels per day per 1000 sq. miles metric (Johnson 1970; Wilson *et al.* 1979; Agard *et al.* 1993).

Various studies have examined macrofaunal species distribution at oil seeps and used various indices to assess the effects of long-term exposure to oil on these organisms (Spies and Davis 1979; Davis and Spies 1980; Giammona 1980; Montagna *et al.* 1987; Steichen *et al.* 1996). However, many of these (Shannon-Wiener, log normal distribution models) indices were only based on macrofaunal distributions (Platt and Lambshead 1985) and do not indicate whether these organisms exist in a stressed environment. These univariate methods typically have no defined scale, and they do not consider information on the relative distinctness of taxa (i.e., how closely or distantly species are related to each other). Only a few of these studies have also reported on nematode numbers, but have not identified nematode species.

Clarke and Warwick (1998) showed that the average taxonomic distinctness (Δ^+) has a number of desirable properties as a measure of biodiversity in the context of environmental impact assessment, notably its lack of dependence on sample size and sampling effort. This enables comparisons to be made between studies in which the sampling varies. This is not the case with most of the traditional species diversity measures which are very sample size dependent. Comparisons can also be made on the basis of taxonomic distinctness which has been found to decrease with increasing levels of environmental impacts (Warwick and Clarke 1998).

This paper provides a first report on nematodes from a natural oil seep at La Brea, Trinidad. It also uses taxonomic distinctness measures of nematodes to assess whether a natural oil seep is a stressed environment. Nematodes may serve as a good indicator of environmental quality because of their high abundance, high diversity and their intimate contact with pore water (Lambshead 1986; Bongers 1990; Kennedy and Jacoby 1999; Platt *et al.* 1984; Danovaro 2000).

MATERIALS AND METHODS

Sediment samples were collected at ten stations (Fig.1) using a hand-operated multiple corer which consisted of three 60 mL syringes mounted on a circular platform; a design that allowed for the easy removal and

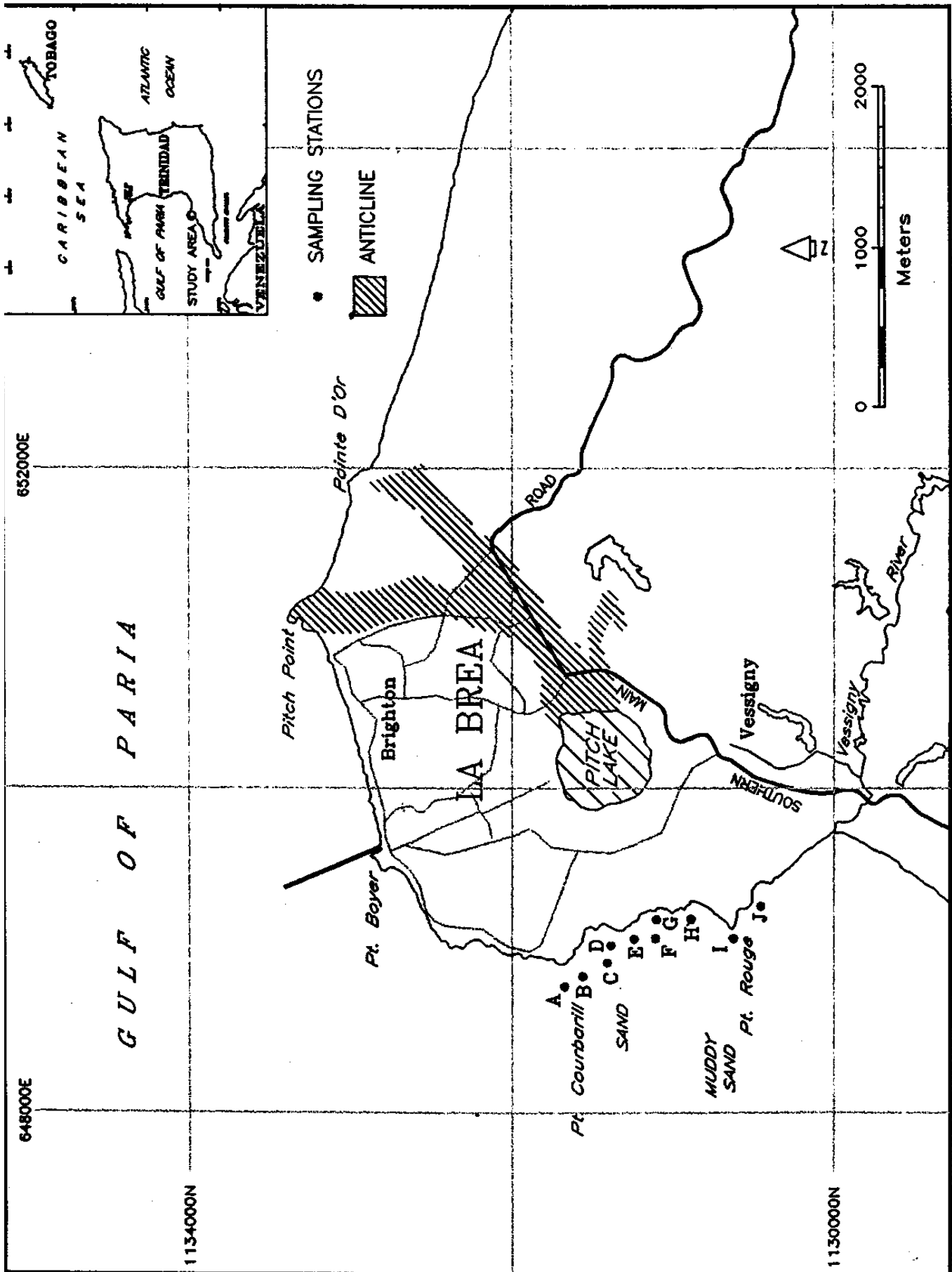


Fig. 1. La Brea Seep, showing the location of the sample stations.

replacement of syringes between sample stations. The tapered end of each syringe was removed, forming a hollow cylinder, the edge of which was sharpened to allow easy penetration into the sediment. Two small holes drilled at the top of each syringe allowed water to be displaced as the cylinders plunged into the sediment.

Sediment cores were placed in plastic bags, preserved in 10% formalin and taken back to the laboratory where they were individually washed through a 63 μm sieve. The meiofauna were separated by flotation using Ludox TM (Platt and Warwick 1983). Each of the samples was then washed and dehydrated for 24 h at 45°C in a 10% glycerol solution and mounted onto glass slides. Nematodes were counted and identified to genus, or species level where possible, using taxonomic keys by Platt and Warwick (1983, 1988).

Data Analysis

Shannon diversity indices (H') and average taxonomic distinctness (Δ^+) (Warwick and Clarke 1995, 1998, 2001) were calculated for nematode assemblages from all the sample stations. Average taxonomic distinctness (Δ^+) is a measure of the degree to which the species in a sample are related taxonomically to each other, and is the average path length between every pair of species traced through a taxonomic tree. From data consisting only of presence or absence of species (i.e. species lists), it is defined mathematically as,

$$\Delta^+ = \left[\sum \sum_{i < j} \omega_{ij} \right] / [s(s-1)/2]$$

where s is the number of species present, the double summation is over the set $\{i=1, \dots, s; j=1, \dots, s, \text{ such that } i < j\}$ and is the 'distinctness ω_{ij} weight' between species i and j . Equal step lengths are normally assumed between each level in the taxonomic hierarchy (species to genus, genus to family, etc.), and Clarke and Warwick (1998) suggest that they be standardised so that the distinctness of two species connected at the highest level (the taxonomically most distant pairing used in a batch of analyses) is set equal to 100. In the present case we have used the six taxonomic levels of species. The taxonomic distinctness determined for the La Brea seep was also compared with the values calculated for other polluted and unpolluted sites in the United Kingdom (Northumberland Coast and Tyne) and in Chile.

RESULTS

One hundred and forty-nine nematodes belonging to 16 families and comprised of 32 species were identified. Many of these appeared to be previously undescribed species (Warwick, pers. comm.). The nematode species

list for the La Brea seep is shown in Table 1. Nematodes belonging to the family Chromadoridae were the most dominant, accounting for about 20% of the total number identified at the seep site, while the most dominant species was *Daptonema* sp.1. Five families - Chromadoridae, Comesomatidae, Linhomoeidae, Monohysteridae and Ethmolaimidae - comprised approximately 75% of the total abundance. Two families - Richtersiidae and Sphaerolaimidae - were previously reported as new records for Trinidad and Tobago (Gobin 2007). Nematode abundances were similar for all stations except for Station 6 where it was highest. Following Wieser's classification, four feeding groups of nematodes were identified. The epigrazers and deposit feeders accounted for 44% and 48% respectively of the total number of nematodes with the remainder (approx. 6%) consisting of predators and omnivores.

The species diversity (H') for all the stations at the La Brea seep was 3.09 with a range of 1.39 to 2.83 for the individual stations. Taxonomic distinctness (Δ^+) values for La Brea was determined to be 73.28, with the lowest value at Station 6 (64.81). When compared to values from different areas in the United Kingdom and Chile (Fig. 2), the value for the La Brea seep site was the lowest. The highest value of taxonomic distinctness for offshore soft sediments was reported in Chile (78.18), a relatively unpolluted site, while the most polluted site in the UK (Liverpool Bay) had a value of 74.67. The taxonomic distinctness value (73.28) for all the sites in La Brea were indeed lower than those reported from Chile and the United Kingdom (Fig. 2).

DISCUSSION

The present study provides a first list of free-living marine nematodes associated with a natural oil seep in Trinidad and Tobago. The nematode densities (149 per 114 cm^2) were higher than that reported for other species such as polychaetes (*Pararionospiso pinnata* 182 m^{-2} , *Nerereis micromma* 182 m^{-2}) and the bivalve *Corbula caribea* (46 m^{-2}), at the La Brea seep. However, it was lower than that reported by Davis and Spies (1980) for Coal Oil Point, Santa Barbara (8.5 per core) and Steichen *et al.* (1996) reported higher nematode densities (229 per 78 cm^2) for the Isla Vista seep. Most such studies have focused on assemblages of organisms or total numbers, with few studies actually identifying the nematode species present.

The La Brea seep is biologically productive despite the high levels of hydrocarbon (26 - 200 $\mu\text{g g}^{-1}$ chrysene equivalents) reported by Agard *et al.* (1988) who described the area as "contaminated". Under such conditions, it is expected organisms in that environment may

Table 1. Nematode (phylum Nematoda) families and species list for the La Brea oil seep coastal area. Family totals are in bold.

Family and Species	No. of Individuals
AXONOLAIMIDAE	
* <i>Paraodontophora</i> sp. 1	7
CERANOMATIDAE	
<i>Pselionema</i> sp. 1	2
CHROMADORIDAE	
<i>Ptycholaimellus</i> sp. 1	5
* <i>Innocuonema</i> sp. 1	12
* <i>Graphonema</i> sp. 1	1
Chromadorid sp. 1	6
Chromadorid sp. 2	1
* <i>Metachromadora</i> sp. 1	3
<i>Hypodontolaimus</i> sp. 1	2
COMESOMATIDAE	
<i>Comesoma</i> sp.1	10
<i>Sabatieria</i> sp. 1	8
* <i>Dorylaimopsis</i> sp. 1	4
* <i>Cervonema</i> sp. 1	1
CYATHOLAIMIDAE	
<i>Paracanthochus</i> sp. 1	1
DESMODORIDAE	
<i>Desmodora</i> sp.1	6
DIPLOPELTIDAE	
* <i>Campylaimus</i> sp. a	1
ENOPLIDAE	
* <i>Mesacanthion</i> sp. a	1

be negatively impacted. However, chronic exposure to hydrocarbons does not always appear to have a toxic effect on organisms as might be expected. Mohammed and Agard (2004) reported that *Corbula caribea* from the seep site had higher levels of NADPH-ferrihaemoprotein reductase when compared to organisms from other sites, which allowed them to tolerate the high levels of hydrocarbons at the seep. Within hydrocarbon-rich environments such as seeps, numerous oil-degrading bacteria have been identified which may utilize the hydrocarbons as a source of energy, and serves as a supplementary food source for meio- and macro-benthic organisms in shallow water (Spies and DesMarais 1983; Bauer *et al.* 1990).

Family and Species	No. of Individuals
ETHMOLAIMIDAE	
* <i>Neotonchus</i> sp. 1	1
* <i>Filitonchus</i> sp. 1	10
* <i>Neotonchus</i> sp. 2	1
LINHOMOEIDAE	
* <i>Terschellingia</i> sp. 1	15
* <i>Terschellingia</i> sp. 2	6
* <i>Terschellingia</i> sp. 3	2
* <i>Terschellingia</i> sp. 4	1
MICROLAIMIDAE	
* <i>Aponema</i> sp. 1	4
ONCHOLAIMIDAE	
<i>Viscosia</i> sp.1	1
OXYSTOMINIDAE	
<i>Halalaimus</i> sp. 1	3
*RICHTERSIIDAE	
* <i>Richtersia</i> sp. 1	9
*SPHAEROLAIMIDAE	
* <i>Sphaerolaimus</i> sp. 1	2
MONOHYSTERIDAE	
<i>Daptonema</i> sp. 1	17
<i>Paramonohystera</i> sp.1	2
<i>Steineria</i> sp. 1	3
Total nematodes	148

* - first records for Trinidad and Tobago

Although the oily layer over sediment may be directly responsible for the reduced abundances and diversity, this stress may be moderated by the nutritive benefits of abundant bacteria. Steichen *et al.* (1996) at Coal Oil Point found that of all the taxonomic groups studied, only nematode abundances were positively correlated with oil contamination. The direct toxic effects of oil or its by-products as well as the deleterious effects of increased sulfide levels generated by microbial activity may be responsible for this (Steichen *et al.* 1996).

Assessment of natural oil seeps have focused primarily on the diversity of species assemblages (Spies and DesMarais 1983; Bauer *et al.* 1990; Davis and Spies

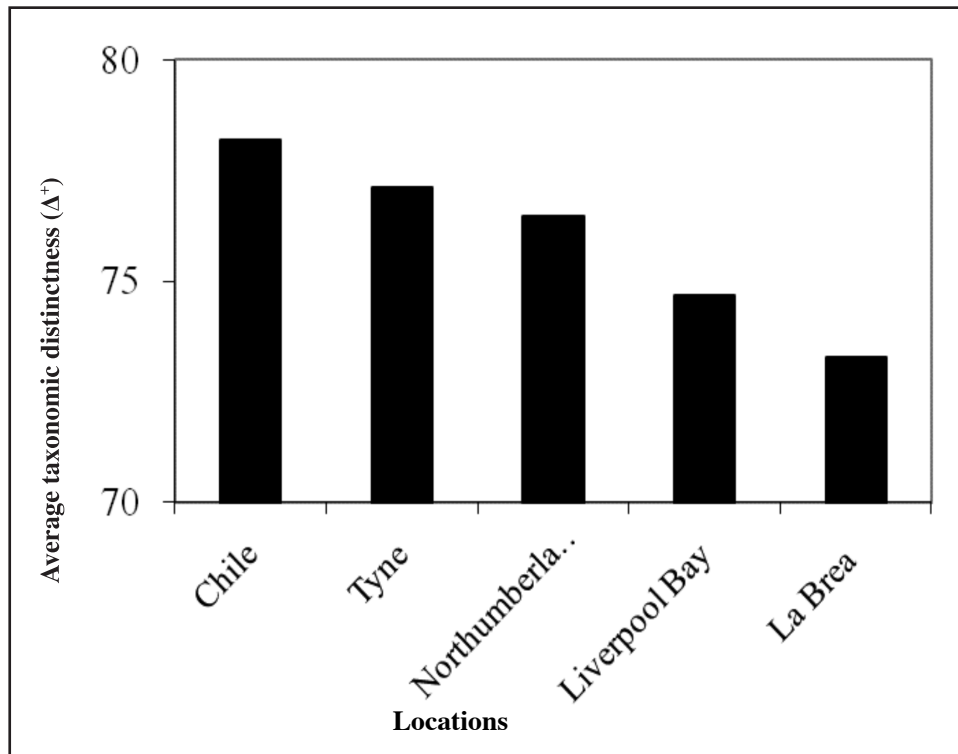


Fig. 2. Taxonomic distinctness (Δ^+) values for temperate (Tyne, Northumberland, Liverpool Bay - UK) and tropical (Chile, La Brea - Trinidad) soft sediment nematode communities.

1980; Montagna *et al.* 1987; Steichen *et al.* 1996) or hydrocarbon levels (Agard *et al.* 1988). However, these assessments give no real indication as to the extent of stress within these environments. Conventional Shannon diversity (H') index for the nematode assemblage from the La Brea seep yielded a value of 3.09, which was higher than the 1.69 - 1.71 reported by Spies and Davis (1979) for all the species recorded at Coal Oil Point, Santa Barbara. Though diversity indices for soft bottom sediments from the Caribbean are not available, nematodes of rocky substrates had a similar diversity index (2.60) (Gobin 1994). However, these values do not give a good indication as to the extent of environmental stress within the seep. These univariate indices show greater diversity when abundance is more evenly distributed among the species sampled, and when the number of species in a sample increases. They have no defined scale, which compromises comparison between studies; they vary naturally between habitats and within habitats over time and they do not consider information on the relative distinctness of taxa (i.e., how closely or distantly species are related to each other). Although nematodes are relatively abundant in the sediment samples from La Brea, their extremely low taxonomic distinctness is indicative of severe environmental stress. These values are the lowest recorded globally for any nematode assemblage from sublittoral habitats so far studied (Fig. 2).

Taxonomic distinctness (Δ^+) measures of the taxonomic spread of species in the assemblage and these observations add further weight to the utility of Δ^+ as a comparative measure of environmental stress. It is difficult to see any other way in which environmental stress at the community level of biological organisation could be measured on such a unified comparative scale. The data used for this comparison are by no means standardised or controlled in terms of the number of location, the core size etc., so that relative diversity measures based on the number of species would be unsuitable as a basis for comparisons.

The La Brea seep has the lowest Δ^+ value when compared to other sublittoral habitats (Fig. 2), which suggest that it is the most stressed of the sites. Northumberland is a relatively pristine mud

and sandy mud area (Warwick and Buchanan 1970). The Tyne, a sewage sludge dumping ground, is a dispersive site where environmental impact on both meiobenthos and macrobenthos is relatively slight (Sommerfield *et al.* 1993), while Liverpool Bay is a heavily industrialised and sewage polluted area (Sommerfield *et al.* 1995). These observations add further weight to the utility of Δ^+ as a comparative measure of environmental stress.

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