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Hydrocarbon Concentrations in the American Oyster, *Crassostrea virginica*, in Laguna de Terminos, Campeche, Mexico

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Laguna de Terminos is a 2,500 km^2 coastal lagoon in the southern Gulf of Mexico, located between 18° 20' and 19° 00' N, and 91° 00' and 92° 20' W (Figure 1). It is a shallow lagoon, with a mean depth of 3.5 m and connected to the Gulf of Mexico through two permanent inlets, Puerto Real to the east and Carmen to the west (Yáñez-Arancibia and Day 1988a). Several rivers, most of them from the Grijalva-Usumacinta basin (the largest in Mexico and second largest in the Gulf of Mexico), drain into the lagoon with a mean annual discharge of 6 x 10^9 m^3/year. This lagoon has been studied systematically, and is probably one of the best known in Mexico. An excellent overview of this lagoon can be found in Yáñez-Arancibia and Day (1988a).

The continental shelf north of Terminos, the Campeche Bank, is the main oil-producing zone in Mexico with a production of about 2 x 10^6 barrels/day. It is also the main shrimp producer in the southern Gulf, with a mean annual catch of 18,000 tonnes/year, which represents 38 to 50 % of the national catch in the Gulf of Mexico (INEGI 1990). The economic importance of this region, along with its extremely high biodiversity, both in terms of species and habitats, has prompted the Mexican government to study the creation of a wildlife refuge around Terminos. Thus, it is very important to know the current levels of pollutants in this area, as a contribution to the management plan of the proposed protected area.

MATERIALS AND METHODS

Oysters (*Crassostrea virginica*) were collected at six sampling stations (Fig. 1) in Laguna de Terminos once a month from February to July, 1991. This period covers the three climatic seasons reported for this area (Day *et al.* 1982; Yáñez-Arancibia and Day 1988b): the cold fronts (February and March), the dry season (April and May) and the rainy season (June and July). Oysters were collected by diving, and transported under refrigeration to CINVESTAV in Merida for further analysis.

Hydrocarbon concentrations in the soft tissues of the oysters were determined following the method adopted by IOCARIBE (1987). The organisms were pooled

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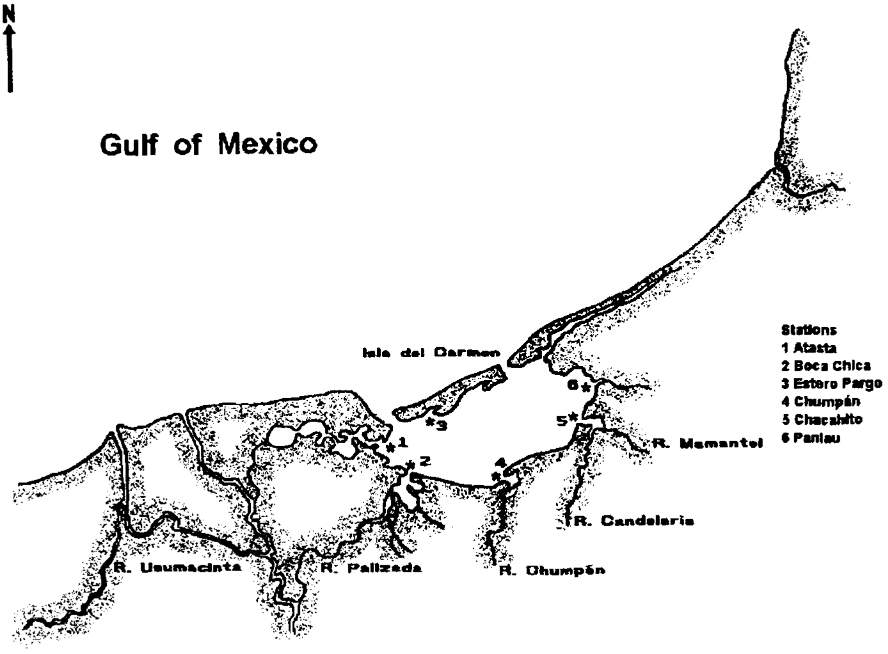

Figure 1. Map of Laguna de Terminos, showing the location of the sampling stations.

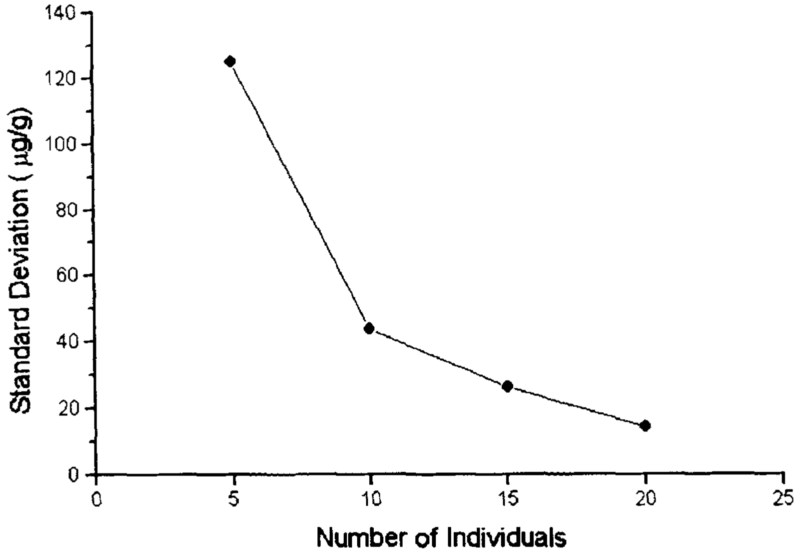

Figure 2. Variability of the total hydrocarbon analysis of oysters *Crassostrea virginica* of pooled samples as a function of the number of individuals.

at each station. The optimum number of individuals in a pooled sample was determined by analyzing three groups of 5, 10, 15 and 20 individuals each and looking for the sample size with minimum standard deviation (Flores and Galindo, 1989; Alvarez-Legorreta *et al.* 1994).

Oysters were digested with 6 M NaOH and the non-saponifiable material extraced with dimethyl-ether. The extract was purified and divided into the aliphatic and aromatic fractions by column chromatography with eight grams each of partially deactivated (with 5 % water) silica gel/alumina. The fractions were analyzed by capillary gas chromatography with a Hewlett-Packard 5890 Series II gas chromatograph equiped with a flame ionization detector, a Hewlett-Packard 200 μm *i.d.* x 25 m, 0.33 μm film Ultra 2 (5 % phenyl-methyl-silicone) column, and HP Chemstation 3365 Series II data acquisition software. Chromatographic conditions were: initial temperature 60°C for one minute, then at 6°C/min to 290°C and held for 20 minutes. The aliphatic fraction was quantified with a n-C_{26} standard and the aromatic fraction with chrysene. Within the aromatic fraction, polynuclear aromatic hydrocarbons were identified by comparing their retention times with those of analytical standards.

RESULTS AND DISCUSSION

The minimum value for the standard deviation, 14 μg/g, corresponds to pooled samples of 20 individuals (Fig. 2). However, most of the peaks from the aliphatic fraction disappeared in the sample with 20 individuals (Fig. 3), probably due to homogenization problems (Flores and Galindo, 1989), and it was decided to use 15 individuals for all the analysis. Using the same method, Alvarez-Legorreta *et al.* (1994) found that the optimum number for the marsh clam, *Rangia cuneata*, is 10 individuals. The mean total hydrocarbon concentrations for each group were found to be statistically the same according to the Kruskal-Wallis non-parametric analysis of variance ($H_{2,9}$=5.95; P≤0.0509).

Hydrocarbon concentrations, in μg/g dry weight, for the american oyster *Crassostrea virginica* are summarized in Table 1. The average (± one standard deviation) concentrations of total hydrocarbons ranged from 157 ± 144 μg/g in February, 52.8 ± 21 μg/g in March, 140 ± 73 μg/g in April, 92 ± 96 μg/g in May, 70 ± 20 μg/g in June and 36 ± 14 μg/g in July. The observed differences were found to be highly significant by the non-parametric (Kruskal-Wallis) analysis of variance ($H_{5,34}$=19.2; P≤0.0017). Similar results were found for the other hydrocarbon fractions. The average Carbon Preference Index (CPI), ranged from 1.12 ± 0.27 in February, to 0.96 ± 0.45 in March, 0.79 ± 0.23 in April, 1.37 ± 0.41 in May, 1.07 ± 0.47 in June, and 0.91 ± 0.24 in July. It was always close to one, which indicates that the hydrocarbons are petrogenic (UNEP/IOC/IAEA 1991). The observed differences in the monthly means were not statistically different ($H_{5,34}$=7.75; P≤0.17). The concentrations of the methyl- derivatives of naphthalene and phenanthrene were always higher than those of the parent compounds, confirming the petrogenic origin of the hydrocarbons (UNEP/IOC/IAEA 1991).

The results obtained in this study are compared in Table 2 to total hydrocarbon

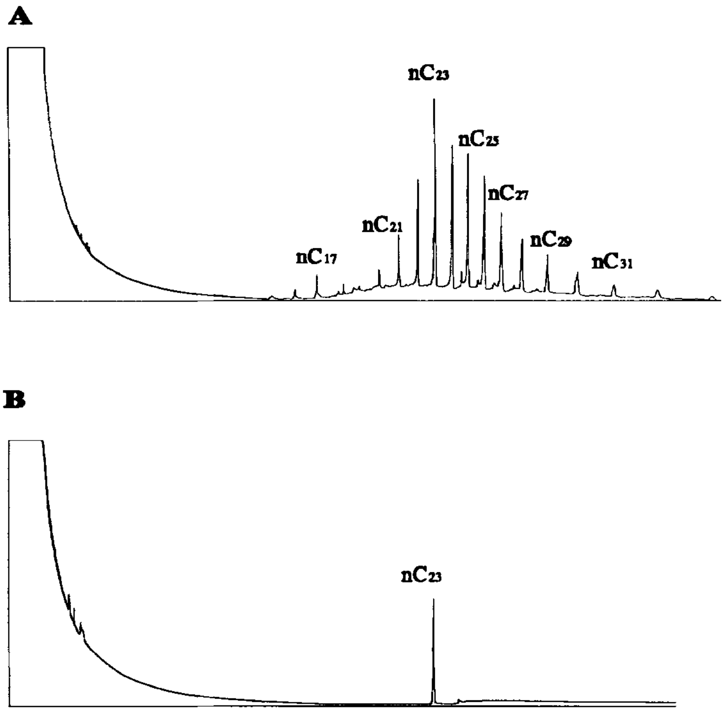

Figure 3. Gas chromatograms of the aliphatic fraction of pooled samples with a) 15 individuals, and b) 20 individuals.

concentrations found by other authors in coastal lagoons from the southern Gulf of Mexico. The mean concentration of total hydrocarbons obtained here (91.1 ± 85.7 μg/g) for Laguna de Terminos falls within the range of average values found previously by Botello (1979; 1980), indicating no temporal trend in over a decade. These concentrations are slightly higher than those found at Mecoacan and Carmen-Machona, two coastal lagoons in Tabasco, Mexico, where there are extensive activities of the oil industry. The very high concentrations reported by Castro (1981) are probably because hydrocarbons were quantified gravimetrically.

The geometric mean concentration of PAH's was 0.24 μg/g, which is slightly lower than the value obtained by the NOAA *National Status and Trends Program* (O'Connor 1992) of 0.26 μg/g, and much lower than the concentration considered "high" of 0.89 μg/g.

The relatively high concentrations of petrogenic hydrocarbons found in this work, as compared to those found in nearby coastal lagoons with direct activities of the oil industry, are cause for concern for the health of this valuable ecosystem. These results are confirmed by the recent report of high concentrations of hydrocarbons in both sediments and marsh clams (*Rangia cuneata*) in Laguna de Pom, a lagoon adjacent to Laguna de Terminos (Alvarez-Legorreta *et al.* 1994).

Table 1. Hydrocarbon concentrations, in µg/g dry weight, in the oyster *Crassostrea virginica*, from Laguna de Terminos, Mexico.

Month	Station	Aliphatics (µg/g)	UCM* (µg/g)	Aromatics (µg/g)	Total (µg/g)	PAH** (µg/g)	CPI
February	Atasta	14	63	46	123	0.43	0.91
	Boca Chica	8	57	67	132	0.40	1.02
	Estero Pargo	14	289	146	448	0.48	1.17
	Chumpán	1	42	40	83	0.27	0.80
	Chacahito	2	35	38	74	0.17	1.57
	Panlau	2	52	30	84	0.38	1.23
March	Atasta	1	28	19	49	0.02	0.67
	Boca Chica	1	23	16	40	0.04	0.79
	Estero Pargo	3	60	25	88	0.39	1.75
	Chumpán	1	33	19	53	0.13	0.86
	Chacahito	1	24	8	33	0.08	0.74
April	Atasta	2	72	7	81	0.11	0.61
	Boca Chica	5	92	31	128	0.35	0.90
	Estero Pargo	4	59	43	106	0.20	1.02
	Chumpán	3	36	76	115	0.12	0.5
	Panlau	5	225	37	267	0.31	0.94
May	Atasta	1	21	15	37	0.29	1.30
	Boca Chica	2	24	21	47	0.33	0.81
	Estero Pargo	2	25	20	47	0.33	1.14
	Chumpán	6	88	189	283	1.24	1.52
	Chacahito	1	20	27	47	0.27	2.05
	Panlau	2	61	27	90	0.75	1.39
June	Atasta	2	23	31	56	0.20	0.68
	Boca Chica	1	19	26	47	0.15	0.75
	Estero Pargo	1	24	37	62	0.15	0.59
	Chumpán	2	24	43	69	0.91	1.80
	Chacahito	3	38	53	93	0.42	1.35
	Panlau	3	54	37	94	0.46	1.24
July	Atasta	1	13	20	34	0.24	1.05
	Boca Chica	1	8	16	25	0.17	0.81
	Estero Pargo	1	18	8	27	0.31	1.08
	Chumpán	1	11	20	32	0.29	1.19
	Chacahito	1	14	49	64	0.18	0.52
	Panlau	1	19	17	37	0.15	0.83

* Unresolved Complex Mixture. ** Polynuclear Aromatic Hydrocarbons

Table 2. Comparison of the concentrations, in µg/g, of total hydrocarbons in oysters found in this study with published values.

Hydrocarbons	C P I	Place	Reference
37	1.2	Terminos	Botello, 1980
115	1.3	Terminos	Botello, 1979
2,580	Not given	Mecoacan	Castro, 1981
77	0.9	Mecoacan	Gold, 1992
87	0.7	Carmen-Machona	Gold, 1993
91	0.9	Terminos	This Work

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