

## Hydrocarbon Concentrations in Oysters (*Crassostrea virginica*) and Recent Sediments from Three Coastal Lagoons in Tabasco, Mexico

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The Mexican state of Tabasco, located in the southern Gulf of Mexico, is one of the main producers of Eastern Oysters, *Crassostrea virginica*, with an annual gross production of 10,000 tonnes. It is also one of the major producers of crude oil, with 20 to 25% of Mexico's domestic production. The high visibility of the oil industry has produced conflicts with other users of the coastal zone, such as fishermen, who blame the industry for an increased mortality of oysters in 1992- 1993.

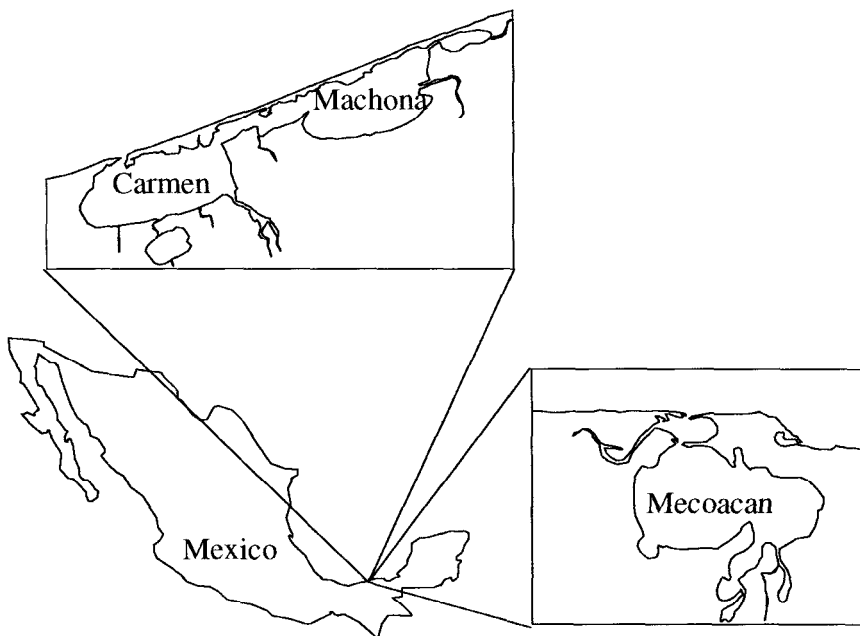
To determine the impact of oil pollution on oyster production, hydrocarbon residue concentrations were determined in oysters and recent sediments in the three main coastal lagoons in Tabasco : El Carmen, La Machona and Mecoacan. To look for pollution sources other than petroleum, other contaminants were analyzed, such as trace metals (Marin *et al.*, 1997).

### MATERIALS AND METHODS

El Carmen is located 18° 18' 30" and 18° 14' 30" North latitude and 93° 44' 30" West longitude, with a surface area of 91 km<sup>2</sup>. La Machona is located between 18° 25' 30" and 18° 20' 00" North latitude, and 93° 33' 50" and 93° 40' 00" West longitude, with a surface area of 76 km<sup>2</sup>. These lagoons communicate through a shallow channel, El Pajonal. The river Santana drains into La Machona, and river San Felipe into El Carmen. Mecoacan is located at 93° 10' North latitude and 18°28' West longitude, with a surface area of 50 km<sup>2</sup>. Three rivers drain into this lagoon : Rio Seco, Cuxcuchapa and Escarbado (Vázquez-Gutiérrez, 1994).

Oysters (*Crassostrea virginica*) and recent sediments were collected at 35 sampling stations in the coastal lagoons Mecoacan, El Carmen and La Machona (Fig. 1), in June, September and November 1992, and May 1993. Sediments were collected with a

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**Figure 1.** Map of the study zone, showing the three lagoons studied.

0.1 m<sup>2</sup> Van Veen grab and the oysters either by diving (in Mecoacan) or with an oyster rack. All samples were transported to the laboratory to CINVESTAV in Merida for further analysis. Since oysters were not always available at all stations, the total number of samples analyzed was 108.

Hydrocarbon concentrations in sediments were determined according to IOC/UNESCO (1982), and organisms following the method adopted by IOCARIBE (1987). The organisms were pooled at each station, taking 15 individuals per station to minimize the variance (Gold et al., 1995a). Oysters were digested with 6 M NaOH and the non-saponifiable material extracted 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 equipped with a flame ionization detector, a Hewlett-Packard 200  $\mu$ m i.d. x 25, 0.33  $\mu$ m 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 an n-C<sub>25</sub> 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.

Sediments were refluxed for 1.5 hr with 3% KOH in methanol, and the non-saponifiable material extracted with hexane. The extract was purified and divided into aliphatic and aromatic fractions by column chromatography with totally activated alumina. Gas chromatographic analysis was the same as for organisms.

The results were highly non-gaussian and heterocedastic. Since variances could not be stabilized using the usual transformations, central values are reported as medians, and differences between means are tested by randomization of the between-groups sum of squares (Manly, 1991).

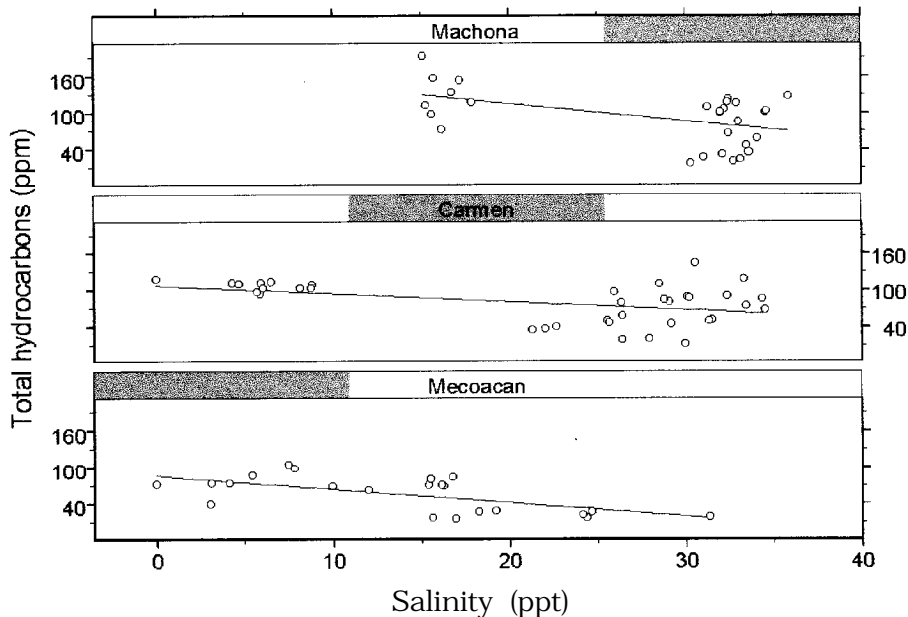
## RESULTS AND DISCUSSION

The median concentration of each hydrocarbon fraction, in the Eastern Oyster for each lagoon and sampling trip are given in Table 1.

**Table 1.** Median concentrations, in µg/g dry weight, of hydrocarbons in the Eastern Oyster, *Crassostrea virginica*, from Mecoacan, Carmen and Machona, Mexico.

Month	Lagoon	Aliphatic c	UCM <sup>†</sup>	Aromatic c	Total	PAH
June 1992	Mecoacan	1.18	13.0	56.0	69.4	0.203
	Carmen	1.66	14.3	51.0	65.6	0.127
	Machona	2.27	15.8	91.5	107.2	0.141
Sept 1992	Mecoacan	2.74	35.1	54.1	87.9	0.192
	Carmen	4.60	49.7	29.5	85.4	0.323
	Machona	4.89	42.7	57.8	102.9	0.615
Nov 1992	Mecoacan	1.68	17.5	44.0	71.2	0.334
	Carmen	2.36	44.9	54.2	107.4	0.316
	Machona	2.36	42.0	70.2	117.3	0.402
May 1993	Mecoacan	3.49	5.12	14.9	21.1	0.145
	Carmen	2.58	9.80	18.7	34.4	0.162
	Machona	2.50	8.14	9.61	26.4	0.459

<sup>†</sup> Unresolved Complex Mixture.



**Figure 2.** Total Hydrocarbons in oysters as a function of salinity for each lagoon.

Total hydrocarbons and PAHs were always higher in Machona, except for the May 1993 sampling for total hydrocarbons and the June 1992 sampling for PAHs. The observed differences between lagoons ( $P=0.0014$  for total hydrocarbons, and  $P=0.026$  for PAHs) were highly significant. The unresolved aliphatics (UCM) were always higher than the resolved aliphatics, which indicates that the hydrocarbons have been degraded. Median concentrations of aromatics were higher than the corresponding medians of both aliphatics and UCM.

The median concentrations of all hydrocarbon fractions, with the exception of aliphatics, tend to be lower in May than in the other months. The observed differences between months ( $P=0.000$  for total hydrocarbons, and  $P=0.0036$  for PAHs) were highly significant. May is at the peak of the dry season in this region, and the observed decrease in concentration might be related to salinity. This can be seen in Fig. 2, which shows the concentrations of total hydrocarbons (in  $\mu\text{g/g}$ ) as a function of salinity (in ppt) for each lagoon. The decrease in hydrocarbon concentrations with increasing salinities indicates that there is a riverine input of hydrocarbons. The same effect was observed for trace metals in these lagoons (Marin *et al.*, 1997).

The overall median concentration of total hydrocarbons obtained here, 79.8  $\mu\text{g/g}$ , is lower than the mean concentration, 91  $\mu\text{g/g}$ , found for the same oysters in Laguna de Terminos, a neighboring lagoon where no increased oyster mortalities have been reported (Gold *et al.*, 1995a).

Using a two population distribution model, Jackson *et al.* (1994) reported median PAH concentrations (for the low concentration population) in oysters (*C. virginica*) in the northern Gulf of Mexico between 208 and 353  $\text{ng/g}$ , for the years 1986- 1990, which compares well with the overall median obtained here (for the same species) of 272  $\text{ng/g}$ . This value is very close to the 240  $\text{ng/g}$  found in Laguna de Terminos (Gold *et al.*, 1995). The median concentrations in this work are in the range reported by Sericano *et al.* (1995) for the International Mussel Watch. The highest median concentration obtained here, 614.7  $\text{ng/g}$  in Machona in September 1992, does not approach the median values reported by Jackson *et al.* (1994) for their population with high values, of 1075 to 1910  $\text{ng/g}$ , which indicates that there seems to be no problem with PAH pollution in the three coastal lagoons analyzed here.

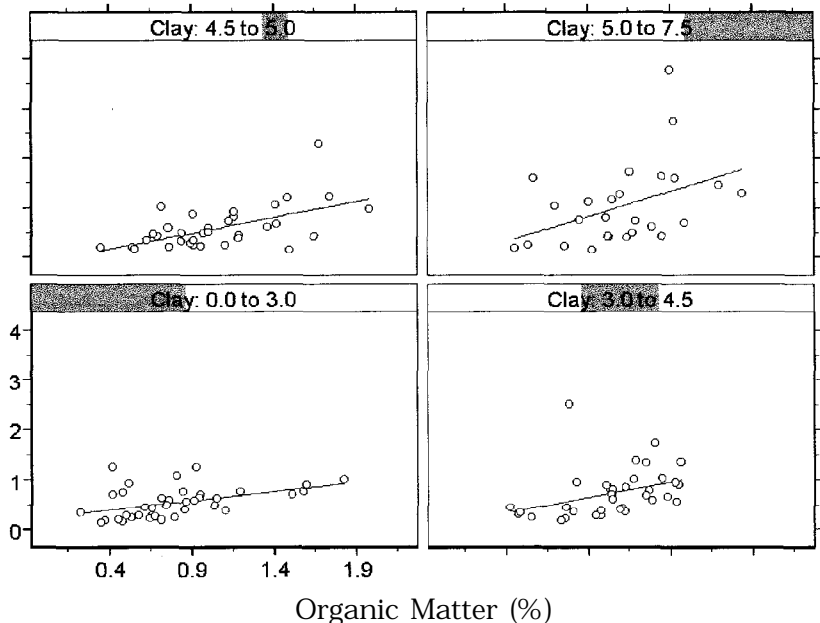
The median concentrations of hydrocarbons in sediments, by lagoon and sampling trip, are given in Table 2. Also, the median clay fraction is given.

**Table 2.** Median concentrations of hydrocarbons, in  $\mu\text{g/g}$  dry weight, and clay fraction in sediments from Mecoacan, Carmen and Machona, Mexico.

Month	Lagoon	Aliph.	UCM	Arom.	Total PAH <sup>†</sup>	Clay <sup>‡</sup>
June 92	Mecoacan	0.565	5.36	1.42	6.89	3.00
	Carmen	0.490	6.08	1.32	8.11	6.24
	Machona	0.625	4.98	0.775	6.33	4.51
Sept 92	Mecoacan	0.339	3.02	0.333	3.77	2.00
	Carmen	0.110	4.52	0.400	5.07	5.00
	Machona	0.138	2.82	0.415	3.55	5.00
Nov 92	Mecoacan	0.279	2.28	0.429	2.95	2.25
	Carmen	0.375	4.41	0.624	5.60	4.50
	Machona	0.278	2.25	0.341	3.32	5.00
May 93	Mecoacan	0.214	2.32	0.241	2.80	5.00
	Carmen	0.523	7.54	0.584	8.78	4.50
	Machona	0.457	5.07	1.01	6.66	5.00

<sup>†</sup> In  $\text{ng/g}$ . <sup>‡</sup> In %.

The median concentrations of the unresolved hydrocarbons were higher than those of the resolved aliphatics, which indicates



**Figure 3.** Aromatic hydrocarbon concentrations in sediments as a function of the organic matter content, given clay.

weathering of the oil. As opposed to hydrocarbons in oysters, the concentration of aromatics was of the same order as the concentration of aliphatics. Total hydrocarbons and PAHs were always higher in El Carmen. These differences were highly significant for total hydrocarbons ( $P=0.0086$ ), but not for PAHs ( $P=0.165$ ). Total hydrocarbons were significantly higher in June and May ( $P=0.0002$ ), and PAHs were lowest in September ( $P=0.0076$ ). The differences between months do not seem to be related to salinity.

The overall median PAH concentration found here is 12.0 ng/g, which is much lower than the critical concentration of Long and Morgan (1990) of 35,000 ng/g and the unadjusted “high” concentration of 2,400 ng/g found by O’Connor (1990) for the northern Gulf of Mexico. This value is close to the concentrations of PAHs found at the unimpacted areas in Sarasota Bay, Florida (Sherblom *et al.*, 1995).

The concentration of hydrocarbons in sediments seems to be controlled by the organic matter content and the clay fraction, as can be seen in Figure 3, which shows the concentration of aromatic hydrocarbons as a function of organic matter,

conditioned on clay content using the equal counts algorithm (Cleveland, 1993). There is a strong linear dependence of hydrocarbon concentration on organic matter, and the slope increases as the clay fraction increases.

From the data presented here, there seems to be no evidence that the hydrocarbon concentrations found in these coastal systems are producing the oyster mortality. The concentration of pollutants, particularly metals such as cadmium, and salinity were correlated with the prevalence of histopathological lesions in different tissues.

However, these lesions were not causing the increased mortalities (Gold *et al.*, 1995b). The presence of oyster pathogens (*Perkinsus marinus*) is another possible confounding factor (Burreson *et al.*, 1994).

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