

Table 1. Percentage of starry flounder with pleomorphic nuclei.^a

Station	% with NP rating			N
	1	2	3	
Berkeley	55	29	17	42
Oakland	65	22	13	23
San Pablo Bay	66	31	3	29
Vallejo	64	36	0	22
Santa Cruz	77	23	0	13
Russian River	97	3	0	29

^aAbbreviations: NP, pleomorphic nuclei; N, sample size. Ratings: 1, <5% of erythrocytes were pleomorphic; 2, 5 to 50% of erythrocytes were pleomorphic; 3, >50% of erythrocytes were pleomorphic.

In this study, Jeffrey N. Cross of SCCWRP and Jo Ellen Hose of Occidental College quantified micronuclei in peripheral circulating erythrocytes of starry flounder (*Platichthys stellatus*) and correlated micronucleus frequencies with body burdens of chlorinated organic contaminants. The flounder were collected by otter trawl from four sites in San Francisco Bay and two sites along the outer coast of central California (Figure 1) during two sampling periods in the winter of 1986-87.

Micronuclei are small, secondary nuclei formed after chromosome breaks (Schmid 1976). They may arise spontaneously, but the induction of micronuclei above background levels is a sensitive indicator of genotoxic

Micronuclei in Starry Flounder from San Francisco Bay

damage resulting from exposure to mutagens (Heddle et al. 1983).

Blood was collected from each fish immediately after capture, and smears were prepared on the ship. The smears were then stained in the laboratory, and the number of micronucleated erythrocytes per 1000 erythrocytes was determined. The erythrocytes were studied for degree of pleomorphism (loss of the usual elliptical shape of the nucleus) (Table 1) and for the presence of detached and at-

tached micronuclei (Figure 2). If a pleomorphic nucleus had a projection greater than about one-fourth the nuclear diameter and the projection terminated in a chromatin mass, it was considered an attached micronucleus.

Micronuclei (detached and attached) and nuclear pleomorphism are manifestations of cytotoxicity. Detached micronuclei are formed after chromosomal breakage or spindle damage. Attached micronuclei and severe nuclear pleomorphism may be

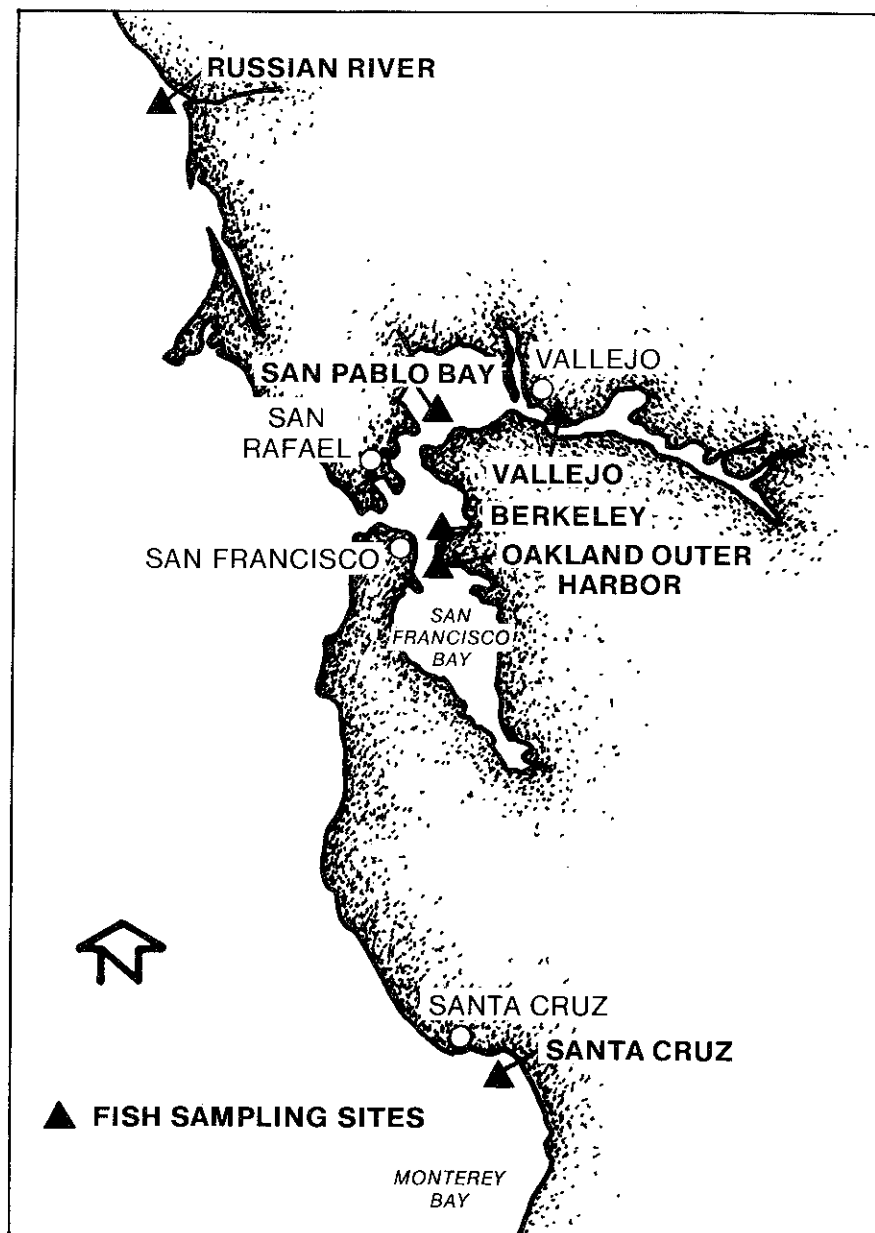


Figure 1. Map of the study sites.

the result of (1) chromosomal breakage or spindle damage, (2) destabilization of the nuclear membrane by certain chlorinated organics or intermediates of polynuclear aromatic hydrocarbon metabolism, or (3) agents that cause chromatin clumping (chromocenter formation) or margination (chromatin adhering to nuclear membrane).

Cross and Hose found that micronucleus frequencies were significantly higher among starry flounders collected within San Francisco Bay than among fish collected on the outer coast (Table 2). A significantly higher proportion of fish from the outer coast had no visible micronuclei. Within San Francisco Bay, there were no significant differences in

micronucleus frequency. Fish with high micronucleus frequencies also had a high incidence of misshapen nuclei (nuclear pleomorphism).

In addition, micronucleus frequencies (1) were not significantly different among male starry flounders from the bay and the outer coast; (2) were significantly higher among females from the bay than among females from the outer coast; and (3) were positively correlated with fish size among females, but not among males, from San Francisco Bay stations.

The composition of organic contaminants in the livers of starry flounders was very similar among the four sites within San Francisco Bay and was slightly less similar between fish collected within the bay and on the outer coast (Figure 3). Fish from central San Francisco Bay had higher mean levels of contaminants than did fish from northern San Francisco Bay, and fish from the northern bay had higher mean levels than did fish from the outer coast (Table 3). Except for chlordane and lindane, contaminant concentrations were not correlated with fish size. Chlordane and lindane concentrations were higher in females collected in northern San Francisco Bay; these fish were generally smaller than females collected in the central bay.

The lack of compositional differences among fish from the

different sites and the presence of fish with low organic contaminant concentrations at all of the sites suggest that (1) chlorinated hydrocarbons are present throughout the bay system and on the outer coast and (2) some starry flounders move throughout the bay and between the bay and the outer coast. In a review of data on chlorinated hydrocarbons in sediments and biota, Phillips and Spies (manuscript in review) concluded that some compounds, particularly polychlorinated biphenyls (PCBs), are widespread in the San Francisco estuarine system.

Micronucleus frequencies were not correlated with organic contaminant concentrations in this study. The results are consistent with the non-chromosome-breaking properties of DDTs and

Table 2. Starry flounder micronucleus frequencies per 1000 erythrocytes.^a

Station	Mean	SD	Median	N	PROP
Berkeley	1.9	2.33	1.3	42	0.119
Oakland	1.5	2.06	0.5	23	0.348
San Pablo Bay	1.3	1.19	1.0	29	0.172
Vallejo	2.2	1.49	0.5	22	0.091
Santa Cruz	0.6	0.77	0.5	13	0.462
Russian River	0.4	0.74	0.0	29	0.655

^aAbbreviations: SD, one standard deviation; PROP, proportion of zeros.

Table 3. Summary of starry flounder liver contaminant concentrations ($\mu\text{g/kg}$ wet weight) by site. Data are means and one standard deviation (in parentheses).^a

Station	N	Total DDT	Total Pesticides	Total PCB
Berkeley	18	202 (145)	50 (32)	422 (306)
Oakland	16	189 (120)	47 (34)	438 (312)
San Pablo Bay	14	161 (90)	48 (32)	110 (53)
Vallejo	14	160 (112)	30 (17)	110 (73)
Santa Cruz	4	73 (73)	119 (142)	89 (63)
Russian River	14	152 (245)	34 (44)	152 (253)

^aN is the sample size; Total DDT is the sum of *p,p'*-DDE and *p,p'*-DDD; Total Pesticides is the sum of chlordane, dieldrin, heptachlorepoide, and lindane;

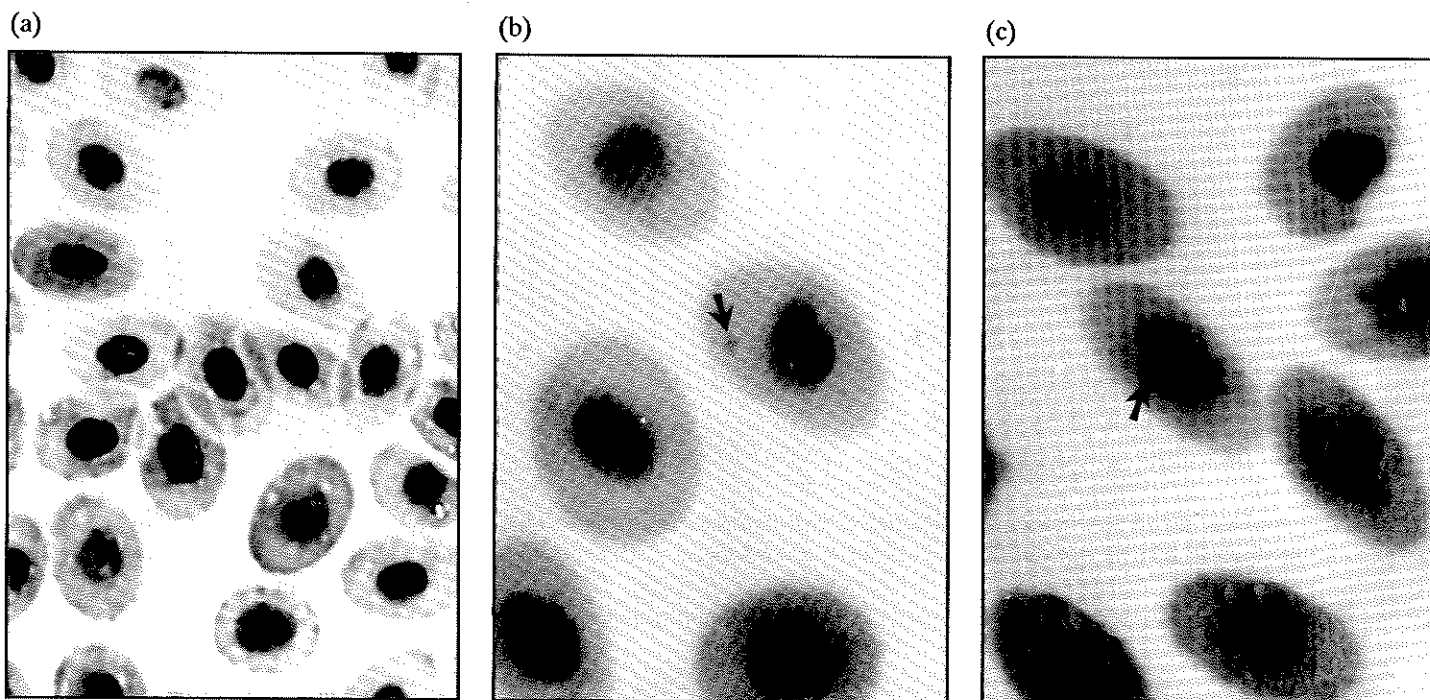


Figure 2. Erythrocytes (stained with May Grunwald-Giemsa) of starry flounder. (a) Sample from fish from the Russian River. Note the uniform elliptical appearance of the nuclei. White circles in the cytoplasm are artifacts. Magnification, X 5600. (b) One erythrocyte contains a detached micronucleus (arrow). Magnification, X 9450. (c) Sample from fish from San Pablo Bay shows an attached micronucleus (arrow). Many nuclei are pleomorphic. Magnification, X 9450.

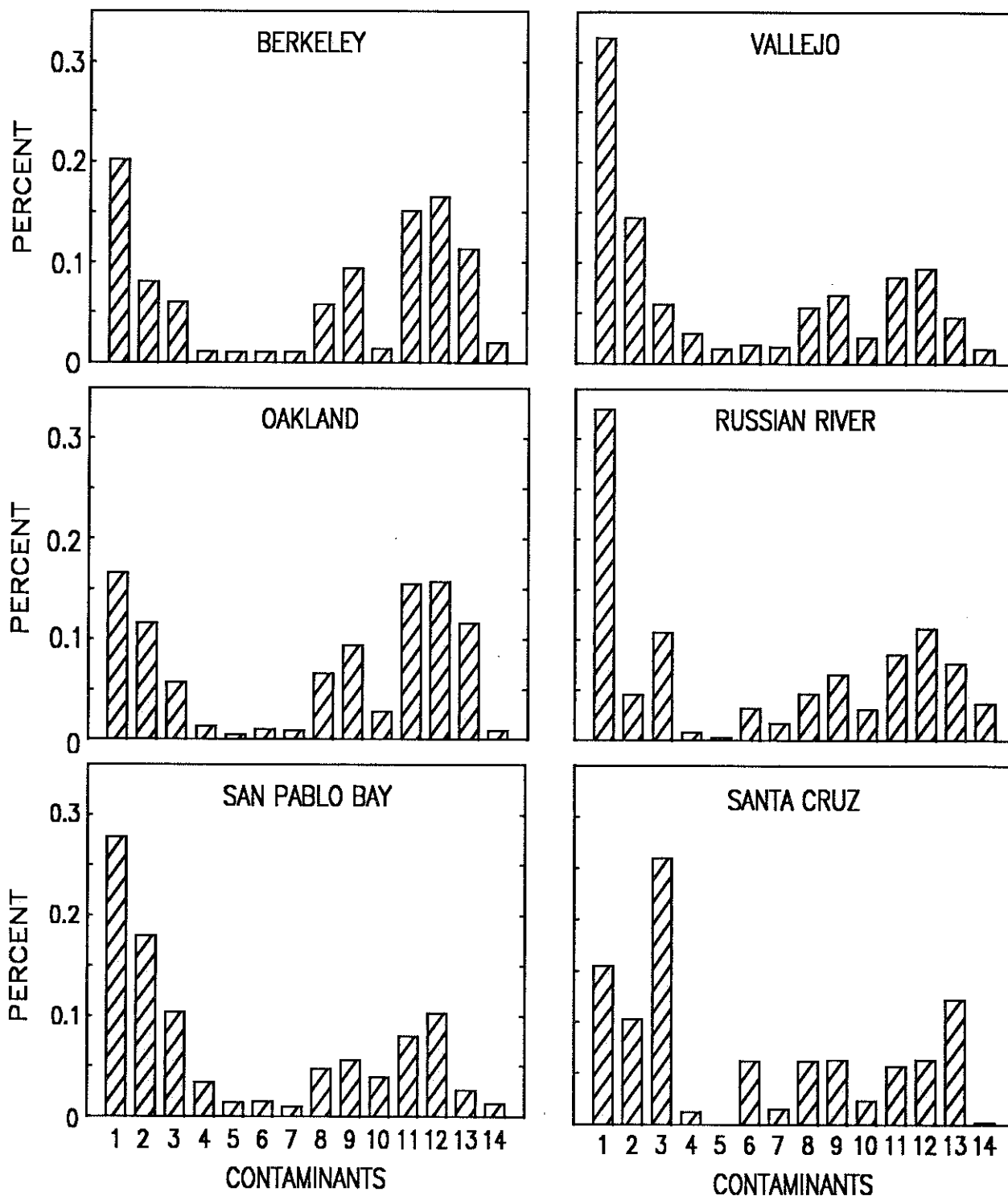


Figure 3. Normalized mean percentage of chlorinated hydrocarbons found in fish livers. Contaminant numbers on the abscissa are as follows: 1, *p,p'*-DDE; 2, *p,p'*-DDD; 3, dieldrin; 4, chlordane; 5, lindane; 6, heptachlorepoide; 7, PCB44; 8, PCB101; 9, PCB118; 10, PCB128; 11, PCB138; 12, PCB153; 13, PCB180; 14, PCB206. PCB numbers indicate specific identities of the congeners (Ballschmiter and Zell 1980). Numbers increase with increasing molecular weight.

PCBs (Green et al. 1975, Heddle et al. 1983). It is well known from many mammalian and a few fish experiments that compounds such as metabolites of mutagenic polynuclear aromatic hydrocarbons (e.g., benzo[a]pyrene), mutagenic nitroaromatics and phenols, and carcinogenic volatile monoaromatics (benzene) enhance micronucleus formation (Schmid 1976, Hoofman and de Raat 1982, Heddle et al. 1983). These compounds were not measured in this study but are present in San Francisco Bay and may be responsible for the micronucleus frequencies observed.

Micronucleus frequencies of starry flounder from stations within San Francisco Bay (mean = 1.7‰ , median = 0.5‰) were lower than micronucleus frequencies of white croaker (mean = 3.4‰ , median = 2.5‰) and kelp bass (mean = 6.8‰ , median = 4.3‰) from contaminated sites near Los Angeles (outer San Pedro Bay and eastern Palos Verdes Peninsula, respectively) (Hose et al. 1987).

Acknowledgments

This work was funded by National Oceanic and Atmospheric Administration Grant No. 50ABNC700091.

Chemical analyses were performed by Dr. R. Spies and D. Rice of Lawrence Livermore National Laboratory.

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