between the location of the spots detected by iodine and the repellency as determined in the olfactometer. We have not yet identified the repellent substances. The significance of skin lipids in terms of their effect on the behavior of mosquitoes can only be conjectured. Perhaps the attraction of the host to the mosquito depends on a balance between naturally occurring repellents and attractants.

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Clostridium botulinum Type F from Marine Sediments

Abstract. Clostridium botulinum *Type F has been demonstrated in two samples of marine sediments. One sample was taken 83 kilometers off the coast of California; the other, 100 kilometers off the coast of Oregon. Cultures of this type have not been reported previously in the United States, and only once before in the whole world.*

In a survey on the incidence of *Clostridium botulinum* in the coastal areas of Alaska, Washington, Oregon, and California, two samples of marine sediment have yielded cultures of *Cl. botulinum* Type F. The first and only previous culture of *Cl. botulinum* Type F was isolated from a homemade liver paste connected with an outbreak of human botulism on the Danish island Langeland (*1*). One of the five persons who ate the liver paste suffered no harm, but three had severe attacks of botulism, and the fifth person died three days later (*2*). This culture has since been described as a prototype strain designated as *Cl. botulinum* Type F (*3*).

The marine sediments that yielded cultures of *Cl. botulinum* Type F in this laboratory were collected 83 kilometers from the coast of California (sample A) and 100 kilometers from the coast of Oregon (sample B). Sample A came from a depth of 1646 meters at 42°N latitude and sample B from a depth of 1326 meters at 43°N latitude.

Portions (approximately 5 g) of these mud samples were inoculated into 25 ml of broth containing glucose, peptone, trypticase, beef infusion, and ground meat, a modification of Dolman's medium (*4*). The inoculated tubes were incubated anaerobically (95 percent nitrogen and 5 percent carbon dioxide) at 25°C for 5 days. A portion of the broth was then centrifuged at 10,000 rev/min (12,000*g*). The supernatant was tested for toxicity by injecting two mice (Swiss Webster strain) intraperitoneally with 0.4 ml of a 1:2 dilution of the supernatant and gelatin-phosphate buffer. Characteristic symptoms of botulism, if present, occurred within 20 hours after injection.

The toxin was identified by injecting pairs of mice with 0.5 ml of mixtures composed of 0.4 ml of a 1:2 dilution of the supernatant and 0.1 ml of one of the following antitoxins: a polyvalent antitoxin (Types A, B, C, E, and F in equal concentrations); or an individual antitoxin (Types A, B, C, D, E, or F). Heat lability of the toxin was determined by injecting a pair of mice with 0.4 ml of a 1 : 2 dilution of the sample supernatant that had been heated for 10 minutes at 100°C and then cooled. All mice were observed for at least 6 days.

The data supporting the identification of the culture as *Cl. botulinum* Type F are summarized in Table 1. Neutralization of the toxin was achieved in both samples only by the Type F antitoxin and by a polyvalent antitoxin containing Type F. Heating the supernatant for 10 minutes at 100°C inactivated the toxin. The toxin of sample A was not neutralized by Types A, B, C, D, or E antitoxin, since the mice injected with this toxin died within 19 to 23 hours and had characteristic symptoms of botulism. The same results were obtained for sample B with the exception (not shown in Table 1) that Type E antitoxin showed some neutralization of the toxin, but only when the Type E antitoxin was in large excess. Cross neutralization between Type F and Type E was also reported from the Denmark isolate (*1, 3*). The titer of the toxin from both cultures was 6 to 20 minimum lethal doses (mouse) per milliliter of medium.

Table 1. Neutralization reaction pattern of cultures identified as *Clostridium botulinum* Type F. Results are given as number of mice dead out of number tested. S is supernatant; HS, heated supernatant.

Source of toxin	Anti-toxin	Result	
		Sediment sample A	Sediment sample B
S	None	6/6	6/6
S	ABCEF	0/2	0/2
S	A	4/4	4/4
S	B	4/4	4/4
S	C	4/4	4/4
S	D	2/2	2/2
S	E	4/4	4/4
S	F	0/6	0/6
HS	None	0/2	0/2

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Deformity of Forelimb in Rats: Association with High Doses of Acetazolamide

Abstract. *Deformities of the right forelimb occurred in a number of offspring of rats given high doses of the carbonic anhydrase inhibitor, acetazolamide, during pregnancy. In most cases this was the only deformity found. More than 20 times the usual therapeutic dose rate used for humans was required to produce this effect in rats.*

We have found a remarkably specific and reproducible malformation of the right front extremity in a number of offspring of rats given a diet containing large concentrations of acetazolamide during pregnancy. Acetazolamide (2-acetylamino-1,3,4,-thiadiazole-5-sulfonamide) is a potent carbonic anhydrase inhibitor which has been used as a diuretic in human patients since 1953.