

## Isolation of Non-O1 *Vibrio cholerae* Serovars from Oregon Coastal Environments†

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Water, sediment, and shellfish from three Oregon estuaries were cultured for pathogenic *Vibrio* species. Non-O1 serovars of *V. cholerae* were the most common pathogenic *Vibrio* species recovered. Non-O1 *V. cholerae* were isolated from all three estuaries sampled, covering an area of about 170 miles along the Oregon coast. Non-O1 *V. cholerae* were isolated from water and sediment, but not shellfish, at temperatures ranging from 11 to 19°C and salinities of 2.3 to 26‰. Sixteen isolates representing 12 different non-O1 serovars were identified, while four non-O1 *V. cholerae* isolates failed to react with any of the 54 antisera tested. These results indicate that non-O1 *V. cholerae* serovars can be found over a large geographic area and under a variety of environmental conditions. These organisms are apparently an autochthonous component of these estuarine microbial communities.

There has been heightened interest in the occurrence of potentially pathogenic *Vibrio* species in U.S. coastal environments since the 1978 outbreak of 11 cases of *V. cholerae* O1 in Louisiana (3). A number of infections by non-O1 serovars of *V. cholerae* have been documented in the United States, most of which were associated with U.S. Atlantic and Gulf coastal areas (5, 7). The isolation of non-O1 *V. cholerae* strains from California coastal waters has recently been reported (6). Reports of pathogenic *Vibrio* species from Pacific Northwest coastal areas have been limited to *V. parahaemolyticus* (2, 8). We report here the results of field studies of three Oregon estuaries from which numerous non-O1 serovars of *V. cholerae* were isolated.

Samples of water, sediment, oysters, and crabs were collected from Coos, Yaquina, and Tillamook Bays. Water was collected from approximately 2 cm below the surface in sterile 5-liter containers, and sediment was collected with a Ponar dredge. Temperature and salinities were measured electronically with a Yellow Springs Instrument Co. combination probe and meter (YSI model no. 33). Samples were processed as described by Roberts and Seidler (9). This included an 18-h enrichment in alkaline peptone broth at 35°C followed by inoculation onto thiosulfate-citrate-bile salts-sucrose (TCBS) agar (Oxoid Ltd.) and incubation at 35°C for an additional 18 h. Yellow, sucrose-fermenting colonies were picked from TCBS plates, and *V. cholerae* strains were identified as described previously (9). *V. cholerae* isolates were identified serologically with antisera prepared by Adams and Siebeling (1, 11). Non-O1 serovars of *V. cholerae* were identified based on the system developed

at Louisiana State University as described previously (1) and correlated with the *V. cholerae* serotyping system of Sakazaki and Shimada when possible.

Non-O1 *V. cholerae* was isolated from all three estuaries sampled from February through July 1980 at temperatures ranging from 11 to 19°C and salinities of 2.3 to 26‰ (Table 1). Non-O1 serovars of *V. cholerae* were isolated from water, sediment, and algae (seaweed), but not from crabs, clams, or oysters. Each isolate was serotyped with anti-O sera produced against 54 non-O1 *V. cholerae* serovars. Sixteen isolates agglutinated in 12 different serovar-specific antisera, while four isolates failed to agglutinate in any of the 54 antisera. Of the 16 isolates, 2 agglutinated in two anti-O sera (N and X; Z and AA), a finding which suggests that non-O1 *V. cholerae*, like O1 isolates, may express at least two O-antigen determinants (1, 11). Two of the serovars, H and J, were isolated from both Coos and Tillamook Bays, which are approximately 170 miles apart.

The public health significance of non-O1 *V. cholerae* in Oregon coastal waters remains to be determined. The concentrations of non-O1 *V. cholerae* from the three estuaries sampled ranged from less than 1 to 110 organisms per liter of seawater based on most-probable-number estimates as previously described (9). These concentrations are similar to those described for *V. cholerae* from other U.S. coastal waters (4). The facts that these concentrations are very low and no isolates were obtained from shellfish indicate that the risk of infection from Oregon shellfish was probably small, at least under those conditions under which samples were obtained. Numerous wound infections caused by non-O1 *V. cholerae* acquired from saline or brackish water have been documented (5), however. The potential for acquiring these types of infections from Oregon coastal waters exists, since the data presented here show that non-O1 *V. cholerae* can be isolated from geographically distant sites, from a wide range of salinity and temperature, and during various times of the year. These results suggest that non-O1 *V. cholerae* are autochthonous components of these Pacific Northwest estuarine communities in a manner similar to that described for *V. cholerae* in other U.S. coastal environments (4).

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† Technical paper 7765 from the Oregon Agricultural Experiment Station, Corvallis, OR 97311.

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TABLE 1. Non-O1 *V. cholerae* isolated from Oregon estuaries

Site (date)	Source	Temp (°C)	Salinity (%)	Serovar by:	
				LSU <sup>a</sup>	Sakazaki and Shimada
Yaquina Bay (February)	Water	12	16.2	A	26
Yaquina Bay (February)	Water	12	2.3	Ukn <sup>b</sup>	Ukn
Yaquina Bay (April)	Water	11	26	F	Ukn
Tillamook Bay (May) <sup>c</sup>	Water	17	19	G	8
Tillamook Bay (May) <sup>c</sup>	Algae	NA <sup>d</sup>	NA	G	8
Coos Bay (June) <sup>c</sup>	Water	19	21	J	6
Coos Bay (June) <sup>c</sup>	Water	19	21	Ukn	Ukn
Coos Bay (June)	Water	16	26	H	Ukn
Tillamook Bay (July)	Sediment	ND <sup>e</sup>	ND	J H Ie W J	6 Ukn 39 51 6
Tillamook Bay (July)	Sediment	17	26	H K	Ukn Ukn
Tillamook Bay (July)	Water	18.5	6.5	H N/X <sup>f</sup> Ukn Ukn Z/AA <sup>f</sup>	Ukn 41/57 Ukn Ukn 44/Ukn

<sup>a</sup> LSU, Louisiana State University.

<sup>b</sup> Ukn, Unknown; no reaction with available antisera.

<sup>c</sup> Each of these two pairs of samples came from a single site; all other samples were from different sites.

<sup>d</sup> NA, Not applicable.

<sup>e</sup> ND, Not determined.

<sup>f</sup> Cross-reactions between antisera prepared against different vaccine strains.

This work was supported in part by the Oregon State University Sea Grant College Program, National Oceanic and Atmospheric Association Office of Sea Grant, U.S. Department of Commerce, under grant NA 79AA-D00106, project R/FSD8.

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