

**Occurrence of enteroviruses in the West-Istrian Coastal Area  
of the Northern Adriatic, Croatia**

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Viral contamination of seawater occurs mainly through discharges of sewage effluents into rivers that ultimately find their way to the sea, or through offshore disposal of sewage outfalls.

A control of the presence of viruses in seawater, mussels and sediments collected in the Limski Kanal (mariculture area) and in the touristic centers along the west-istrian coast was performed during the 1989-1990 period. Simultaneously, a sanitary quality control of recreational and mariculture waters as well as mussels was conducted. The concentration of viruses in seawater was done by adsorption in the quartz sand (SCHWARTZBROD and LUCENA, 1978). The elution of viruses from sediment particles was done according to GERBA *et al.* (1977). The minced mussel meat was centrifuged in a buffered solution, followed by a supernatant inoculation on GMK cell line and precipitate reconcentration for hepatitis A virus antigen detection. The enteroviruses level was determined by plaque-assay using GMK cell line (LENETTE and SCHMIDT, 1979). Hepatitis A virus was detected by immune enzyme test (ELISA). The sanitary quality of waters and mussels was assessed according to WHO/UNEP (1977) guidelines.

In water samples only the hepatitis A virus was detected. Meanwhile, in controlled mussels several types of viruses were found (Hepatitis A virus, Coxsackievirus B3, Echovirus types 4 and 11). The following viruses appeared in the sediments: Hepatitis A virus, Coxsackievirus types B2 and B3 and Poliovirus type 3.

Concerning the proportion of virus positive samples (Table 1) waters and sediments from recreational areas were more polluted compared to Limski Kanal area. Meanwhile, a higher viral contamination of mussels was recorded in Limski Kanal area compared to recreational coastal waters.

Table 1. Occurrence of enteroviruses in different matrices.

Study area	Sea water			Sediments			Mussels		
	N	(+)	%	N	(+)	%	N	(+)	%
Limski Kanal area	27	4	14.8	17	6	35.3	19	6	31.6
Coastal recreational areas	23	7	30.4	33	15	45.5	40	11	27.5

N - number of tested samples, (+) - number of virus-positive samples

Concerning the seasonal aspect of viral contamination the highest number of virus' positive samples was recorded in the mariculture area (35.7 %) as well as in recreational areas (58.3 %) during springtime (Table 2). Due to the karstic type of the Istrian Peninsula and rainy winter-spring period an increased number of virus positive samples during the springtime is expected. The fall increase of viral contamination in recreational areas probably resulted from quadruple effluents enlargement of the touristic centers during the summer-fall period. Most touristic and municipal centers along the coast dispose their domestic wastewaters by traditional methods, which are quite inadequate to destroy viruses. Viruses survive in the marine environment for a prolonged time, especially if protected by particulate organic matter.

Table 2. Enteroviruses occurrence during the seasons of the 1989-1990 period.

Study area	Spring			Summer			Fall			Winter		
	N	(+)	%	N	(+)	%	N	(+)	%	N	(+)	%
Limski Kanal area	28	10	35.7	22	4	18.2	6	1	16.7	7	1	14.3
Coastal recreational areas	12	7	58.3	66	20	30.3	18	6	33.3	-	-	-

N - number of tested samples, (+) - number of virus-positive samples

The most frequent virus detected in water samples was the hepatitis A virus, particularly in Limski Kanal area. Due to the influence of polluted groundwaters from the nearby springs, periodically the internal part of this area did not meet adequate sanitary quality of water and shellfish (FUKS and DEVESCOVI, 1989). A simultaneous sanitary ambient quality control and virus presence detection revealed, that although the water sanitary quality was acceptable according to WHO standards (1983), viruses were detected in the water, mussels and sediments, respectively. The inevitable conclusion, based on an initial screening for detection of enteroviruses, particularly in Limski Kanal, is that the mariculture area is surrounded by potential sources of viral contamination. Undoubtedly, the present finding on the Kanal's utilization for shellfish and fish raising purposes should cause an alarm among all interested and responsible for preventing its contamination. Consequently, water and food sanitary surveillance, besides the present bacterial control, become inadequate. The introduction of virological tests should be considered, especially in cases where the present knowledge indicates that they are indispensable.

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