REVIEW OF FIELD APPLICATIONS OF THE MICROTOX TEST IN GREAT LAKES WATERS

by

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Management Perspective

Title: Review of field applications of the Microtox test in Great Lakes waters

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Perspective:

This manuscript reports new research results on a Microtox test survey of St. Lawrence River water, undertaken in October 1985 and June/July 1986. The results indicate the existence of zones of low to moderate toxicity in several areas of the river and the riverine lakes of the system. Areas so identified include the Brockville to Ogdensburg stretch, an area below Cornwall and Massena, the Lac St. Louis - Montreal and Montreal Harbour zone, nearshore samples below Sorel, and the southern part of Lac St. Francois, downstream from the mouths of the Yamaska and St. Francois Rivers. Most other samples, such as from the Ottawa River, Lac des Deux Montagnes and Quebec City areas showed no toxicity in this test.

The Microtox toxicity test and analyzer are evaluated for such field investigations on the basis of the above and our previously published work on the Detroit River and on Lake St. Clair. Although the correlations of the Microtox data with concentrations of specific organic and inorganic contaminants are not highly significant, it is concluded that this test is useful for the quick screening of large areas. Some correlations appear to exist with certain chemical groups, such as phenols. Therefore, this test is useful as one of a battery of biological and chemical tests for the determination of the health or impairment of a river or lake ecosystem.

Le présent manuscrit présente les nouveaux résultats de recherches portant sur des relevés d'essais Microtox pour l'eau du fleuve St-Laurent, entrepris en octobre 1985 et en juin-juillet 1986. Ces résultats indiquent l'existence de zones de toxicité faible à moyenne dans plusieurs parties du fleuve et des expansions lacustres du bassin. Les zones ainsi identifiées comprennent le parcours de Brockville à Ogdensburg, une zone en aval de Cornwall et de Massena, ainsi que la zone du lac St-Louis - Montréal et celle du Port de Montréal, des sites d'échantillonnage près du rivage en aval de Sorel, et la partie sud du lac St-François, en aval de l'embouchure de la Yamaska et de la St-François. La plupart des autres sites d'échantillonnage, comme ceux de la rivière des Outaouais, du lac des Deux-Montagnes et de la ville de Québec, ont été jugés non toxiques d'après cet essai.

L'essai de toxicité et l'analyseur Microtox sont évalués pour de telles études in situ d'après les critères ci-dessus et d'après nos travaux déjà publiés portant sur la rivière Détroit et le lac Ste-Claire. Bien que les corrélations des données Microtox avec les concentrations particulières de contaminants organiques et inorganiques ne soient pas très significatives, on peut conclure que cet essai est utile pour un tri rapide des échantillons de régions étendues. On note certaines corrélations avec certains groupes de produits chimiques comme les phénols. Par conséquent, cet essai est utile s'il fait partie de la panoplie d'essais biologiques et chimiques utilisés pour déterminer l'état de santé de l'écosystème d'un cours d'eau ou d'un lac.

ÉTUDE DES APPLICATIONS DE L'ÉPREUVE MICROTOX DANS LES EAUX DU LAC ST. CLAIR

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RÉSUMÉ

L'épreuve de toxicité Microtox, une épreuve biologique faisant appel à la bactérie marine luminescente Photobacterium phosphoreum, est utilisée sur place et au laboratoire pour déterminer l'état de contamination de l'eau dans la rivière Détroit, le lac St. Clair et le Fleuve Saint-Laurent. Les résultats de cette épreuve indiquent les endroits et les zones où il y a une diminution de la qualité de l'eau, et s'avèrent utiles en rapport avec d'autres études sur les mêmes endroits.

Cette étude présente de nouveaux résultats relatifs au lac St. Clair et au Fleuve Saint-Laurent et les discute à la lumière d'autres résultats biologiques, chimiques et physiques obtenus par la même occasion. On y présente les avantages et les limites de cette épreuve biologique dans les travaux préliminaires sur la contamination de l'eau dans les Grands Lacs et ailleurs.

REVIEW OF FIELD APPLICATIONS OF THE MICROTOX TEST IN GREAT LAKES WATERS[1]

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ABSTRACT

The Microtox™ toxicity test, a bacterial bioassay employing the luminescent marine *Photobacterium phosphoreum* as the test organism has been used in field and laboratory research to test for water contamination in the Detroit River, Lake St. Clair and the St. Lawrence River. The test results have indicated locations and zones of impaired water quality and have been found useful in connection with other studies on the same areas.

This review presents new data on the St. Lawrence River and discusses the results in conjunction with other biological, chemical and physical data collected at the same time. It assesses the advantages and limitations of this bioassay for exploratory work on water contamination in the Great Lakes and elsewhere.

INTRODUCTION

The Microtox™, [2] toxicity test is a bacterial bioassay which uses the luminescent marine *Photobacterium phosphoreum* as the test organism. Originally developed for quick toxicity determinations of industrial effluents (Bulich and Isenberg, 1981), we have used this test in field surveys to delineate contamination plumes in rivers (Ribo et al., 1985) and in lakes (Kaiser et al., 1988) as well as in the laboratory to determine the acute toxicities of pure chemical substances (Kaiser and Ribo, 1988; Kaiser, 1987).

The Microtox test has been described in detail by Bulich and coworkers (Bulich and Isenberg, 1981; Bulich et al., 1981) and a recent review by Ribo and Kaiser (1987) presents an evaluation of its applications and practical aspects. It should be noted here that this test, as other bioassays, measures the overall effect on the organisms of the substances and/or conditions present in the sample and, therefore, may or may not be correlated with the effects in another bioassay or with the presence or absence of a specific contaminant factor.

We describe here new results on water samples from the St. Lawrence River, evaluate these in conjunction with other environmental data from the same area, and review the applicability and usefulness of this bioassay for field investigations in the Great Lakes system.

EXPERIMENTAL

Over 370 surface (1m depth) and bottom (1m off bottom) water samples were collected from 191 stations in the St. Lawrence River between Kingston, Ontario and the mouth of the Saguenay River, Quebec, during the periods of 30 Sep. to 18 Oct. 1985, and 16 June to 9 July, 1986. The samples were stored at 4 °C and were analyzed within a few hours onboard the research vessel CSS *Limnos*. All samples were non-turbid and could be analyzed without prior filtration. For the analysis, 0.5 mL aliquots of osmotic adjustment solution (20 % NaCl in double distilled water) were added to 4.5 mL of sample in order to reach a 2% saline concentration in the samples to provide osmotic protection for the test bacteria. Then, 0.5 mL of this adjusted sample was added to a suspension of 0.01 mL of the reconstituted bacteria in 0.5 mL of ultrapure water. The light output was recorded before and after 15 min exposure at 15 °C. All measurements given are single determinations; in our experience, the combined instrumental and analytical variability of such samples is less than 0.03 Γ units. Further details of the procedure and data reduction are given by Ribo et al. (1985).

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1) Presented in part at the 23rd Can. Symp. on Water Pollution Research, Burlington, 18 Feb. 1988.
2) Microtox is a registered trademark of Microbics Corporation, Carlsbad, California, USA.

RESULTS

Table 1 gives the classification and enumeration of the samples in four ranges of Γ values for each sampling period. Of the 161 samples collected in October 1985, a total of 48% fall into classes 0 and 1, and 52% into classes 2 and 3. Of the latter, 40 samples (25%) had Γ values of 0.10 or greater with the highest observed value of Γ= 0.45 (station 75). In June 1986, both surface and bottom water samples were taken, some at the same stations as in 1985, but the majority from locations further downstream. Of the 191 samples (excluding the three estuary stations 6E100, 6E300 and 6E400), a total of 54 % fall into the classes 0 or 1, the remaining 46% into classes 2 or 3. Classes 2 and 3 include a range of $0.05 < \Gamma < 0.50$, which represents low to moderate acute toxic effects on *Photobacterium phosphoreum*. Of above 46%, only 20 samples (11%) had Γ values of 0.10 or greater, with the highest observed value of Γ = 0.25 (station 251S). Table 2 gives the Γ values and classification for each sample with the Γ values rounded to the nearest one hundredth, resulting in values of Γ= 0.05 being assigned either to class 1 or 2.

TABLE 1. Classification and enumeration of samples exclusive of estuary stations.

Gamma range	Class	Number of samples		
		All samples 1986	Surface water 1986	Surface water 1985
$\Gamma < 0.00$	0	21 (11%)	17 (12%)	25 (16%)
$0.00 < \Gamma < 0.05$	1	83 (43%)	60 (43%)	52 (32%)
$0.05 < \Gamma < 0.10$	2	67 (35%)	45 (32%)	44 (27%)
$0.10 < \Gamma < 0.50$	3	20 (11%)	17 (12%)	40 (25%)
$0.00 < \Gamma < 0.05$		191 (100%)	139 (99%)	161 (100%)

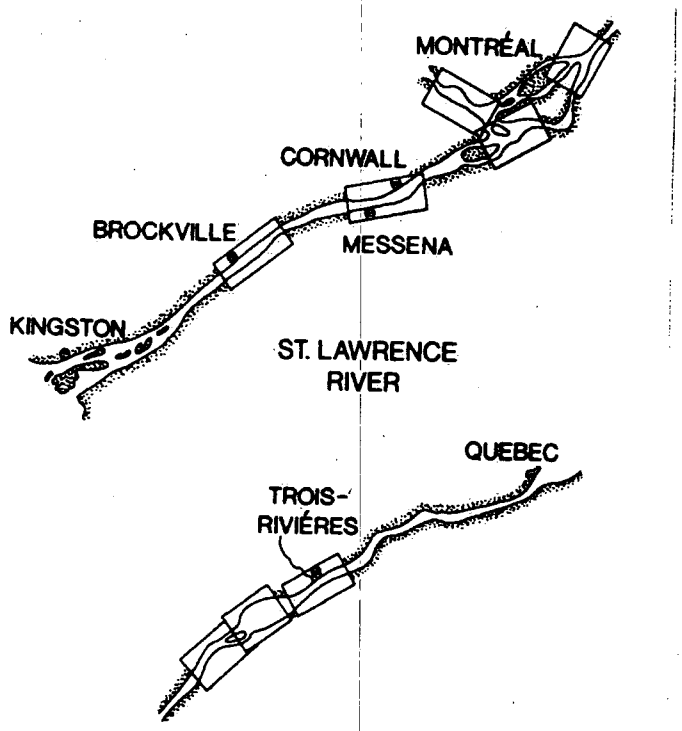


FIGURE 1. General map of the St. Lawrence River. Sampling areas indicated are shown enlarged in Figure 2.

TABLE 2. Gamma (Γ) values and classifications of St. Lawrence River water samples as determined by the Microtox test for 15 min exposure.

Station No.	Γ 1985	Γ 1986	Class 1985	Class 1986	Station No.	Γ 1985	Γ 1986	Class 1985	Class 1986
27	0.29	0.07	3	2	145	0.07	--	2	-
28	0.24	0.11	3	3	146	0.14	0.05	3	1
29	0.26	0.13	3	3	147	0.09	--	2	-
30	0.20	--	3	-	148	0.04	--	1	-
31	0.04	--	1	-	149	<0	--	0	-
32S(*)	0.13	0.10	3	3	150	<0	--	0	-
32B(*)	-	0.02	-	1	151	0.06	--	2	-
34	0.13	--	3	-	153	0.09	--	2	-
35	0.10	--	2	-	154	0.33	--	3	-
37	0.16	--	3	-	155	0.04	--	1	-
41	0.11	--	3	-	156	0.02	--	1	-
42	<0	--	0	-	157	0.02	--	1	-
43	0.03	--	1	-	158	0.01	--	1	-
44	0.04	--	1	-	159	0.07	--	2	-
45	<0	--	0	-	160	0.03	--	1	-
46	0.11	--	2	-	161	0.04	--	1	-
48	0.06	--	2	-	162	0.05	--	1	-
50	0.07	--	2	-	163	0.09	--	2	-
51	0.02	--	1	-	164	0.02	--	1	-
53	0.07	--	2	-	165	0.00	--	1	-
54	0.05	--	1	-	166	0.05	--	1	-
56S	<0	0.08	0	2	167	0.08	--	2	-
56B	--	0.05	-	2	168	0.07	--	2	-
57	<0	--	0	-	169	0.02	--	1	-
59	0.04	--	1	-	170	<0	0.19	0	3
61	0.07	--	2	-	171	0.04	0.07	1	2
62	0.06	--	2	-	172	0.07	0.02	2	1
63	0.03	--	1	-	173	0.04	0.09	1	2
65	0.06	--	2	-	174	0.06	0.05	2	2
66S	<0	0.07	0	2	175	0.02	<0	1	0
66B	--	0.07	-	2	176	<0	0.02	0	1
71	<0	--	0	-	177	0.01	0.03	1	1
73	0.45	--	3	-	178	<0	--	0	-
79	0.01	--	1	-	179	0.04	--	1	-
82	0.12	--	3	-	180	0.04	0.02	1	1
85	0.23	--	3	-	181	<0	--	0	-
95	0.04	--	1	-	182	0.09	0.02	2	1
104	0.04	--	1	-	183	0.03	0.02	1	1
108	<0	--	0	-	184	0.04	0.05	1	1
111	<0	--	0	-	185	0.05	0.07	2	2
112S	<0	0.09	0	2	186	<0	--	0	-
112B	--	0.01	-	1	187	0.18	0.03	3	1
113	0.06	--	2	-	188	0.06	0.03	2	1
115	<0	--	0	-	189	0.06	0.03	2	1
117	0.16	--	3	-	190	0.09	0.07	2	2
123	0.12	--	3	-	191	0.03	--	1	-
124	0.17	<0	3	0	192	0.09	--	2	-
131	0.14	--	3	-	193	0.10	--	3	-
132	0.10	--	3	-	194	0.03	--	1	-
133	0.20	--	3	-	195	0.10	--	3	-
134	<0	--	0	-	196	0.08	--	2	-
135	0.05	--	2	-	197	0.08	--	2	-
136	0.00	--	1	-	198	0.03	--	1	-
137	0.08	--	2	-	199	0.19	--	3	-
138	0.15	--	3	-	200	0.01	--	1	-
139	0.10	--	3	-	201	0.13	--	3	-
140	0.16	--	3	-	202	0.01	--	1	-
141	0.24	--	3	-	203	0.01	--	1	-
142	0.12	0.06	3	2	204	0.07	--	2	-
143	0.09	0.04	2	1	205	0.08	--	2	-
144	0.05	--	2	-	206	<0	--	0	-

Table 2. Continued

Station No.	Γ 1985	Γ 1986	Class 1985	Class 1986	Station No.	Γ 1985	Γ 1986	Class 1985	Class 1986
207	0.12	--	3	-	300S	-	0.04	-	1
208	0.14	--	3	-	300B	-	0.05	-	2
209	0.08	--	2	-	302S	-	0.04	-	1
210	0.09	--	2	-	302B	-	0.03	-	1
211	<0	--	0	-	303S	-	0.14	-	3
212	0.12	--	3	-	303B	-	0.05	-	2
213	0.11	--	3	-	310	-	0.04	-	1
214	0.01	--	1	-	313	-	0.05	-	2
215	0.07	--	2	-	314	-	0.07	-	2
216	0.16	--	3	-	323S	-	0.07	-	2
217	0.05	--	2	-	323B	-	0.05	-	1
218	0.10	--	2	-	324S	-	0.05	-	2
219	0.06	--	2	-	324B	-	0.11	-	3
220	0.09	--	2	-	326	-	0.16	-	3
221	0.07	--	2	-	327	-	0.00	-	1
222	0.14	--	3	-	328	-	0.00	-	1
224	0.08	--	2	-	329	-	0.00	-	1
225	0.01	--	1	-	331S	-	0.05	-	1
226	0.04	--	1	-	331B	-	0.06	-	2
227	0.19	--	3	-	332	-	0.07	-	2
228	0.06	--	2	-	343	-	0.09	-	2
229	0.11	--	3	-	352S	-	0.06	-	2
230	<0	--	0	-	352B	-	0.07	-	2
231	0.28	--	3	-	355S	-	0.06	-	2
232	0.36	--	3	-	355B	-	0.05	-	2
233	<0	--	0	-	357	-	0.06	-	2
234	0.03	--	1	-	358	-	0.22	-	3
235	<0	--	0	-	359S	-	0.16	-	3
236	0.03	--	1	-	359B	-	0.05	-	2
237	0.01	--	1	-	360	-	0.11	-	3
238	0.02	--	1	-	361	-	0.05	-	1
239	<0	--	0	-	365S	-	0.03	-	1
240	0.03	--	1	-	365B	-	0.07	-	2
241	0.02	--	1	-	367S	-	0.04	-	1
242	0.01	--	1	-	367B	-	0.05	-	2
243S	0.02	0.05	1	1	373	-	0.10	-	2
243B	--	0.05	-	2	374S	-	0.10	-	2
244	0.01	--	1	-	374B	-	0.08	-	2
245	<0	--	0	-	376S	-	0.08	-	2
246	0.15	--	3	-	376B	-	0.02	-	1
247	0.09	--	2	-	377	-	0.09	-	2
248	0.04	--	1	-	382S	-	0.08	-	2
249	0.01	--	1	-	382B	-	0.08	-	2
250	0.03	--	1	-	383S	-	0.09	-	2
251S	--	0.25	-	3	383B	-	0.08	-	2
251B	--	0.10	-	3	384	-	0.03	-	1
252S	--	0.08	-	2	385	-	0.04	-	1
252B	--	0.06	-	2	387S	-	0.05	-	2
254	--	0.14	-	3	387B	-	0.07	-	2
264	--	0.07	-	2	389	-	0.06	-	2
266S	--	0.05	-	1	390	-	0.10	-	2
266B	--	0.06	-	2	391	-	0.09	-	2
267	--	0.12	-	3	392	-	0.05	-	1
271	--	0.07	-	2	393	-	0.07	-	2
273	--	0.05	-	2	394	-	0.07	-	2
275	--	0.19	-	3	395S	-	0.07	-	2
290S	--	0.07	-	2	395B	-	0.06	-	2
290B	--	0.01	-	1	401S	-	0.09	-	2
293	--	0.19	-	3	401B	-	0.09	-	2
296	--	0.00	-	1	402S	-	0.02	-	1
297	--	<0	-	0	402B	-	0.04	-	1
298	--	0.04	-	1	404	-	0.03	-	1

Table 2. Continued

Station No.	Γ 1985	Γ 1986	Class 1985	Class 1986	Station No.	Γ 1985	Γ 1986	Class 1985	Class 1986
406	--	0.02	-	1	526	-	<0	-	0
413S	--	0.04	-	1	527	-	0.03	-	1
413B	--	0.04	-	1	529	-	<0	-	0
420	--	0.03	-	1	530S	-	<0	-	0
421S	--	0.05	-	1	530B	-	0.02	-	1
421B	--	0.02	-	1	531	-	<0	-	0
430	--	0.01	-	1	534	-	0.03	-	1
431	--	0.04	-	1	535	-	0.03	-	1
432	--	0.01	-	1	537	-	0.05	-	1
451	--	0.05	-	1	538S	-	<0	-	0
454	--	<0	-	0	538B	-	0.00	-	1
455	--	0.10	-	2	539S	-	0.01	-	1
456S	--	0.00	-	1	539B	-	0.04	-	1
456B	--	0.05	-	1	540	-	0.01	-	1
458S	--	<0	-	0	541	-	<0	-	0
458B	--	<0	-	0	553S	-	0.01	-	1
460	--	0.03	-	1	553B	-	0.07	-	2
461S	--	0.04	-	1	555S	-	0.11	-	3
461B	--	0	-	0	555B	-	0.00	-	1
463S	--	0.05	-	1	557S	-	0.01	-	1
463B	--	0.01	-	1	557B	-	0.02	-	1
479S	--	<0	-	0	559S	-	<0	-	0
479B	--	0.11	-	3	559B	-	0.01	-	1
481S	--	0.01	-	1	561S	-	0.05	-	2
481B	--	0.03	-	1	561B	-	0.00	-	1
484	--	0.08	-	2					
485	--	0.02	-	1	GMTime(day,month) [1986]				
488S	--	0.10	-	3					
488B	--	0.04	-	1	6E100S	1300(27,6)	0.04	1	
490S	--	0.08	-	2	6E100S	1900(27,6)	0.03	1	
490B	--	0.07	-	2	6E100S	2100(27,6)	0.00	1	
491S	--	0.01	-	1	6E100S	0100(28,6)	0.07	2	
491B	--	0.02	-	1	6E100S	0900(28,6)	0.02	1	
492S	--	0.04	-	1	6E100S	1100(28,6)	0.05	1	
492B	--	0.09	-	2	6E100B	1330(27,6)	0.03	1	
494S	--	0.07	-	2	6E100B	1930(27,6)	0.03	1	
494B	--	0.03	-	1	6E100B	2130(27,6)	0.00	1	
496S	--	0.01	-	1	6E100B	0130(28,6)	0.03	1	
496B	--	<0	-	0	6E100B	0930(28.6)	0.03	1	
497	--	0.04	-	1	6E100B	1130(28,6)	0.02	1	
498	--	<0	-	0					
499	--	0.13	-	3	6E300S	2200(28.6)	<0	0	
501	--	0.08	-	2	6E300S	1600(29,6)	<0	0	
504S	--	<0	-	0	6E300S	1800(29,6)	<0	0	
504B	--	<0	-	0	6E300S	2000(29,6)	<0	0	
506	--	<0	-	0	6E300B	2230(28,6)	<0	0	
507	--	0.01	-	1	6E300B	1630(29,6)	<0	0	
510S	--	0.00	-	1	6E300B	1830(29,6)	0.02	1	
510B	--	0.00	-	1	6E300B	2030(29,6)	<0	0	
512	--	<0	-	0					
514	--	0.07	-	2	6E400S	0500(02,7)	<0	0	
517	--	0.03	-	1	6E400B	0530(02,7)	0.01	1	

(*) Code: B: bottom, S: surface; samples not coded are surface water samples.

Figure 1 gives an overview of the St. Lawrence River with the sample locations and test results shown in the enlargements given in Figure 2. This figure shows the test results on the basis of the classification scheme given in Table 1. For the purpose of this figure, the data have been arranged to show classes 0 and 1 as one category and classes 2 and 3 as another. Also, sample classes were averaged between 1985 and 1986 results, where both determined, and between closely adjacent stations. Furthermore, for stations were both surface and bottom waters were tested, Figure 2 gives the surface water tests only. As evident from the data enumeration in Table 1, there is very little effect on the sample distribution from this selection.

Due to the length of the river, the graphic representation of the data requires a breakup into smaller areas, as shown in Figure 1. These areas cover stretches with known and suspected larger municipal and industrial effluent outfalls to the St. Lawrence River. Higher densities of samples were collected in those areas. In particular, such zones are in the Brockville/Prescott area (Fig. 2a), the Cornwall/Massena area (Fig. 2b), the Ottawa River/Lac des Deux Montagnes area (Fig. 2c), the Lac St. Louis area (Fig. 2d), the Montreal Harbour area (Fig. 2e), the river stretch near Sorel (Fig. 2f), the Lac St. Pierre (Fig. 2g) and the Trois-Rivieres (Fig. 2h) areas.

DISCUSSION

Over the last few years, the Microtox toxicity bioassay with *Photobacterium phosphoreum* has become widely used for the quick determination of the acute toxic effects of effluents on aquatic organisms. Although this bacterium is of marine origin, its use in freshwater systems poses no problem through the simple addition of NaCl to the sample for osmotic protection of the organisms. We have previously used this bioassay as part of multidisciplinary research studies on Lake St. Clair (Kaiser et al., 1988) and the Detroit River (Ribo et al., 1985). The experience gained from these surveys indicates that plumes and zones of toxic effects from land-based sources can be detected in receiving waters with this bioassay. Although individual samples may be difficult to interpret, repetitive and/or grid sampling will delineate such zones. For example, in Lake St. Clair, zones of class 2 and 3 Γ values were found in the nearshore water of much of the lake, while offshore water showed mostly class 0 and 1 values.

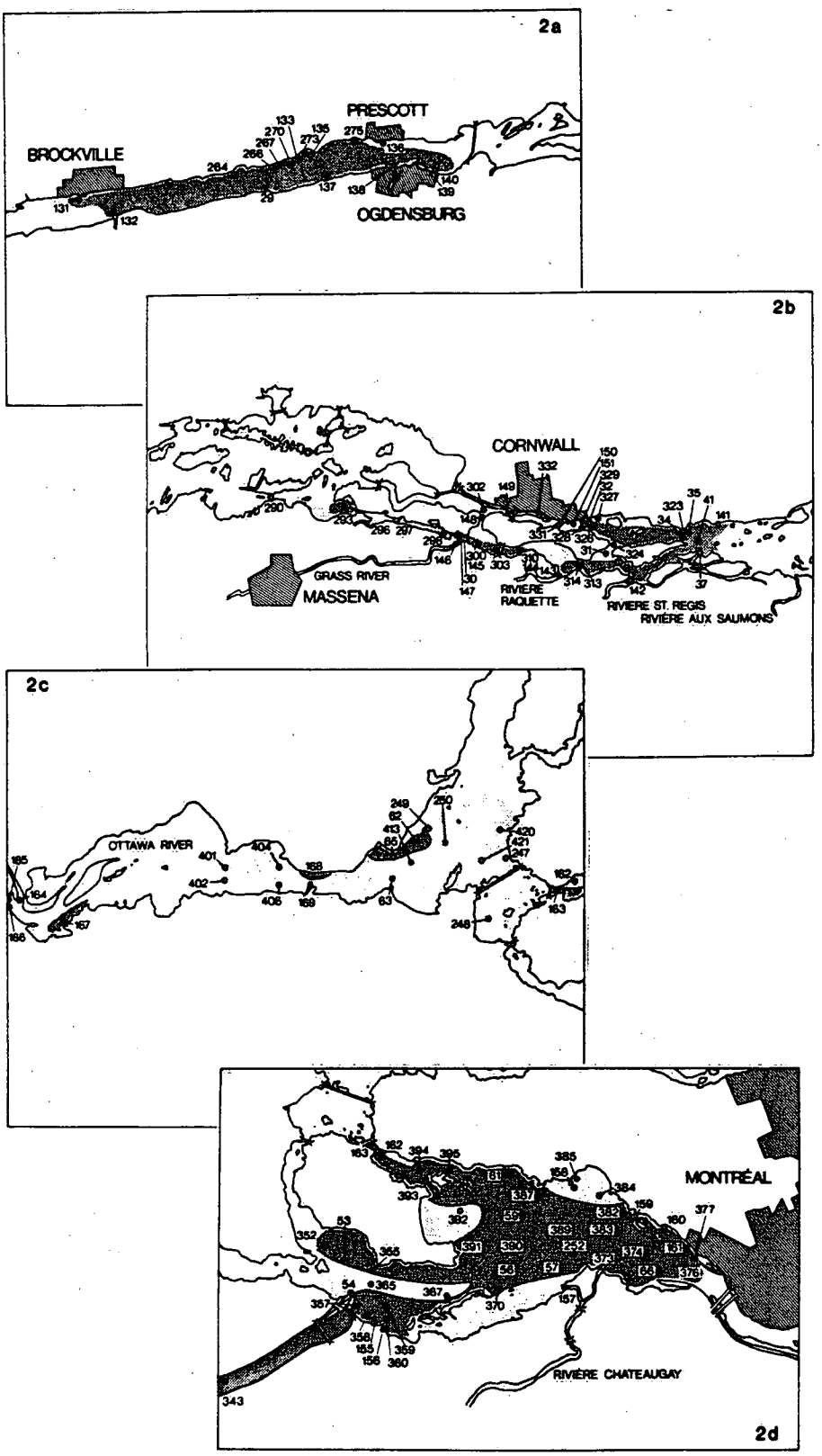
In fast flowing rivers of the size of the Detroit and St. Lawrence Rivers, with discharges of approximately 6,000 and 13,000 m^3sec^{-1}, respectively, any toxic effects attributable to point sources will be difficult to observe due to both the large dilution factor and the difficulty in positioning and sampling relative to very narrow and possibly intermittent and/or meandering effluent plumes. Consequently, sample values can easily change from one observation to another, whether separated in time or in space, even by relatively small distances.

St. Lawrence River

As evident from the data given in Table 2, which are selectively shown in Figure 2, areas of elevated Γ values are mainly found at or below locations with higher industrial and municipal densities along the river. Exceptions to these observations appear to be the stations 27 (in the north channel), 28 (in the south channel), and 29 (midriver station) near Wolfe Island at the head of the river, where Γ values ranged from 0.07 to 0.29 in both surveys. The comparatively high values for these stations appear inexplicable at this time but similarly high Γ values were found in the Brockville to Prescott area (Fig. 2a).

In the Cornwall/Massena area (Fig. 2b), the river flow is parted by islands into several channels. In addition, a number of tributaries enter at close intervals on the south shore below Massena, namely the Grass, Raquette and St. Regis Rivers. Γ vales of up to 0.19 were found at several stations in the north and south channels in that area in 1986. Measurements of volatile halocarbons undertaken during the same surveys at the mouths of these tributaries recorded values as high as 190,000 $ng.L^{-1}$ of tetrachloroethylene (Lum and Kaiser, 1986).

In Lac St. Francois, the riverine lake just downstream of the Cornwall/Massena area (not shown in Fig. 2), Γ values were generally below 0.10 in 1985. Two stations with class 3 level responses were at station 254 in the shipping channel near the eastern end of the lake and off the mouth of the Riviere au Baudet tributary near the Ontario/Quebec border. Other stations in the eastern part of the lake had mostly class 2 levels.


2a
PRESCOTT
BROCKVILLE
OGDENSBURG
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CORNWALL
GRASS RIVER
MASSENA
RIVIERE RAQUETTE
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RIVIERE AUX SAUMONS
2c
OTTAWA RIVER
MONTREAL
RIVIERE CHATEAUGAY
2d

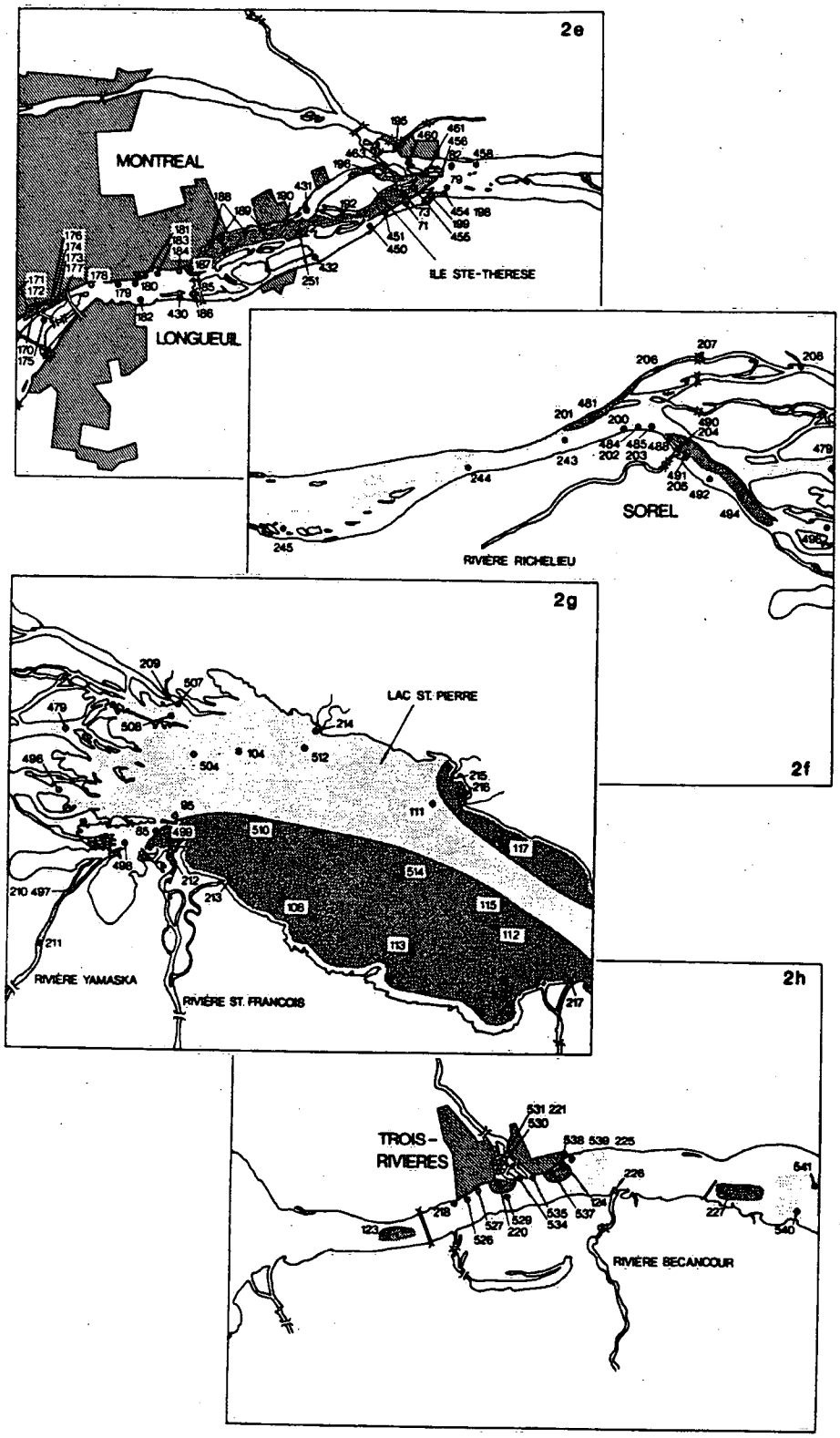

FIGURE 2. Presence and absence of agents and/or conditions in St. Lawrence River water samples causing low acute toxicity to *Photobacterium phosphoreum*; see Table 1 for definition of classes and Fig. 1 for locations.

In Lac St. Louis (Fig. 2c) and the Ottawa River/Lac des Deux-Montagnes (Fig. 2d) areas, all values were below $\Gamma = 0.09$ in both years, however, most of the samples were collected in 1985. At stations further downstream, in the Montreal Harbour (Fig. 2e) area to near Repentigny, several values were above 0.10, including stations 170, 187, 193, 195, 199, and 73. At station 73, just off Ile Sainte Therese, the value of $\Gamma = 0.45$ (in 1985) was the highest recorded in both surveys. This area received partially treated municipal waste effluents from Montreal at that time.

In the area of Sorel (Fig. 2f), class 2 levels were observed near and downstream of the mouth of the Richelieu River. Class 3 level responses were found at several stations along the northern shore of the St. Lawrence River. In Lac St. Pierre (Fig. 2g), the highest value of $\Gamma = 0.23$ was observed at station 85 at the mouth of the Yamaska River tributary in 1985. Elevated levels were also found at stations 117 and 216 off the mouths of the Yamachiche Rivers and at station 213 off the Riviere St. Francois. Further downstream, in the Trois-Rivieres to Quebec City reach of the St. Lawrence River, high Γ values were found at the mouths of Riviere Portneuf and Riviere Jacques-Cartier (Fig. 2h). At Quebec City (not shown in Fig. 2), all except one station showed very low toxicity with values in the class 0 and 1 ranges.

On average, the 1985 samples showed somewhat higher Γ values than those of 1986. This is particularly evident from the much higher proportion of samples in class 3, namely 25% in 1985, compared to 12% (surface water only) in 1986 (Table 1). Moreover, it appears that the higher proportion of class 3 samples in 1985 is mainly due to a lower proportion (32%) of class 1 samples in 1985, compared to the 43% found in 1986.

Interpretation of these findings must consider any variations in sampling areas. In both years, the sampling covered the river stretch between Lake Ontario and Quebec City, with emphasis on the upper reaches in 1985. In the following year, some of the same stations were resampled and many additional stations along the river, particularly in the lower river reaches were added. If one selects the toxicity data obtained in 1986 for only those stations which were covered in 1985, the samples are distributed as follows: Class 0: 3 stations (6%), class 1: 15 stations (44%), class 2: 13 stations (38%), and class 3: 4 stations (12%), respectively. The distribution of these data is quite similar to that found for all 1986 data. It can therefore be concluded that there is a significant difference between the toxicity levels observed in the two sampling periods. Contributing to this difference could be one or more of the following reasons: (i) Reduced flow in the St. Lawrence River which reaches its annual maximum in April/May and usually declines from June/July to September/October by approximately 10% (Pocklington and Tan, 1987), (ii) stations selected in 1985 may be less evenly distributed and more representative of onshore discharges than those in 1986, and (iii), colder water temperature in October (1985) versus June (1986) results in slower degradation, adsorption and volatilisation of toxic substances entering the river.

It is obvious that a 10% decrease in flow alone could not account for the observed increase in toxicity values. Also, when considering the station locations as shown in Figure 1, there is no reason to suspect any bias in sampling locations between the two sampling periods. Therefore, both potential reasons (i) and (ii), above, do not appear to be applicable here. In contrast, the water temperature reaches up to 23 °C in July and drops to approximately 8 °C by September or October. Coinciding with that decrease in temperature is an increase in dissolved organic carbon from June/July means of 3.6 to October/November means of 4.3 $mg.L^{-1}$, found by Pocklington and Tan (1987) for the 1981 to 1985 period. They found also that the total load of organic carbon (particulate and dissolved) increases within the river from 1.0×10^6 t/a at the outflow of Lake Ontario to some 1.8×10^6 t/a at Quebec City. In the summer months, this increase stems from the primary production in the river in contrast to fall, when it is primarily of terrigenous origin (Pocklington and Tan, 1987). Much of this allochtonous material is likely to be leaf litter and other wood products of the hardwood forests within the St. Lawrence River watershed. Obviously, this decaying litter will release a significant amount of lignin-derived products, such as phenolics and tannins. These types of compounds have been shown to exhibit strong toxic effects to *Photobacterium phosphoreum* (Konishi et al., 1987; Ribo and Kaiser, 1983) and we feel that this allochtonous material is likely to be a contributing factor for the higher number of class 2 and 3 samples observed in the fall of 1985 compared to summer 1986.

Also given in Table 2 are data for three stations in the upper St. Lawrence River estuary, namely 6E100 (just east of Isle d'Orleans), 6E300 (off Kamouraska), and 6E400 (at the mouth of the Saguenay River). At these stations, the observed Γ values are generally close to zero. For station 6E100, which is near Quebec City, a difference is noted between the surface and bottom water samples. At this station, the bottom water is strongly influenced

by the salt water intrusion at times of flood tide. The predominantly negative Γ values for stations 6E300 and 6E400 indicate a stimulating effect of these samples on the bacterium's metabolism; probably a result of the relatively unpolluted, nutrient-rich marine water mixed in at these stations.

General observations

As mentioned above, the Microtox test is designed to measure the acute toxicity of effluents in a fast and efficent way. Its applicability and usefulness in aquatic freshwater systems of generally high water quality requires operation of the instrument close to its detection limit with the consequent possibility for erroneous data due to experimental variations. While the occurence of such problems cannot be excluded entirely, a survey involving hundreds of samples, analyzed under identical conditions, provides the necessary statistical basis for a reliable interpretation of the results. We have now used this bioassay for initial studies on one large lake (Lake St. Clair) and on two major North American rivers (Detroit River and St.Lawrence River) with a total of approximately 500 water samples analyzed over three years.

One goal of these investigations was to determine whether or not the Microtox test could be considered a useful tool for the rapid screening of the water quality of these waters. In addition, it was anticipated that plumes of toxic effluents from municipal or industrial point sources could be observed in the receiving waters and that this test would therefore provide initial clues as to the need for further, more detailed investigations of particular areas.

While the majority of the total of 496 samples from the three areas in the Great Lakes basin had Gamma values between 0 and 0.5, there were some 20% with $\Gamma < 0$ and another 1% with $\Gamma > 1.0$. The latter few samples proved to be highly toxic effluents and leachates not expected before the investigation (Ribo et al., 1985). Moreover, many of the samples with $0 < \Gamma < 0.5$ values indicated plumes of effluents which either were already known to exist or recognized through these observations. Consequently, the results obtained were useful in a variety of aspects in these multidisciplinary investigations. As mentioned before, correlations of the results with concentrations of several contaminant groups, including volatile halocarbons, organochlorine compounds, polychlorinated biphenyls and polynuclear aromatic hydrocarbons proved to be of limited success only (Ribo et al., 1985). Of the contaminant groups investigated, the best correlation with the Microtox values appeared to be with chlorinated phenols; however no other phenols were measured.

Recently, Dutka et al. (1986, 1987, 1988) have embarked on countrywide surveys of natural waters in Canada with the Microtox and other bioassays. These authors use routinely a ten-fold concentration for the investigated water samples through flash evaporation at 45 °C. Ribo et al. (1985) in their study of Detroit River water samples have used a vacuum distillation method with a rotary evaporator for concentrating the samples at 40 °C and have found that the toxicity of samples containing significant amounts of phenols (ug/L levels) is greatly reduced after distillation while the distillates show increased toxicity. As phenols are frequent constituents of industrial and municipal wastewaters which are easily lost by that sample concentration method, no such concentration was undertaken in the work reported here. Moreover, unless the loss of such volatile materials is of no consquence or the toxicity of the distillates is also determined,this method of concentrating samples appears inadvisable.

Although not quite as "portable" as a typewriter or small portable computer, we have successfully used the instrument and associated recorder in motel rooms and onboard the research vessel CSS *Limnos*. With some additional precautions against vibration interference, it could certainly be used in a mobile laboratory as well. Under very humid conditions, however, excessive condensation may interfere with the operation of the instrument.

As the toxicity of a sample can be masked, at least partly, by the presence of compounds stimulating the bacteria's metabolism, hence also light emission, low or negative Γ values do not necessarily indicate the absence of any toxic substances. Therefore, one should not rely on the results of a few isolated "clean" samples. However, given a good spread of samples, spatially and/or temporalily, it appears that this bioassay can help to define zones of impaired water quality in a comparatively quick and easy fashion. It is therefore recommended as one of a variety of tools for such initial investigations. In addition, it may be useful for the delineation of effluent mixing zones in receiving waters where other tests are more time and/or resource consuming.

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