

Environmental Pollution 104 (1999) 401-410

ENVIRONMENTAL POLLUTION

Aquatic bryophytes for a spatio-temporal monitoring of the water pollution of the rivers Meuse and Sambre (Belgium)

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Received 5 February 1998; received in revised form 18 September 1998; accepted 18 September 1998

Abstract

Jaccard's distances between the aquatic bryophyte assemblages recorded in 1972–1973 and in 1997 in nine study sites along the Belgian course of the rivers Meuse and Sambre were correlated with the differences of concentrations of 13 physico-chemical factors $(O_2, N-NO_2^-, N-NH_4^+, N-NO_3^-, P-PO_4^{3-}, K^+, pH, Ca^{++}, Fe, Cu, Pb, Zn and Mn)$ between 1972–1973 and 1996. The strong decrease in heavy metals pollution and the decrease of the trophic level are correlated with the recolonization of the most polluted sites in the 1970s by the pollution-sensitive species present in the valleys at the beginning of the century. Conversely, N-NO₂⁻, N-NO₃⁻ and K⁺ have increased between the two surveys. They currently reach much higher concentrations in the Sambre than in the Meuse, as well as N and micro-pollutants such as pyrene and anionic detergents. The different loads of these factors in the studied rivers might explain their different recolonization patterns. Although some species might have developed a tolerance to certain factors, an effort to improve the quality of the effluents coming from industrial areas still remains to be made, especially in the frame of the projects of river flooding of the riverine areas. \bigcirc 1999 Elsevier Science Ltd. All rights reserved.

1. Introduction

The large European rivers have progressively been regulated to provide protection from flooding, to produce hydroelectricity and to facilitate the movement of shipping. Their alluvial floodplains have been strongly industrialized. However, this industrialization was undertaken in total disregard of ecological consequences. As a result, water quality very quickly decreased, giving rise to spectacular ecological disasters such as the pollution of the Rhine by xenobiotic factors following the fire of the chemical industry Sandoz in 1986. These ecological disasters revealed the vulnerability of large rivers. Several international agreements for improving water quality, such as the Rhine Action Program between Germany, France, The Netherlands, Luxembourg and Switzerland, were signed. Water quality has consequently improved for about 15 years (Hellmann, 1994; Beurksens et al., 1994; Malle, 1996; Tittizer and Krebs, 1996). Exactly as the recolonization of cryptogamic epiphytes thanks to the decrease of sulphur dioxide concentrations (Gilbert, 1992; Sjögren, 1995), aquatic macrophytes have been reacting very closely to these

2. Methods and study area

The Belgian part of the river Meuse has a length of 153 km. The river comes from the French part of the Ardennes and passes through the cities of Dinant, Namur, from where its banks become industrialized,

changes of water quality (Holmes and Whitton, 1977; Kohler and Schiele, 1985). Aquatic bryophytes proved to integrate in time the pollutions of different origins and were combined in efficient methods of bioindication at basin scale (Descy and Empain, 1981). A survey of the pollution level of the rivers Meuse and Sambre in Belgium using aquatic bryophytes was undertaken in the 1970s (Empain, 1973, 1977). This survey will be compared with recent bryological and physico-chemical data in this paper to answer the following questions: to what extent have the changes of water quality caused a change in aquatic bryophyte assemblages, what are the physicochemical factors best explaining these changes and what are the species concerned, what are the physico-chemical factors currently explaining the segregation of the floristic assemblages, and to what extent can aquatic bryophytes be used for long-term monitoring of water quality?

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and Liège, which is the third biggest city of Belgium (Fig. 1(a)). Its mean annual discharge at the beginning of its Belgian course is $131 \text{ m}^3/\text{s}$. The Meuse receives the waters of the river Sambre in Namur and the waters of the river Ourthe in Liège. In Visé, near The Netherlands,

its mean annual discharge reaches $314 \text{ m}^3/\text{s}$. The river Sambre is the largest tributary of the Meuse, having a mean annual discharge of $25.5 \text{ m}^3/\text{s}$ in Namur. It crosses several large industrial areas that have more or less declined for 30 years such as that of Maubeuge in



Fig. 1. (a) The study area. (b) The Walloon basin of the river Meuse.

 Table 1

 Comparison of the minima, mean and maxima concentrations of the physico-chemical factors between 1972–1973 (on the left) and 1996 (on the right)

	% sat. O ₂		[N-NO ₂] µg/l		$[N-NH_4^+] \mu g/l$	
1	91-94.3-104	86-103-153	17-22-29	12-25-41	0-60-280	< 10-50-120
2	91-97.9-103	81-107-144	22210-18-33	12-26-60	0-95-340	10-70-260
3	86-94.5-102	52-95131	27-61-185	36-86-199	0-810-2160	140-460-860
4	70-90.4-105	63-91-113	63-70-154	36-88-544	310-980-2400	170-350-630
5	50-78.1-100	38-77-109	33-85-151	40-203-1134	600-1420-2800	200-750-1370
6	30-64.7-99	45-80-110	38-92-161	41-181-602	340-1530-2800	200-820-1850
7	26-55-93	56-78-98	105-393-1400	86-288-506	960-6550-19600	800-3200-3400
8	0-48.5-100	24-54-93	64-435-1530	112-451-1120	980-4970-8960	1200-2980-5430
9	0.8-55-92	56-68-84	90-347-660	150-328-509	1170-5880-12260	1640-3070-4310
-						
	[N-NO ₃] mg/l		$[P\text{-}PO_4{}^3]\;\mu g/l$		pН	
1	0.84-1.74-2.9	0.25-2-3.9	11-139-343	10-70-110	7.8-7.85-7.9	7.89-8.22-8.56
2	0.6-1.6-2.8	0.65-2.25-4.03	12-116.5-215	10-60-130	7.7-7.9-8.3	7.03-8.09-8.73
3	0.8-1.61-3.1	1.14-2.55-4.48	19-146-241	30-100-180	7.7-7.9-8.1	7.47-7.84-8.57
4	1-1.77-3	1.61-2.98-4.63	41-743-1435	60-390-970	7.4-7.6-7.9	7.35-7.81-8.32
5	0.7-1.86-3.2	1.74-2.9-4.37	82-992-2508	110-400-820	7.2-7.4-7.7	7.24-7.56-8.17
6	0.1-1.77-3.3	0.86-2.84-4.57	82-800-2244	120-330-860	7.2-7.4-7.7	7.13-7.52-8.08
7	0.2-1.52-3.2	1.04-3.56-6.43	42-362-528	180-420-740	7.5-7.8-8.1	7.12-7.47-7.71
8	0.4-2.08-4.2	< 0.02-3.19-6.59	28-292-469	210-430-720	7.2-7.7-7.9	7.27-7.57-8.18
9	0-1.85-3.7	2.06-3.49-5.4	50-348-538	290-450-640	7.5-7.8-8	7.23-7.47-7.81
	[Ca ^{+ +}] mg/l		[Fe] $\mu g/l$		[K ⁺] mg/l	
1	55-82-117	53 4-68 9-78 1	95-215-239	114-286-805	2 2-2 9-4	2 2-3-4 1
2	46-74-101	54 6-66 4-74 1	0-230-500	103-294-834	0.7-2.5-5.1	2 2-2 9-3 5
3	55-86-117	57 7-73 2-91 6	125-285-640	134-351-1172	1 1-4-6 7	3-4 1-5 8
4	55-90 3-117	59 5-76 2-89 8	123 203 040	133-330-794	1.1 4 0.7	3-4 2-5 8
5	45-74 5-101	55 7-67 7-81 9	189-582-1140	122-287-884	1 3-3 5-7 3	3-4 4-5 9
6	50-75 4-104	56 3-68 7-85 1	280-553-854	59-366-1285	1.3 3.9 7.5	3 1-4 6-6 7
7	65-146-217	67 2-112 3-145 4	130-652-1620	211-501-1208	3 1-9 9-21 8	6 1-10 1-14 2
8	54-88 6-106	73 6-117-161 6	705-1318-2005	310-557-1270	26-6 5-9 7	6 1-8 8-10 9
9	72-147-216	84.2-122.8-185.4	141-712-1592	242-449-661	3.1-9.45-20.9	8.2-10.6-13
	[Cu] ug/l		[Mn] ug/1		[Pb] ug/l	
			[[11]] µg/1		[10] µg/1	
1	0-5.5-13	1-1.8-2.8	28-68-170	18-52-93	0-28.6-44	1.4-3.9-13.2
2	0-11.6-65	1.2-2.4-3.7	12-63-130	16-56-111	0-29-55	< 0.5-1.7-4.2
3	0-8.1-17	1.5-2.8-6.5	18-92-139	38-77-122	3-42.3-89	1.2-3.9-8.2
4	3-15-37	2-4.1-10.8	77-124-198	31-67-93	40-66-84	2.6-11.2-20.8
5	6-16-52	2.7-4.4-6.9	66-195-358	41-68-96	32-68-118	3.5-9.4-50.5
6	3-13.5-37	2.2-5.3-15.6	110-191-312	35-97-320	22-62.6-118	0.8-5.3-20.6
7	3-15-42	1.8-4-9.9	176-265-382	113-199-279	43-117-305	3.7-8.2-24.4
8	12-24.8-62	3.6-4.9-8.1	188-258-436	111-181-304	96-229-506	5.8-12.2-34.6
9	5-22-48	1.9-4-5.7	228-305-382	126-175-231	53-129-251	3-6.2-9.7
	$[Zn] \; \mu g/l$		[cyanures] (mg/l)		[As] (mg/l)	
1	0-38-66	< 25-41-69	_	0.002-<0.002-0.002	_	< 0.2-0.4-0.8
2	0-22-60	< 25- < 25-30	_	0.002-<0.002-0.002	_	< 0.2-0.5-1.1
3	0-76-327	< 25-27-55	_	0.002-<0.002-0.002	_	< 0.2-0.6-1.2
4	193-472-788	35-75-297	_	_	_	0.3-0.7-1.2
5	200-506-1859	32-54-109	_	0.002-0.013-0.03	_	0.3-0.8-1.8
6	98-308-610	< 25-52-144	_	< 0.002-0.007-0.019	_	0.3-0.9-2.7
7	42-467-994	39-72-143	_	< 0.002-0.01-0.037	_	0.4-1.4-2
8	235-758-1300	44-94-198	_	0.002-0.033-0.096	_	0.3-1.3-1.9
9	71-543-1027	39-59-97	_	_	_	0.3-1.2-1.5
	[anionic degerge	ents] (ng/l)	[phenol] (mg/l)		[pyrene] (ng/l)	
1		< 0.012.0.027.0.042	/	0.02.0.000.0.027	=- /	5 11 21
1	—	< 0.012-0.027-0.045	-	0.02-0.009-0.02/	_	D-11-51
∠ 2	—	< 0.012-0.021-0.041	-	< 0.002-0.01-0.035	_	3-10-31
3	—	\0.012-0.027-0.05	-	< 0.002-0.01-0.035 0.002-0.011-0.021	_	3-10-32
4	-	0.029-0.046-0.103	-	0.003-0.011-0.031	-	_

	[anionic degers	gents] (ng/l)	[phenol] (mg	/l)	[pyrene] (ng/l)						
5	_	0.023-0.046-0.074	_	0.004-0.017-0.049	_	5-20-46					
6	-	0.024-0.048-0.081	_	0.004-0.013-0.031	_	1-14-61					
7	-	0.045-0.086-0.129	_	0.002-0.014-0.058	_	3-35-160					
8	-	0.05-0.113-0.198	_	0.006-0.022-0.093	_	13-101-241					
9	_	0.085-0.107-0.122	_	0.006-0.013-0.028	_	_					
	[atrazine] (µg/l)	[hexachloroc	yclohexane] (ng/l)							
1	_	0.04-0.135-0.466	_	2-7-16							
2	_	0.034-0.151-0.553	_	3-8-19							
3	_	0.029-0.277-1.571	_	3-14-40							
4	_	_	_	_							
5	_	0.029-0.177-0.739	_	2-19-99							
6	_	0.021-0.208-0.670	_	5-7-60							
7	_	0.034-0.235-1.193	_	9-23-86							
8	_	0.048-0.452-2.43	_	3-16-45							
9	_	-	_	-							

Table 1 (Continued)

1: Meuse in Anseremme; 2: Meuse in La Plante; 3: Meuse in Andenne; 4: Meuse in Ivoz-Ramet; 5: Meuse at lle Monsin; 6: Meuse in Visé; 7: Sambre in Namur; 8: Sambre in Charleroi; 9: Sambre in Floreffe; – no data available.

France and that of Charleroi in Belgium. Being intensively navigated, both rivers present a dense network of weirs and sluices regulating the water level.

The nine weirs studied by Descy and Empain (1976) and Empain (1977) from both bryological and physico-chemical points of view in the rivers Sambre and Meuse (12 repetitions of the measures of the physico-chemical factors in 1972–1973) and where a physico-chemical survey of water quality by the Walloon Region was available (13 repetitions of the physico-chemical factors in 1996), were bryologically surveyed in 1997 (Fig. 1(b)). The aquatic bryophytes (nomenclature of Corley et al., 1981Corley and Crundwell, 1991) were recorded by using a coefficient of presence–absence. Only the species occurring in more than 30% of the sites were retained in the analysis. The collections of Empain (1977) were checked in order to get a homogeneous floristic data set, i.e. to ensure that the taxonomic conceptions in difficult genera such as *Amblys*-*tegium*, in which intermediate specimens between *fluviatile* and *tenax* occur, are the same. To assess the floristic changes between the 1970s and the 1990s, the variation of occurrence of each species, i.e. the sum of its occurrences in the 1990s divided by the sum of its occurrences in the 1970s, was calculated as well as Jaccard's coefficient J between the floristic assemblages of each of the *i* sites (i = 1, ..., 9) surveyed in the 1970s and in the 1990s:

$$J_i = P_i / (P_i + N_i)$$

where P_i is the number of species in common in the site *i* in the 1970s and in the 1990s, and N_i is the number of species recorded in the course of one of the survey and not during the other one in the site *i*.

Table 2

Comparison of the bryophyte assemblages between 1972-1973 (on the left) and 1997 (on the right)

	1		2		3		4		5		6		7		8		9		f
Amblystegium fluviatile	0	1	0	1	0	1	0	1	0	1	0	0	0	0	0	1	0	0	n
Amblystegium humile	0	1	0	1	0	0	0	1	0	1	0	0	0	0	0	0	0	0	n
Amblystegium riparium	1	1	1	1	1	1	1	1	1	1	1	1	1	1	0	1	1	1	1.1
Amblystegium tenax	1	1	0	1	0	1	0	1	0	1	0	0	0	1	0	0	0	1	7
Cinclidotus danubicus	1	1	1	1	1	1	1	1	1	1	0	1	0	0	0	0	0	0	1.2
Cinclidotus riparius	1	1	1	1	1	1	1	1	1	1	1	1	0	0	0	0	0	0	1
Cratoneuron filicinum	0	1	0	1	0	1	0	0	1	0	0	1	0	0	0	0	0	0	4
Fissidens crassipes	1	1	1	1	0	1	0	1	0	1	0	0	0	1	0	0	0	0	3
Fontinalis antipyretica	1	1	1	1	1	1	0	0	0	0	0	0	0	0	0	0	0	0	1
Hygrohypnum luridum	0	1	0	1	1	1	0	0	0	0	0	0	0	0	0	0	0	0	3
Octodiceras fontanum	1	1	1	1	1	1	0	0	0	0	0	0	0	0	0	0	0	0	1
Rhynchostegium riparioides	1	1	1	1	1	1	0	0	0	0	0	0	0	1	0	1	0	1	2
J	0.6	6	0.5	8	0.6	53	0.4	2	0.4	2	0.5	50	0.2	25	0.0	00	0.	33	

1: Meuse in Anseremme; 2: Meuse in La Plante; 3: Meuse in Andenne; 4: Meuse in Ivoz-Ramet; 5: Meuse at Ile Monsin; 6: Meuse in Visé; 7: Sambre in Namur; 8: Sambre in Charleroi; 9: Sambre in Floreffe; *J*: Jaccard's coefficient; *f*: number of occurrences in 1997/ in 1972–1973; *n* = species recorded in 1997 and not in 1972–1973.

Table 3

Correlation coefficients between the differences of the mean, the minima and/or the maxima concentrations of the physico-chemical factors measured in 1972–1973 and in 1996 and Jaccard's coefficients between the floristic assemblages recorded in 1972–1973 and in 1997

Factor	J
% sat. O ₂	NS
[N-NO ₂]	NS
[N-NH4 ⁺]	0.74*
max [N-NO3-]	0.75*
$\log [P-PO_4^3]$	0.74*
[K ⁺]	-0.81^{**}
pH	NS
max [Ca ⁺⁺]	0.75*
[Fe]	0.91***
[Cu]	0.88**
[Mn]	NS
[Zn]	0.91***
[Pb]	0.95***

NS: not significant. * Significant at 0.05 level. ** Significant at 0.01 level. *** Significant at 0.001 level.

The differences between the mean concentrations of the physico-chemical factors measured in the 1970s and in the 1990s for each of the *i* sites were calculated and correlated with the Jaccard's coefficients to test whether the changes in the physico-chemistry of the waters caused a floristic change. The same operation was performed with the maxima, the logarithmic means and the logarithmic maxima concentrations of the physicochemical factors, and the variables giving the best correlation coefficients were retained. A numerical classification of the nine study sites according to their floristic assemblages in 1972-1973 and in 1997 based on Jaccard's distances between the sites and using Ward's minimum variance criterion was undertaken to assess the degree of similarity of the floristic assemblages along a temporal gradient on the one hand and along an upstream-downstream gradient on the other hand. The patterns of floristic clusters related to water chemistry were studied along the first axes of a correspondence analysis performed on the floristic data set, whose site scores were correlated with the physico-chemical factors. Due to data availability, the correlation coefficients for atrazine, pyrene, cyanures, As and anionic detergents were calculated with the site scores of the survey 1997 only.

3. Results

The mean, minima and/or maxima of the physicochemical factors measured in 1972–1973 and in 1996 are given in Table 1. The rivers have a strong buffer-capacity due to high concentrations of calcium with annual means greater than 65 mg/l, hence high pH values always greater than 7.5. The concentrations of the trophic factors ammonium and phosphates increase upstream to downstream. Ammonium, phosphates and heavy metals concentrations measured in 1973–1973 were nearly always greater than those of 1996, whereas the mean concentrations of potassium and the maxima concentrations of nitrates measured in 1996 were always greater than those measured in 1972–1973. These concentrations reach much higher values in the Sambre than in the Meuse, which is also true for N, Ca⁺⁺, heavy metals, cyanures, anionic detergents, pyrene and atrazine.

The comparison of the bryophyte data set between 1972-1973 and 1997 is presented in Table 2. Two species, Amblystegium fluviatile and A. humile, were not recorded during the survey 1972-1973. The frequency of occurrence of *Cinclidotus danubicus*, *Fissidens crassipes*, Cratoneuron filicinum, Hygrohypnum luridum, Rhynchostegium riparioides and Amblystegium tenax increased between 1972-1973 and 1996. In the Meuse, the sluice of Visé has always proved to be the site with the lowest number of species, including Amblystegium riparium, recorded in almost all the study sites in the course of both floristic surveys. In the Sambre, the area of Charleroi, without any bryophyte living in the 1970s, has been recolonized. Jaccard's coefficients have a tendency to decrease upstream to downstream, i.e. the floristic differences between the surveys 1972-1973 and 1997 increase along the course of the river. Jaccard's coefficients are significantly correlated with the differences of mean concentrations of N-NH4⁺, P-PO4³⁻, K⁺, Cu, Pb, Zn and Fe and with the differences of maxima of N- NO_3^- and Ca^{++} (Table 3). A summary of the cluster analysis of the sites based on their floristic composition is given in Fig. 2. Five groups emerge from this classification:

- in the Meuse, the sites Anseremme, La Plante, Andenne, Ivoz-Ramet and Monsin of the survey 1997, clustered at a semi-partial R-squared of 0.045 (cluster 1);
- in the Meuse, the upstream sites Anseremme, La Plante and Andenne of the survey 1972–1973 (cluster 2), clustered at a semi-partial R-squared level of 0.01, and joined with the first cluster at a semi-partial R-squared level of 0.076;
- all the sites of the Sambre of the survey 1997 (cluster 3), clustered at a semi-partial R-squared level of 0.01, and joined with the first two clusters at a semi-partial R-squared level of 0.21;
- in the Meuse, the downstream sites Ivoz-Ramet, Monsin of the survey 1972–1973 and the site Visé of both surveys (cluster 4), clustered at a semipartial R-squared level of 0.03;
- the sites Namur and Floreffe in the Sambre of the survey 1972–1973 (cluster 5), clustered at a semi-partial R-squared level of 0.00 and joined with the former cluster at a semi-partial R-squared level of 0.13.

The pattern of the floristic clusters along the first two axes of the correspondence analysis of the floristic data Name of Observation or Cluster

					_	_	•	•	0	0	0	0	0	0	0	0 B	0 B	0 B	0 B
		0	0	0	0	0	0	0	0	В	в 1	-	-	D	1	1	1	1	1
		В	В	В	В	В	В	В	В	1	1	-7		0	0	1	י 2	1	י פ
		1	3	5	7	9	2	4	6	3	5	/	0	8	0	1	2	-	0
	0.3	+																	
		l																	
		1																	
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		XX>	(XXX)	(XXX)	XXXX)	XXXX	XXXX	XXXX)	(XXX)	<b>XXX</b>	~~~~~	~~~~	\\\\ /\\\	·~~`	~~~~	~~~~	~~~~/ ~~~~	\^^^/	~~~ VVV
		XX>	(XXX)	(XXX)	XXXX	XXXX	XXXX.	XXXX)	(XXX) //////////////////////////////////	< X X X /	~~~~	~~~/ ~~~~	~~~	~~~	~~~~	~~~^ ~~~~	~~~~/ ~~~~	\^^^/	~~~ YYY
		XX>	(XXX)	(XXX)	XXXX	XXXX)	XXXX.	XXXXX	XXXX/ //////	~~~~	~~~~	~~~ <i>~</i> / ~~~~	\^^ /VV	 	~~~~. ~~~~~	~~^^ VVVV	~~~~/ YYYYY	(YYY)	<u> </u>
	0.25	+XX>	(XXX)	(XXX)	XXXX)	XXXXX	XXXX.	XXXX/ VVVV	~~~~/ ~~~~	~~~~	~~~~	~~~~/ ~~~~~	~~~ /VY	 	~~~~ YYYY	~~~~ YYYY	****	XXXX	XXX
			(XXX)	XXXX	XXXX	XXXXX	XXXX	~~~~/ ~~~~	~~~/ ~~~~	~~~~ /////	~~~~ ~~~~	~~~~/	~~~ / Y Y	XX.	XXXX.	XXXX	XXXXX	XXXX	XXX
_			(XXX)	(XXX)	XXXX	XXXX/	XXXX. 	~~~~	~~~// //////	~~~~	~~~~ ~~~~	~~~~/	~~~ ~~~	XX	YYYY	XXXX	XXXXX	XXXX	XXX
S			(XXX)	XXX.	XXXX	XXXX/	~~~~	~~~~/ ~~~~	~~~~/ ~~~~	~~~~. ~~~~	~~~~ ~~~~	~~~~/	~~~ ~~~	XX	XXXX	XXXX	XXXXX	XXXX	XXX
е			(XXX)	XXX.	XXXX	****	~~~~	~~~ <i>~</i>	~~~// ~~~~	~~~~ ~~~~	~~~~ ~~~~	~~~~/ ~~~~	~~~ ~	XX.	XXXX	XXXX	XXXXX	XXXX	XXX
m			(XXX)	XXXX.	XXXX. VVVV	~~~~	~~~~	~~~~/ ~~~~	~~~ <i>^</i>	~~~^ ¥Y	<u>, , , , , , , , , , , , , , , , , , , </u>	****	XXX	XX	XXXX	XXXX	XXXXX	XXXX	XXX
1				<b>ΚΧΧΧ</b> . Ζ <u>ννν</u> γ	~~~~	~~~ <i>~</i> / ~~~~	~~^^ ~~~~	~~~~ <i>`</i>	~~~ YYY	XX	YYYY YYYY	XXXXX	XXX	XX	XXXX	XXXX	XXXXX	XXXX	XXX
-	0.2	+XX/	(XXX)	~~~~	~~~~ ~~~~	~~~~/	~~~~ ~~~~	~~~~ <i>`</i>	~~~ YYY	YY	XXXX	XXXXX	XXX	XX	XXXX	XXXX	XXXXX	XXXX	XXX
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Fig. 2. Summary cluster diagram of the study sites according to the bryophyte assemblages using Ward's minimum variance clustering criterion. Dissimilarity, indicated on the vertical axis, is the between clusters semi-partial R-squared. OB1: Meuse in Anseremme, 1997; OB2: Meuse in Anseremme, 1972–1973; OB3: Meuse in La Plante, 1997; OB4: Meuse in La Plante, 1972–1973; OB5: Meuse in Andenne, 1997; OB6: Meuse in Andenne, 1972–1973; OB7: Meuse in Ivoz-Ramet, 1997; OB8: Meuse in Ivoz-Ramet, 1972–1973; OB9: Meuse at Ile Monsin, 1972–1973; OB11: Meuse in Visé, 1997; OB12: Meuse in Visé, 1972–1973; OB13: Sambre in Namur, 1997; OB14: Sambre in Namur, 1972–1973; OB15: Sambre in Charleroi, 1997; OB16: Sambre in Charleroi, 1972–1973; OB17: Sambre in Floreffe, 1997; OB18: Sambre in Floreffe, 1997; OB18: Sambre in Floreffe, 1972–1973.

Table 4 Correlation coefficients between the site scores of the correspondence analysis of the bryophyte data set and the chemical factors

Factor	Axis 1	Axis 2
% sat. O ₂	NS	0.81***
N-NO ₂ ⁻	0.51*	-0.64 **
N-NH4 ⁺	NS	$-0.86^{***}$
max N-NO ₃ ⁻	0.67**	NS
P-PO4 ³⁻	NS	-0.52*
K ⁺	0.61**	-0.64 * *
pH	NS	NS
max Ca ⁺⁺	NS	-0.84***
Fe	NS	-0.83***
Cu	NS	-0.76***
Mn	NS	$-0.85^{***}$
Zn	NS	-0.73***
Pb	NS	-0.81***
(2) atrazine	0.78*	NS
(2) max pyrene	0.93**	NS
(2) max cyanures	0.82**	NS
(1) As	0.67*	-0.85**
(1) anionic detergents	0.77**	-0.87**

(1), (2): correlation coefficient with the site scores of the survey 1997 only, including nine or seven sites in function of the data availability, respectively; NS: not significant.

* Significant at 0.05 level. ** Significant at 0.01 level. *** Significant at 0.001 level.

set explaining 28% and 25% of the total variance, respectively, is presented in Fig. 3(a). Along axis one, the site scores of the Sambre increased from the central part of the axis to the extreme right of the axis between the surveys 1972–1973 and 1997. Along axis two, all the site scores of the survey 1972-1973 have increased between the two surveys so that all the sites of the survey 1972-1973 are located beneath their homologues of the survey 1997. Cluster one is characterized by Amblystegium humile and A. fluviatile, cluster 2 by Fon*tinalis antipyretica* and *Fissidens crassipes*, cluster 3 by Amblystegium tenax and Rhynchostegium riparioides, cluster 4 by Cratoneuron filicinum, Cinclidotus danubicus and C. riparius, and cluster 5 by Amblystegium riparium (Fig. 3(b)). On axis one, the site scores are significantly correlated with the mean concentrations of K⁺ and N- $NO_2^-$  and with the maxima concentrations of N-NO₃⁻. The site scores of the survey 1997 are also significantly correlated with the mean concentrations of atrazine, As and anionic detergents and the maxima concentrations of pyrene and cyanures. On axis two, the site scores are significantly correlated with the mean concentrations of N-NH4⁺, P-PO4³⁻, Fe, Zn, Pb, Cu and the maxima concentrations of  $Ca^{++}$  (Table 4).

# 4. Discussion

As in the Lower Rhine (Frahm and Abst, 1993, Frahm, 1997), the diversity of the aquatic bryophyte

assemblages of the rivers Sambre and Meuse has increased during the last decades. In the rivers Sambre and Meuse, the changes in aquatic bryophyte assemblages are best correlated with the changes in heavy metals concentrations and, to a lesser extent, with the changes of the trophic level of the waters. The improvement of water quality concerning heavy metals and trophic level is shown along the second axis of the correpondence analysis of the floristic data set, whose site scores are significantly negatively correlated with these factors, and along which the scores of the same sites increased between the surveys 1972–1973 and 1997. It seems thus that heavy metals and trophic factors, that heavily polluted the Sambre and the Meuse in the 1970s, were limiting for the aquatic bryoflora that was able to spread thanks to the decrease in these factors. This is supported by recent ecophysiological experiments. It was experimentally demonstrated that the growth of Rhytidiadelphus squarrosus decreases with increasing concentrations of heavy metals. The toxicity was shown to be influenced by concentration and exposure: the toxicity sequence on shoots exposed to metal via their bases is Pb < Cu < Ni < Zn < Cd, whereas the toxicity sequence was shown to be concentration-dependant in the case of shoots exposed to metal by pulse-incubation; at 0.01 mM, Ni < Zn < Pb < Cu, whereas at 0.02 mM, Pb < Ni < Zn < Cu, with no growth possible with Cd at either concentration (Sidhu and Brown, 1996). Total metal uptake increases with the supplied concentration up to values that saturate the available anionic exchange sites external to the cell cytoplasm. Indeed, copper and lead are preferentially adsorbed (Rühling and Tyler, 1970) and might be in competition with other cations needed by the plants (Glime, 1992). It was suggested that membrane damage is one of the most significant effect of toxic metal on cryptogamic plants (Brown and Whitehead, 1986). Evidences were brought, however, that substantial reduction in photosynthesis occurs without the loss of comparable proportion of the intracellular K (Brown and Wells, 1990), and that photosynthesis also declines in proportion to intracellular Cd (Wells and Brown, 1995). Concerning the trophic level, it was suggested that high concentrations of N-NH₄⁺ could explain the growth stimulation of *Amblys*tegium riparium in eutrophic conditions (Kelly and Huntley, 1987). Conversely, it was experimentally demonstrated that Fissidens crassipes does not tolerate concentrations of N-NH4⁺ higher than 11.6 mg/l (Frahm, 1975, 1976). The N-NH₄⁺ concentrations in the Sambre currently remain under this critical level, which might be a cause of recovery of this species.

Thanks to this improvement of water quality, the study sites show more floristic affinities along a spatial gradient than along a temporal gradient. In the Meuse, the floristic assemblages presented significant differences along a longitudinal gradient in the 1970s, with *Cinclidotus* 



Fig. 3. (a) Correspondence analysis of the bryophyte data set, representation of the study sites. For explanation of the labels, see Fig. 2. (b) Correspondence analysis of the bryophyte data set, representation of the bryophyte species. Ambflu: Amblystegium fluviatile; Ambhum: Amblystegium humile; Ambrip: Amblystegium riparium; Ambten: Amblystegium tenax; Cindan: Cinclidotus danubicus; Cinrip: Cinclidotus riparius; Crafil: Cratoneuron filicinum; Fiscra: Fissidens crassipes; Fonant: Fontinalis antipyretica; Hyglur: Hygrohypnum luridum; Rhyrip: Rhynchostegium riparioides.

spp. and *Amblystegium riparium* in the downstream part of the river, and *Cinclidotus* spp., *Amblystegium riparium, Fontinalis antipyretica, Fissidens crassipes* and some Amblystegiaceae such as *Hygrohypnum luridum* or *Amblystegium tenax* in the upstream part of the river. The decrease in heavy metals and trophic level, which is especially obvious in the downstream sites, led these two different clusters to converge into a single floristic cluster including species such as *Amblystegium fluviatile, A. humile* and *Cratoneuron filicinum*, that were present in the river at the end of the last century (Delogne, 1883; Mansion and Clerbois, 1894) and strongly decreased or even disappeared from the Meuse in the 1970s. Nowadays, the longitudinal floristic gradient is thus homogeneous until Liège, even downstream from the city of Namur, at the confluence of the heavily polluted river Sambre, probably thanks to the dilution of the Sambre waters in the Meuse. Hence, the first five sites recorded in 1997 present more floristic affinities between them than with their homologues recorded in 1972–1973. Only the site Visé, located downstream from the industrial area of Liège, showed no significant temporal shift of aquatic bryophyte assemblages, which is shown by the proximity of its site scores 1972–1973 and 1997 along the first axes of the ordination and which testifies of a still limiting pollution for its bryoflora. In the river Sambre, water pollution reached such a level in the 1970s, that only *Amblystegium riparium*, which is known to be one of the most pollution-tolerant species (Hussey, 1982Kelly and Huntley, 1987Touw and Rubers, 1989), or even no species, occured. The current floristic assemblages of the Sambre, characterized by *Rhynchostegium riparioides* and *Amblystegium tenax*, show consequently less similitudes with their homologues of the 1970s than with the floristic assemblages of the river Meuse.

The site scores of the Sambre show a second temporal evolution trend, increasing between the surveys 1972– 1973 and 1997 along the first CA axis. The site scores on axis one are significantly correlated with the mean concentrations of K⁺ and with the maxima concentrations of N-NO₂⁻ and N-NO₃⁻. Exactly as in the river Rhine (Hellmann, 1994), these factors, as opposed to heavy metals and trophic level, have shown a continuous tendency to increase in the rivers Sambre and Meuse during the last decades. They currently reach much lower concentrations in the Meuse than in the Sambre, which is also much more loaded in N and micro-pollutants like pyrene, anionic detergents or atrazine. This might explain why the aquatic bryoflora of the two studied rivers has shown two different patterns of recolonization, i.e. the spread of Cratoneuron filicinum, Amblystegium fluviatile, A. humile, Hygrohypnum luridum and Cinclidotus danubicus in the river Meuse and the spread of Rhynchostegium riparioides and Amblystegium tenax in the lower Sambre, where still no Cinclidotus species occur.

The spread of a species such as *Amblystegium tenax* in the still heavily polluted lower Sambre is surprising, because this species is usually found in good quality waters (Werner, 1996Vanderpoorten et al., 1998). However, it seems to have spread in Belgium during the last decades, even in very bad quality waters, where it is present with the only pollution-tolerant *A. riparium* (Vanderpoorten, in prep.). Bryophytes proved to be able to develop pollution-tolerant ecotypes. This was already demonstrated in some species growing in sites with important heavy metals concentrations (Briggs, 1972Brown and House, 1978), and it is likely that the very polymorphic *Amblystegium tenax* presents an ecotype which is pollution-tolerant in the lower Sambre.

# 5. Conclusion

Important changes of aquatic bryophyte assemblages occured in the rivers Sambre and Meuse between the 1970s and the 1990s and are correlated with decreasing concentrations of heavy metals and trophic level. The different loads of micro-pollutants and of N between the two studied rivers might explain the different patterns of recolonization of their bryoflora. The aquatic bryoflora can efficiently be used for the spatio-temporal monitoring of water quality at basin scale, and the presented here data constitute a reference basis for future comparisons. In fact, it remains to be hoped that the process of water quality improvement will continue in the following years and will allow the success of the Belgian– Dutch ecological project "Grenzmaas" of floodings of the riverine areas in the Lower Meuse.

### Acknowledgements

This work was realised thanks to the financial support of the FRIA. Many thanks are due to D. Wylock and to the Walloon Region for providing and for authorizing the publication of the physico-chemical data of 1996, to the Navigation Office for providing authorizations of access to the sluices, to A. Empain for providing unpublished data and for valuable discussions, and to Mrs. Bock who was once more very kind to correct the English of this paper.

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