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Relationship Between Summarizing Chemical Parameters like AOX, TOC, TN_b, and Toxicity Tests for Effluents from the Chemical Production

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The effect assessment of wastewater from the industrial sector of chemical production is based on toxicity data. For the ecological risk estimation it would be valuable to be able to predict toxicity by chemical data especially evaluated by parameters with summarizing characteristics.

Therefore this study was undertaken to investigate, whether there are correlations between summarizing parameters, which potentially indicate hazardous water components, like AOX (adsorbable organic halogen), TOC (total organic carbon) and TN_b (total bound nitrogen) and biological effects, observed from different bioassays conducted in the laboratory. The toxic effects were investigated on luminescent bacteria, microcrustaceans and algae in accordance with "paragraph 7a of the German Federal Water Act". The fish test was not taken into account for this investigation because of its lack of sensitivity.

MATERIALS AND METHODS

Eighty one wastewater samples from five effluents of the chemical industry (Table 1) were collected between 1996 and 1999.

The chemical parameters were analysed in accordance with standardized analytical methods. The TOC content was determined following the German version of the European guideline EN 1484: 1997, AOX following the German version of the European guideline EN 1485: 1996, and TN_b following the German standard method DIN 38409-27: 1992.

The poisonous effects were evaluated by means of daphnids (*Daphnia magna*) according to the German Standard method DIN 38 412-30: 1989 (immobility test after 24 h), by algae (*Scenedesmus subspicatus*) according to DIN 38 412-33: 1991 (growth inhibition after 72 h), and by luminescent bacteria (*Vibrio fisheri*) according to DIN 38 412-341: 1993 (inhibition of bioluminescence after 30 min.).

The ranking of wastewater by toxicity and by analytical results was compared with the Spearman Rank Correlation Test. It is a nonparametric ranking test with correction of ties. According to Basler (1988) this test can also be applied in the case of many ties.

Table 1. Origin of wastewater samples

Effluent	Lines of production
A	Production of photographic chemicals, colours, phosphorous organic compounds, phenole derivatives, and of chemicals for plastics manufacturing
B	Production of caoutchouc, plastics, and of explosives; processing of polyvinylchloride; chlorination of aromatic compounds for fuel additives
C	Processing of mineral oil, diverse aromatic compounds; production of substances for pharmaceutical products, substances for pesticides, and of chemicals for plastics manufacturing
D	Plastics manufacturing; production of anorganic substances, and of basic materials for detergents
E	Production of acetic acid for plastics manufacturing, chlorinated hydrocarbons (as basic material), and of inorganic salts

RESULTS AND DISCUSSION

The results of chemical analysis are summarized in Table 2. The level of AOX was highest in the samples of the effluent C. The AOX values ranged from 0.66 mg/L to 1.31 mg/L, whereas effluent B showed the highest mean concentrations of TOC (71.8 mg/L), and NT_b (44.1 mg/L). The most inconspicuous results were to be found in the effluent D.

Table 2. Wastewater characterisation of the investigated chemical effluents concerning AOX, TOC and TN_b

		AOX (mg/L)			TOC (mg/L)			TN_b (mg/L)		
Ef.	N	Mean	SD	Range	Mean	SD	Range	Mean	SD	Range
A	28	0.74	0.18	0.45–1.12	42.6	8.20	25.7–58.5	36.2	7.38	21.2–50.0
B	11	0.26	0.16	0.09–0.71	71.8	35.3	24.3–153	44.1	10.1	22.7–59.1
C	19	1.05	0.14	0.66–1.31	45.5	6.37	35.6–59.1	32.4	6.92	24.2–44.5
D	10	0.08	0.17	0.01–0.54	6.05	6.68	1.44–17.1	6.46	2.75	3.42–11.3
E	13	0.61	0.39	0.04–1.37	33.3	14.2	14.6–74.7	35.3	14.4	13.3–55.8

Ef., Effluent; n, sample size; SD, standard deviation

The toxicity responses obtained with the wastes are presented in Table 3. The samples were classified in percent of wastewater, in dilution with clean water, which lose toxic effects (100% wastewater = nontoxic sample; 4.2% wastewater = most toxic sample in this study). Of the 81 samples tested, only 11 (13.6%) were found to be nontoxic, 16 samples (19.8%) were determined to be toxic by one, 30 samples (37%) by two toxicity tests and the remaining 24 samples (29.6%) exhibited toxicity to all test organisms.

All effluents displayed, at least occasionally, toxicity to all test organisms. But the degree of toxicity varied greatly between the different effluents. The highest and most frequent toxicity was recorded with samples of the effluent C (containing the highest concentration of AOX)) followed by the effluents A and D. Effluent B only occasionally displayed slight toxicity, although it showed the highest level of TOC and TN_b.

Toxicity is related to the kind of products which are synthesized and released by the industrial sites. This is the reason why no test system could be generally stated as the most sensitive for all effluents (Table 4).

Table 3. Sample classification according to the bioassays results (as wastewater concentration in clean water without hazardous properties to the used testorganisms in %) from the analyzed chemical effluents (100% = nontoxic samples; 4.2% = most toxic samples)

Waste-	Effluent														
water	A			B			C			D			E		
%	D[a]	M[b]	A[c]	D	M	A	D	M	A	D	M	A	D	M	A
100	16	2	16	4	10	9		1	2	2	7	2	7	7	10
50	11	3	7	6	1	1			2	3	1	2	4	1	1
33	1	9	1	1				1	2	5	1	1	2	2	
25		2	2				5	8	5			2			1
17		4	1				11	2	3			1			
13		5	1			1	3	5	2						1
8		2						1	2			2			
6		1						1						3	
4									1		1				

[a]Daphnids; [b]Microtox (30 min.); [c]Algae

The sensitivity ranks for the effluents in Table 4 is comparable to other studies. Effluent A was most toxic to luminescent bacteria. Vasseur & Ferard (1984) also observed that discharge from factories synthesizing colours and phenols are more toxic to luminescent bacteria than to daphnids. In accordance with the data set of Munkittrick & Power (1991) pharmaceutical factories produce wastes (like effluent C), which are more toxic to daphnids than to luminescent bacteria. Effluent D, containing wastes from the production of detergents, is most toxic to algae. This corresponds to investigations of Zimmermann (1992).

Table 4. Sensitivity ranking of the applicated bioassays according to their sensitivity frequency (number of times a bioassay was most sensitive) for each effluent

Effluent	Rank				
A	Microtox	>	Algae	>	Daphnids
B	Daphnids	>	Algae	=	Microtox
C	Daphnids	>	Microtox	>	Algae
D	Algae	>	Microtox	>	Daphnids
E	Microtox	>	Daphnids	>	Algae

Wastewater samples are mixtures of organic and inorganic substances. It must be assumed that it is not possible to predict biological effects from chemical analysis. But the primary objective of this study was to understand, if chemical summarizing parameters have a predictive character concerning toxicity. For this purpose the samples were listed according to their harmful effect (expressed as number of immobile daphnids, as inhibition of algal growth and of bacterial luminescence in percent in the undiluted sample) and according to the chemical results (in mg/L). The correlations were obtained by comparing the toxicity versus the effluent concentration of AOX, TOC and TN_b.

Table 5. Spearman correlation coefficients between toxicity and selected chemical data of effluents from different chemical water treatment plants (n = 81)

		Chemical screening procedure		
Bioassay	Indicator organism	TOC	AOX	TN_b
Microtox (30 min)	*Vibrio fisheri*	0.19	0.52[a]	0.12
Algae	*Scenedesmus subspicatus*	0.0	0.29[b]	-0.03
Daphnids	*Daphnia magna*	0.19	0.41[c]	-0.07

[a]p < 0.01% (two sided), that similar ranking occured by chance
[b]p < 1% (two sided), that similar ranking occured by chance
[c]p < 0.1% (two sided), that similar ranking occured by chance

The correlation coefficients (r), describing the strength of association, are presented in Table 5. Microtox results correlated best with AOX data (r = 0.52). Munkittrick et al. (1991) also found a remarkable strong correlation of Microtox with AOX. Likewise the correlation of toxicity on algae and on daphnids was strongest with AOX. The probability was low that the similar ranking of wastewater by toxicity to daphnids ($p < 0.1\%$), algae ($p < 1\%$), microtox ($p < 0.01\%$) and AOX content could have occured by chance. In a way AOX can be regarded as a predictor of bioassays results. Increasing AOX content in wasterwater from chemical industrial plants can be understood as an indication of increasing toxiciy. But this hint should not be overestimated. The highest correlation coefficient of 0.52 also means that the AOX values explain only 27% of the variation in the toxicity data. Between biological effects and the group parameters TN_b and TOC there are obviously no relations.

Concerning the toxicity of effluents from the pulp and paper industry, luminescent bacteria showed a good correlation to the corresponding AOX values, whereas algae showed a bad and daphnids no correlation (Aschacher 1992). But for O'Connor et al. (1993) the group parameter for chlorinated organic compounds was not a good predictor for the toxicity of treated wastes.

The findings reported here indicate that algal growth has a poorer correlation to AOX content than bacterial bioluminescence or immobility of daphnids. In this study algal growth was not inhibited but stimulated by 48% of the tested samples. Stimulation of growth could also be noted by Walsh et al. (1982) for all wastes from different industries (chemical industry included). None was strongly inhibitory to algal growth, but all samples stimulated growth at low concentrations.

This phenomenon can be linked with the fact that the analyzed effluents contained anorganic nutrients which must be added to the growth medium. This fact leads to an enrichment of the nutrient medium and thus to a weaked phytotoxic response. For Turbak et al. (1986) nutrient-enriched samples were less sensitive to herbicidal compounds. A ranking analysis with nontoxic samples to algae showed that the stimulation response was not reflected in the concentration of inorganic nitrogen compounds (sum of nitrate and ammonium). Also, Mayer et al. (1998) did not find detectable effects on algal growth by ammonium and nitrate as nitrogen source. A significant link could be established ($p < 5\%$ two-sided; n = 30) between growth stimulation and phosphate content. The main nutrient component increasing algal growth was phosphate.

Another factor that can negatively affect algal bioassays with industrial wastes is, apart from nutrient enrichment, the degradation of toxicants by light, that is inevitable for algal growth (Walsh et al 1982).

Table 6. Comparative study of bioassay results evaluated with chemical wastewater samples by Spearman correlation analysis (n = 81)

	Bioassay		
Bioassay	Daphnids	Algae	Microtox (30 min)
Daphnids	-	0.52[a]	0.19
Algae	0.52[a]	-	0.32[b]

[a] $p < 0.01\%$ (two-sided), that similar ranking occured by chance
[b] $p < 1\%$ (two-sided), that similar ranking occured by chance

A marginal aspect of this work is the comparison of the sensitivity pattern between the test systems. It was estimated by the Spearman ranking test and the results are shown in Table 6. A good correlation exists between algal and *Daphnia* assays. There is no great difference in the sensitivity pattern between both tests.

The correlation between effluent toxicity in the Microtox assay and in the algal growth was markly weaker. No correlation was observed between Microtox and *Daphnia* assays. This completely agrees with results presented by Wängeberg et al. (1995). Testing industrial wastewaters they also found the poorest correlation between microtox and daphnids (probability of similar ranking < 95%) and significant correlations (probability of similar ranking > 95%) between microtox and algae and between algae and daphnids.

The study demonstrates that from the AOX chemical data it is to a certain degree possible to predict the toxic property of wastewater originating from the chemical industry. Group parameters like TN_b and TOC have no signification as indicator parameters.

REFERENCES

Aschacher GP (1992) AOX and Toxicity. Holzforschung und Holzverwertung 6: 101-103

Basler H (1988) Equivalence between Tie-corrected Spearman Test and a Chi-Square Test in a Fourfold Contingency Table. Metrika 35: 203-209

O'Connor BI, Kovacs TG, Voss RH, Martell PH (1993) A study of the relationship between laboratory bioassay response and AOX content for pulp mill effluents. J Pulp Paper Sci 19:J33-J39

Mayer P, Frickmann J, Christensen E, Nyholm N (1998) Influence of growth conditions on the results obtained in algal toxicity tests. Environ Toxicol Chem 17: 1091-1098

Munkittrick KR, Power EA (1991) The relative sensitivity of Microtox, daphnid, Rainbow Trout, and feathed minnow acute lethality tests. Environ Toxicol Water Qual 6: 35-62

Turbak SC, Olson SB. McFeters GA (1986) Comparison of algal assay systems for detecting waterborne herbicides and metals. Wat Res 20: 91-96

Vasseur P, Ferard JF (1984) Comparaison des tests Microtox et daphnie pour l'evaluation de la toxicite aigue d'effluents industriels. Environ Pollut A 34:225-235

Walsh GE, Duke KM, Foster RB (1982) Algae and crustaceans as indicator of bioactivity of industrial wastes. Wat Res 16: 879-883

Wängberg S-A, Bergström B, Blanck H, Svanberg O (1995) The relative sensitivity and sensitivity patterns of short-term toxicity tests applied to industrial wastewaters. Environ Toxicol Water Qual 10:81-90

Zimmermann U (1992) Vergleichende Untersuchungen ausgewählter Abwasserproben von industriellen Einleitern mit biologischen Verfahren. Umweltplanung, Arbeits- und Umweltschutz 137: 1-98