BIOMEDICAL AND ENVIRONMENTAL SCIENCES 13, 56-68(2000)

Biological Degradation of Some Organic Compounds Enrolled in Paper Industry—A Pollution Prevention Approach

RIFAAT ABDEL WAHAAB

Water Pollution Research Department, National Research Centre, Dokki, Cairo, Egypt

Evaluation of the elimination and the "ultimate" biodegradation by aerobic microorganisms of some organic compounds commonly used in paper manufacturing technology was investigated. Biodegradation lines of nine organic compounds were determined as percentage removal of chemical oxygen demand (COD) over 7 days incubation. The results of the biodegradability test clearly revealed that some of the organic compounds under investigation are highly biodegradable, while others rank from fairly to even non-biodegradable. Significant biodegradation results were recorded for anti-coating ester (95.0%), basoplast 200D (85.3%) and basoplast PR 8050(87.6%). A bleaching agent (formamidin-sulfinic acid), ukanol BSA and solidurit KM demonstrate moderate biodegradation results of 62.1%, 76.2% and 69.8%, respectively. Poor biodegradation results for Hedifix M/35 (12.7%), basazol orange (34.9%) and basazol brown (29.0%) were recorded. Accordingly, appropriate precaution should be taken into consideration when using these compounds for industrial applications.

INTRODUCTION

During recent years, problems related to the contamination of the environment with different pollutants have become very serious. There has been a steady increase in the number of synthesized chemicals encountered in the environment. However, in spite of new chemicals occurring at a rate of 6 000 per week, the number of chemicals in common use is estimated at about 65 000, of greater than 4 000 000 known (Ames and McCann, 1981). These contaminants enter humans and their environment through complex and interrelated pathways. They often become concentrated through the food chain, with minute quantities magnified thousands of time as they are consumed by higher forms of life (Wahaab, Lubberding and Alaerts, 1995).

The need for standard test procedures to determine the biodegradability of these new products (and of estimating their potential impact upon the environment as well) is becoming self-evident. Many investigators have proposed scenarios for testing and evaluating new products, as discussed in the early extensive review (Ludzack and Ettinger, 1963a).

The Water Pollution Control Federation Subcommittee on Biodegradability has enunciated general principles that should apply in the testing of any substances (WPCF, 1967). Between the two extremes of primary biodegradation and ultimate biodegradation, the Subcommittee offers the concept of "Environmental Acceptable Biodegradation" (ECB). The ECB concept is defined as "susceptibility to biodegradation yielding endproducts which are totally acceptable in the receiving environment". These principles are of considerable value in trea-

0895-3988/2000
CN 11-2914

tability studies of wastewater. Wastewater produced from the paper industry is very complex in nature. The pollution in the wastewater of a paper mill depends on the raw materials, the type and amount of filters and additives applied, and on the degree of circuit closure (Casey, 1980).

The main sources of wastewater in the pulp and paper industry are usually: (Ⅰ) debarking wastewater, (Ⅱ) mechanical pulping wastewater, (Ⅲ) kraft black and spent sulfate liquor (chemical pulping), (Ⅳ) bleaching wastewater, and paper mill wastewater. The hazardous impact of pulp and paper industry effluents can be attributed to a complex mixture of extractive compounds, including resin and fatty acids (Junna and Rintala, 1989); tannins (Temmink *et al.*, 1989); some lignin degradation products and chlorinated phenols, if chlorine bleaching is practiced (Galvao *et al.*, 1987), low molecular weight chlorinated lignin derivatives (Kringstad and Lindstrom, 1984), and a variety of other chemical compounds.

In Egypt, the pulp and paper industry relies on agricultural residues as the source of raw material; rice straw and bagasse represent 90% of the fibers used in pulping. Using rice straw relies on a primitive technology for pulping the residues compared to the new and sophisticated methods for pulping wood. The black liquor, containing about one half by weight of the straw quantity and all of the cooking chemicals, causes serious environmental problems. The problem of appearance of non-biodegradable compounds in the ecosystem is that these pollutants do not decompose or be eliminated from the environment. Therefore, these raw materials and any of the other additives used during the industrial processes have to be certified before use in order to avoid any future environmental contamination problems.

The aim of the present study was to investigate the biological degradability of some chemical compounds commonly used in the paper industry in Egypt.

MATERIALS AND METHODS

Principle

The Zahn-Wellens Test (1991) determines the biodegradation and elimination of water-soluble organic compounds from an aqueous phase by aerobic microorganisms. The organic compounds are the sole source of carbon and energy in the medium other than the sludge. The concentration of the compounds used is such that the initial concentration of chemical oxygen demand (COD) is between 100 $mg \cdot L^{-1}$ and 1000 $mg \cdot L^{-1}$. COD is measured at the beginning and end of the test (7 days) and at seven regular intermediate time intervals. Evaluation of the biodegradability of the compounds was made on the basis of these data.

Experimental Set-Up

Test environment and reagents. Incubation took place in the dark at a temperature of 20°. The solutions used in the experiment were: (Ⅰ) nutrient solutino: anhydrous potassium dihydrogen phosphate (KH_2PO_4), anhydrous dipotassium hydrogen phosphate (K_2HPO_4), disodium hydrogen phosphate dihydrate ($Na_2HPO_4 \cdot 2H_2O$) and ammonium chloride (NH_4Cl), (Ⅱ) magnesium sulfate heptahydrate ($MgSO_4 \cdot 7H_2O$), (Ⅲ) anhydrous calcium chloride ($CaCl_2$) and (Ⅳ) ferric chloride hexahydrate ($FeCl_3 \cdot 6H_2O$).

Procedure. A suitable concentration of 3000 $mg \cdot L^{-1}$ for each solution of the nine test compounds was chosen. The reference compound used in this experiment was sodium benzoate at a concentration of 3000 $mg \cdot L^{-1}$. Preparation of the inoculum was carried out by collecting a sample of activated sludge from the aeration tank of a biological treatment facility. The sample was mixed well and washed by adding tap water, allowing the sample to settle, and then decanting of the supernatant. Before use, determination of the concentration of the suspended solids was calculated. The inoculum was aerated at room temperature (20℃) until it was used.

Two test vessels containing at least 500 ml of the test compound were prepared. A sufficient amount of the test compound solution was added to obtain a COD concentration that ranged from 300 to 1000 $mg \cdot L^{-1}$ in the final mixture. The pH value was adjusted to pH 7 ± 0.5. Activated sludge was added in an amount corresponding to 0.2 $g \cdot L^{-1}$ suspended solids in the final mixture. The test medium was made to a total volume of 2 liters. One blank containing only activated sludge was prepared to operate in parallel with each test series. In order to check the activity of the inoculum, at least one control vessel with a solution of reference compound was prepared to operate in parallel with each series. Another vessel for abiotic control using only the test compound (no inoculum) was prepared. To start the test, all vessels were agitated using stirrers. Samples were taken after 3 hours followed by daily samples over a period of 7 days. The samples were centrifuged and filtered through glass wool. COD concentrations in the filtrate were measured for each vessel in duplicate according to APHA approved methods (1995). Biodegradation of the test compound was measured as the percentage of COD removal and calculated according to the following equation:

$$\text{Degradation}(\%) = \frac{(COD_{cor(0)} - COD_{Bio(0)}) - (COD_{cor(t)} - COD_{Bio(t)})}{(COD_{cor(0)} - COD_{Bio(0)})} \times 100$$

$COD_{cor(0)}$ = The corrected COD value of the sample at zero time.

$COD_{cor(t)}$ = The corrected COD value of the sample at time (t) plus the blank difference. The blank difference = the difference in COD value between time (t) and the preceding value.

$COD_{Bio(0)}$ = The COD of the Biomass at zero time.

$COD_{Bio(t)}$ = The COD of the Biomass at time (t).

RESULTS AND DISCUSSION

The essential steps in the manual papermaking process are sheet forming, pressing, drying, sizing, and calendering. This fundamental process has not changed over time. In sheet forming, the water from a fiber suspension is filtered off on a rotating continuous wire to produce a wet web. The mass distribution in the web should be as uniform as possible. In the press section, the wet web is compressed between rolls and rotating felts by mechanical pressure, and dewatered as far as possible. In the size press, starch, size, or pigments are transferred onto or into the fiber mat.

The use of starch, paper auxiliaries or other organic additives markedly increases of the oxidizable effluent component, measured as BOD or COD. Dyes and fillers can lead to discoloration and/or turbidity of effluents.

Biodegradation of the reference compound (Sodium Benzoate) reached 99.0%. This confirms excellent activity of the inoculum used in these series of experiments. Biodegradation of some of these compounds will be discussed below.

Anti-Coating Ester

This product is used in the manufacturing processes of tissue and hygienic paper only. It is applied directly to the dryer as a releasing agent to prevent the fiber sheet from sticking to the cylinder surface. Chemically, this product is mainly an ester of long chain fatty acids. This product demonstrates good biological degradation. COD removal reached 38.2% after 24 h incubation and increased gradually to 95% on the 7th day of the experiment (Table 1 and Fig. 1). It is worth mentioning that this product is highly biodegradable and could be applied in paper manufacturing processes without any environmental impact.

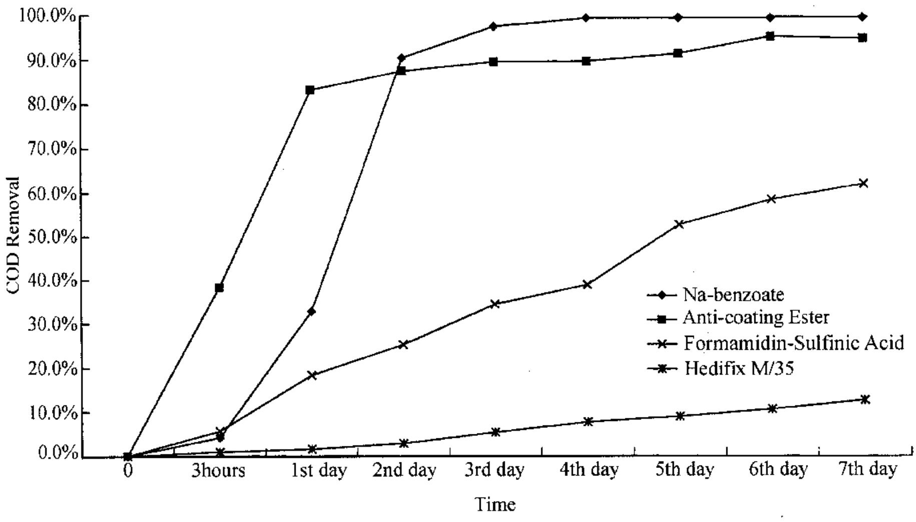

FIG. 1. Biological degradation of paper auxiliaries as a percentage of COD removal.

Formamidin-Sulfinic Acid (*FAS*)

Formamidin-Sulfinic Acid (FAS) is used in paper manufacturing as a bleaching agent. The results recorded in Table 1 and illustrated graphically in Fig. 1 demonstrate poor biological degradation of this product. Only 62.1% removal in COD was achieved after 7 days incubation. The residual COD value is still high (185 $mg \cdot L^{-1}$). These results show clearly that the FAS compound has to be restricted in use and/or substituted by another biodegradable product.

Hedifix-M/35

This product is applied during the paper manufacturing process to avoid any contamination of the wire by sticky precipitation of the fiber suspension (i. e. conditioning the wire). Chemically, this product is an ammonium salt bonded with methyl groups. The results indicated that this product is hardly or even non-biodegradable. Only 12.7% removal in COD was achieved after 7 days (Table 1 and Fig. 1). Therefore, this product should be restricted in use or substituted by another biode-

TABLE 1
Biological Degredation of Paper Auxiliaries

Product	Parameter	Unit	0	3 hours	1st day
Biomass Suspention	COD	$mg \cdot L^{-1}$	22	22	15
Na-benzoate	COD	$mg \cdot L^{-1}$	870	835	585
	Calculation Raw			35	278
	COD Removal	%	0.0%	4.1%	32.8%
Anti-coating Ester	COD Blank	$mg \cdot L^{-1}$	359	328	325
	Blank Difference			31	3
	COD Sample 1	$mg \cdot L^{-1}$	247	115	50
	Sample Correction			146	53
	Calculation Raw			101	187
	Removal Sample 1	%	0.0%	44.9%	83.1%
	COD Sample 2	$mg \cdot L^{-1}$	202	114	45
	Sample Correction			145	48
	Calculation Raw			57	150
	Removal Sample 2	%	0.0%	31.7%	83.3%
	Average		0.0%	38.3%	83.2%
Formamidin-Sulfinic Acid	COD Blank	$mg \cdot L^{-1}$	322	318	314
	Blank Difference			4	4
	COD Sample 1	$mg \cdot L^{-1}$	388	365	326
	Sample Correction			369	330
	Calculation Raw			19	51
	Removal Sample 1	%	0.0%	5.2%	13.9%
	COD Sample 2	$mg \cdot L^{-1}$	455	425	346
	Sample Correction			429	350
	Calculation Raw			26	98
	Removal Sample 2	%	0.0%	6.0%	22.6%
	Average		0.0%	5.6%	18.3%
Hedifix M/35	COD Blank	$mg \cdot L^{-1}$	580	575	573
	Blank Difference			5	2
	COD Sample 1	$mg \cdot L^{-1}$	555	545	540
	Sample Correction			550	542
	Calculation Raw			5	6
	Removal Sample 1	%	0.0%	0.9%	1.1%
	COD Sample 2	$mg \cdot L^{-1}$	570	560	550
	Sample Correction			565	552
	Calculation Raw			5	11
	Removal Sample 2	%	0.0%	0.9%	2.0%
	Average		0.0%	0.9%	1.6%

2nd day	3rd day	4th day	5th day	6th day	7th day
18	22	23	21	27	25
99	43	28	25	24	23
767	827	843	844	844	846
90.4%	97.5%	99.4%	99.5%	99.5%	99.8%
320	313	300	290	285	277
5	7	13	10	5	8
42	42	39	38	39	37
47	49	52	48	44	45
196	198	196	198	208	205
87.1%	88.0%	87.1%	88.0%	92.4%	91.1%
40	38	37	30	30	27
45	4	50	40	35	35
158	164	166	171	177	178
87.8%	91.1%	92.2%	95.0%	98.3%	98.9%
87.4%	89.6%	89.7%	91.5%	95.4%	95.0%
305	300	298	296	290	280
9	5	2	2	6	10
296	263	246	200	179	157
305	268	248	202	185	167
79	120	141	185	208	224
21.6%	32.8%	38.5%	50.5%	56.8%	61.2%
317	294	284	214	193	175
326	299	286	216	199	185
125	156	170	238	261	273
28.9%	36.0%	39.3%	55.0%	60.3%	63.0%
25.2%	34.4%	38.9%	52.8%	58.6%	62.1%
565	560	557	555	550	548
8	5	3	2	5	2
530	520	511	509	499	480
538	525	514	511	504	482
13	30	42	43	56	76
2.4%	5.6%	7.9%	8.1%	10.5%	14.3%
540	537	520	513	511	510
548	542	530	515	516	512
18	28	41	54	59	61
3.3%	5.1%	7.5%	9.9%	10.8%	11.1%
2.9%	5.4%	7.7%	9.0%	10.6%	12.7%

TABLE 2
Biological Degredation of Paper Auxiliaries

Product	Parameter	Unit	0	3 hours	1st day
Biomass Suspension	COD	$mg \cdot L^{-1}$	35	31	27
Na-benzoate	COD	$mg \cdot L^{-1}$	596	540	535
	Calculation Raw			52	53
	% COD Removal		0.0%	9.3%	9.4%
Basaplast 200D	COD Blank	$mg \cdot L^{-1}$	489	475	460
	Blank Difference			14	15
	COD Sample 1	$mg \cdot L^{-1}$	361	110	80
	Sample Correction			124	95
	Calculation Raw			233	258
	Removal Sample 1	%	0.0%	71.5%	79.1%
	COD Sample 2	$mg \cdot L^{-1}$	350	100	75
	Sample Correction			114	90
	Calculation Raw			232	267
	Removal Sample 2	%	0.0%	73.7%	84.8%
	% Average		0.0%	72.6%	82.0%
Basaplast PR 8053	COD Blank	$mg \cdot L^{-1}$	420	416	395
	Blank Difference			4	21
	COD Sample 1	$mg \cdot L^{-1}$	224	80	59
	Sample Correction			84	80
	Calculation Raw			136	136
	Removal Sample 1	%	0.0%	72.0%	
	COD Sample 2	$mg \cdot L^{-1}$	217	82	60
	Sample Correction			86	81
	Calculation Raw			127	128
	Removal Sample 2	%	0.0%	69.8%	70.3%
	% Average		0.0%	70.9%	71.1%
Basazol orange 52L	COD Blank	$mg \cdot L^{-1}$	893	890	860
	Blank Difference			3	30
	COD Sample 1	$mg \cdot L^{-1}$	766	710	670
	Sample Correction			713	700
	Calculation Raw			49	58
	Removal Sample 1	%	0.0%	0.7%	7.9%
	COD Sample 2	$mg \cdot L^{-1}$	750	700	660
	Sample Correction			703	690
	Calculation Raw			43	52
	Removal Sample 2	%	0.0%	6.0%	7.3%
	% Average		0.0%	6.4%	7.6%

2nd day	3rd day	4th day	5th day	6th day	7th day
25	22	20	20	15	15
274	93	38	30	25	20
312	490	543	551	551	556
55.6%	87.3%	96.8%	98.2%	98.2%	99.1%
424	380	340	300	270	220
36	44	40	40	30	50
72	60	59	58	58	40
108	104	99	98	88	90
243	244	247	248	263	251
74.5%	74.8%	75.8%	76.1%	77.6%	77.0%
65	55	45	42	40	35
101	99	85	82	70	85
275	282	290	293	290	295
87.3%	89.5%	92.1%	93.0%	92.1%	93.7%
80.9%	82.2%	83.9%	84.5%	84.8%	85.3%
380	370	355	345	340	328
15	10	15	10	5	12
50	42	41	35	35	27
65	52	35	45	40	39
149	159	168	164	164	165
78.8%	84.1%	88.9%	86.8%	86.8%	87.3%
40	38	37	31	30	25
55	48	52	41	35	37
152	156	150	161	162	160
83.5%	85.7%	82.4%	88.5%	89.0%	87.9%
81.2%	84.9%	85.7%	87.6%	87.9%	87.6%
840	838	825	815	808	803
20	2	13	10	7	5
668	606	602	550	538	486
688	608	615	560	545	491
68	145	136	191	201	225
9.3%	19.8%	18.6%	26.1%	27.5%	34.9%
648	610	603	560	545	475
668	612	616	570	552	480
72	125	119	165	178	250
10.1%	17.5%	16.6%	23.1%	24.9%	35.0%
9.7%	18.7%	17.6%	24.6%	26.2%	34.9%

gradable product to avoid environmental contamination problems.

Basoplast 200D

Basoplast 200D is a fatty alkyl diketene emulsion with a low cationic charge density. This product is a very effective size for paper made in neutral and slightly alkaline media, especially in paper made with furnish that contains chalk. The results indicated that this product is quite biodegradable. It seems that the product is adsorbed on the surface of the activated sludge flocs. COD removal reached 72.6% following only 3 h exposure. The percentage removal in the COD value after 7 days was 85.3% (Table 2 and Fig. 2).

Basoplast PR 8053

This product is a fatty alkyl diketene dispersion with a medium concentration, for sizing paper in a neutral pH range. The product is very effective for sizing papers made from furnish that contains groundwood or recycled paper. No significant difference between Basoplast 200D and Basoplast PR 8053 in biological degradation rates were observed during the experiment. COD removal reached 70.9% following 3 h exposure. The percentage removal in the COD value after 7 days was 87.6% (Table 2 and Fig. 2).

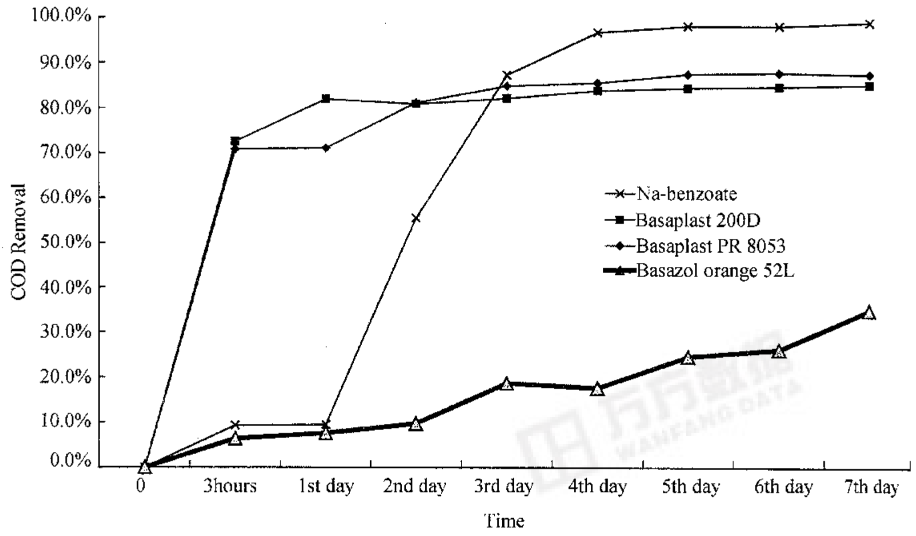

FIG. 2. Biological degradation of paper auxiliaries as a percentage of COD removal.

Basazol Orange 52 L

Basazol Orange 52 L is an effective alternative to acid orange dyes because of its higher affinity for wood-containing furnish and its brilliance on papers made from furnish of this type. Results recorded in Table 2 and illustrated graphically in Fig. 2 demonstrate that this product is poorly biodegradable, even after 7 days exposure. Only 34.9% removal in COD value was achieved. High residual concentrations of this type of dyes in the industrial effluent could cause serious impact on the performance of the biological treatment plant.

Basazol Brown 32 *L*

Basazol Brown 32 L (Methine Brown Dye) can be used to dye paper in the acid to slightly alkaline pH range. Paper made from raw materials that contain lignin, such as groundwood, unbleached chemical pulp and recycled paper can be dyed without any additional chemicals. The largest area of application of this dye is in the production of recycled packaging papers.

Basazol Brown 32 L in an aqueous solution is not easily biodegradable by the microorganisms. A maximum removal value of 29.0% in COD was achieved after 7 days retention time, with a residual concentration of 272 $mg \cdot L^{-1}$ as the non-biodegradable value. Therefore, good dying systems to minimize and/or prevent waste dyes in the industrial effluent must be taken into consideration (Table 3 and Fig. 3).

Ukanol BSA 1

The chemical nature of this product is acrylat-copolymer. It is an adhesive material, used as a special glue for barrier coated and hydrophobic paper surfaces. This product is moderately biodegraded. COD removal began at 36.4% after 24 h followed by 50.1% in the 2nd day and increasing gradually to 76.2% following 7 days digestion (Table 3 and Fig. 3). The residual value is still relatively high (75 $mg \cdot L^{-1}$).

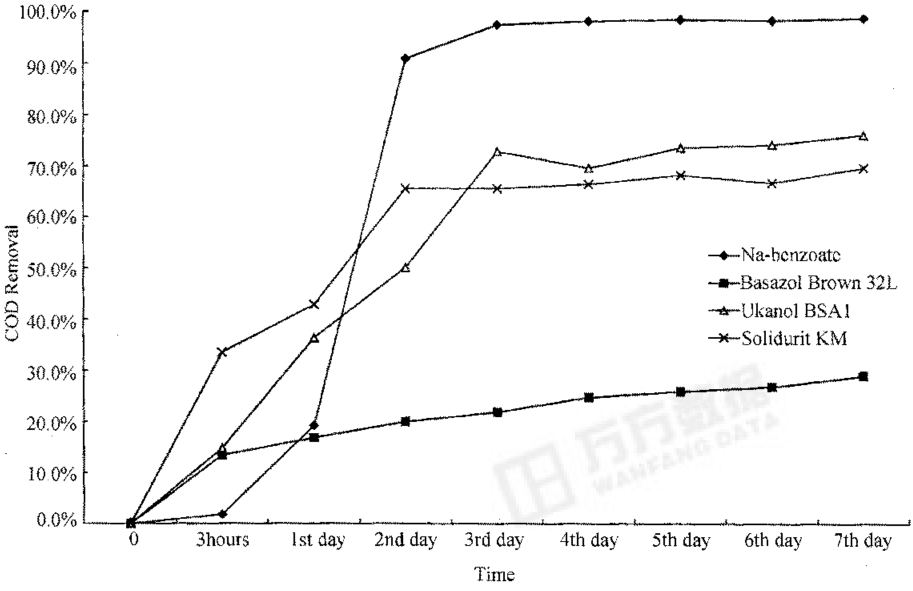

FIG. 3. Biological degradation of paper auxiliaries as a percentage of COD removal.

TABLE 3

Biological Degredation of Paper Auxiliaries

Product	Parameter	Unit	0	3hours	1st day
Biomass Suspension	COD	$mg\cdot L^{-1}$	35	31	27
Na-benzoate	COD	$mg\cdot L^{-1}$	587	573	473
	Calculation Raw			10	106
	% COD Removal		0.0%	1.8%	19.2%
Basazol Brown 32 L	COD Blank	$mg\cdot L^{-}$	535	530	512
	Blank Difference			5	18
	COD Sample 1	$mg\cdot L^{-1}$	375	320	300
	Sample Correction			325	318
	Calculation Raw			46	49
	Removal Sample 1	%	0.0%	13.5%	14.4%
	COD Sample 2	$mg\cdot L^{-1}$	388	332	312
	Sample Correction			337	330
	Calculation Raw			47	68
	Removal Sample 2	%	0.0%	13.3%	19.3%
	% average		0.0%	13.4%	16.8%
Ukanol BSA1	COD Blank	$mg\cdot L^{-1}$	70	67	64
	Blank Difference			3	3
	COD Sample 1	$mg\cdot L^{-1}$	293	239	180
	Sample Correction			242	183
	Calculation Raw			47	102
	Removal Sample 1	%	0.0%	18.2%	39.5%
	COD Sample 2	$mg\cdot L^{-1}$	282	247	189
	Sample Correction			250	192
	Calculation Raw			28	82
	Removal Sample 2	%	0.0%	11.3%	33.2%
	% average		0.0%	14.8%	36.4%
Solidurit KM	COD Blank	$mg\cdot L^{-1}$	223	211	191
	Blank Difference			12	20
	COD Sample 1	$mg\cdot L^{-1}$	200	129	101
	Sample Correction			141	121
	Calculation Raw			55	71
	Removal Sample 1	%	0.0%	33.3%	43.0%
	COD Sample 2	$mg\cdot L^{-1}$	204	131	104
	Sample Correction			143	124
	Calculation Raw			57	72
	Removal Sample 2	%	0.0%	33.7%	42.6%
	% average		0.0%	33.5%	42.8%

2nd day	3rd day	4th day	5th day	6th day	7th day
25	22	20	17	15	15
75	36	30	25	24	21
502	538	542	544	543	546
90.9%	97.5%	98.2%	98.6%	98.4%	98.9%
493	479	474	472	470	463
19	14	5	2	2	7
285	281	270	265	260	250
304	295	275	267	262	257
61	67	85	90	93	98
17.9%	19.7%	25.0%	26.5%	27.4%	28.8%
300	290	286	280	275	265
319	304	291	282	277	272
78	85	87	90	93	103
22.1%	24.1%	24.6%	25.5%	26.3%	29.2%
20.0%	21.9%	24.8%	26.0%	26.8%	29.0%
40	38	18	15	13	12
24	2	20	3	2	1
130	88	88	80	78	74
154	90	35	83	80	75
129	190	190	192	193	198
50.0%	73.6%	73.6%	74.4%	74.8%	76.7%
124	89	85	81	78	74
148	91	105	84	80	75
124	178	162	180	182	187
50.2%	72.1%	65.6%	72.9%	73.7%	75.7%
50.1%	72.9%	69.6%	73.6%	74.2%	76.2%
187	183	178	176	172	170
4	4	5	2	4	2
78	75	72	68	66	64
82	79	77	70	70	66
108	108	108	112	110	114
65.5%	65.5%	65.5%	67.9%	66.7%	69.1%
79	76	70	68	67	63
83	80	75	70	71	65
111	111	114	116	113	119
65.7%	65.7%	67.5%	68.6%	66.9%	70.4%
65.6%	66.5%	66.5%	68.3%	66.8%	69.8%

Solidurit KM

Solidurit KM is a polyacrylamide material. These products are applied in paper manufacturing as wet strengthening agents. The products are intended to increase the tensile stre-ngth, tearing strength, bursting strength and the abrasion resistance of the wet paper.

Results of the biological degradation indicated low removal value in COD (69.8%) after 7 days digestion (Table 3 and Fig. 3). The residual COD value is relatively high (65 $mg \cdot L^{-1}$). It is worth mentioning that 33.5% removal of COD was achieved after only 3 hours incubation. The removal in COD is not actually biodegradation of the product under investigation but may be due to the physical adsorption of that product on the activated sludge floc surface.

CONCLUSION

From the results of the present study, it may be concluded that the problem of hazardous pollutants in the environment is closely connected with increased concentrations of non-biodegradable materials that enter water bodies through industrial and communal waters. There is now little doubt that many diseases are caused by such hazardous environmental pollutants. Therefore, a need to test and certify not only chemical compounds and other additives applied in the paper industry but also any new synthetic chemicals used in the industrial sector in general is self-evident. Accordingly, a clear vision of the biodegradation lines and toxicological impact of these new synthetic compounds should be identified, labeled and assessed.

REFERENCES

Abdel Wahaab, R., Lubberding, H.J., and Alaerts, G.J. (1995). Copper and Chromium (Ⅲ) uptake by duckweeds. *Water Sci. Technol.* **32**, 105-110.

Ames, B.N. and McCann, J. (1981). Chemical characterization of 465 known or suspected carcinogens and their correlation with mutagenic activity in *Salmonella typhimurium* system. *Cancer Res.*, **41**, 4192-4196.

APHA, AWWA, WPEC (1995). *Standard Methods for the Examination of Water & Wastewater*, 18th ed. Washington D.C.

Casey, J.P. (1980). *Pulp and Paper, Chemistry and Chemical Technology*. John Wiley & Sons, New York.

Galvao, J.B., Grieco, V.M., Araujo, R.P.A., Ortelano, M.R., Bertoletti, E., and Ramos, M.L.L.C., (1987). Treatability studies and toxicity reduction in pulp and paper mill effluent. In *Proc. Second IAWPRC Symposium on Forest Industry Wastewater*. Tampere Univ. of Technology, Tampere, Finland.

Junna, J. and Rintala, J. (1989). Evaluation of purification efficiency of activated sludge treatment plants for pulp and paper industry wastewater in Finland. In *Proc. Symp. on Wastewater Management Problems in Agro-industry*, pp.253-260. Istanbul, Turkey.

Kringstad, K.P. and Lindstrom, K. (1984). Spent liquors from pulp bleaching. *Environ. Sci. Technol.* **18**, 236-248.

Ludzack, F.J. and Ettinger, M.B. (1963a). Estimating biodegradability and treatability of organic water pollutants. Biotechnol. *Bioeng.*, **5**, 309-330.

Temmink, J.H.M., Field, J.A., van Haastrecht, J.C., and Merckelbach, R.C.M. (1989). Acute and sub-acute toxicity of bark tannins to carp(*Cyprinus carpiol*). *Water Res.* **23**, 341-344.

WPCF (1967). *Proceeding of the Industrial Conference on Water Pollution Research*, **3**, *WPCF*, Washington D.C.

Zahan-Wellens Method (1991). Water Quality-Evaluation of the aerobic biodegradability of organic compounds in an aqueous medium-static test. *ISO* 9888:1991 (E), 1st *ed. Bonn, Germany*.

(*Received September* 1, 1999 *Accepted January* 18, 2000)