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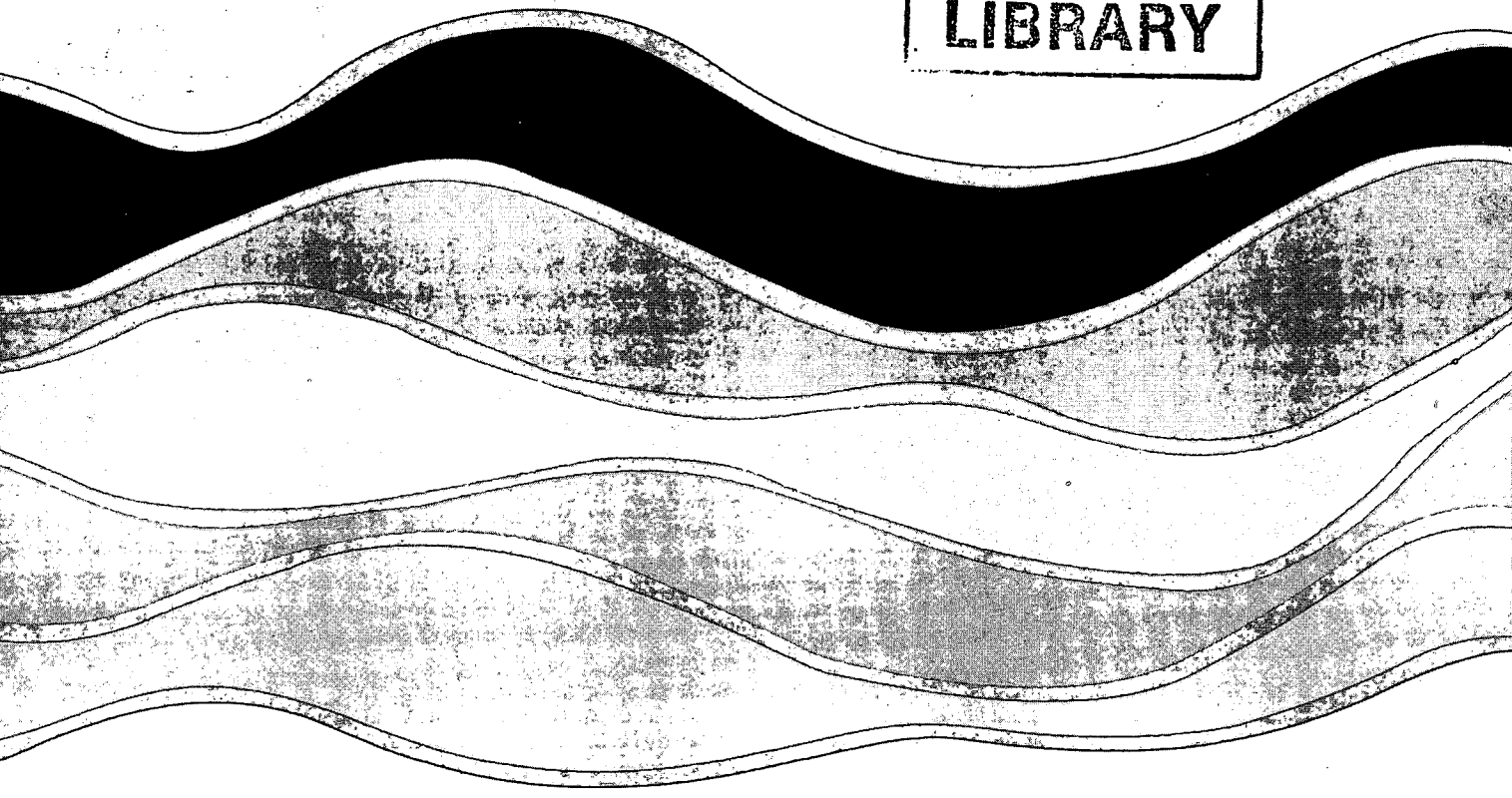

DETERMINATION OF TEXTILE DYES BY HIGH PERFORMANCE LIQUID CHROMATOGRAPHY - DIODE ARRAY SPECTROPHOTOMETRY

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Management Perspective

Dyes and pigments are high volume chemicals which may enter aquatic environments in significant quantities. Interest in the environmental behaviour of dyes arose largely from concerns about carcinogenicity. Very little is known of the environmental occurrence, persistence and fate of dyes, largely because of difficulties in their determination at trace levels in environmental media. There is no environmental information on dyes in Canada. We have initiated a study of the occurrence, persistence, fate and effects of dyes in the Yamaska River in Quebec. This report describes analytical methods developed for various classes of dyes in water, their purity, and a determination of the occurrence of dyes in a textile mill wastewater treatment plant influent at Magog, Quebec.

PERSPECTIVE GESTION

Les colorants et les pigments sont des produits chimiques utilisés en grande quantité dont une proportion importante peut se retrouver dans les milieux aquatiques. L'intérêt que l'on porte au rôle joué par les colorants dans l'environnement a été suscité en grande partie par les préoccupations concernant la cancérogénicité. Nous possédons très peu de données sur la présence, la persistance et le devenir des colorants dans l'environnement; cela est dû en grande partie au fait qu'ils sont difficiles à mesurer à l'état de traces dans les milieux environnementaux. Il n'existe pas de données sur les colorants dans l'environnement au Canada. Nous avons entrepris une étude de la présence, de la persistance, du devenir et des effets des colorants dans la rivière Yamaska, au Québec. Le présent rapport décrit les méthodes d'analyse mises au point pour les diverses catégories de colorants dans l'eau, leur pureté et une détermination de la présence des colorants dans l'influent d'une usine de traitement des eaux usées d'une usine de textile à Magog, au Québec.

ABSTRACT

An ion-pairing high performance liquid chromatographic - diode array spectrophotometric technique has been used to characterize 17 acid, basic, direct, disperse, mordant and reactive textile dyes. The analysis revealed that many of the dye formulations contain more than one component, and in some cases as many as six components. Analysis of samples of a textile mill water treatment plant influent revealed the presence of four red-orange dyes, one of which was tentatively identified by retention time and spectral matching as Acid Red 73. The four dyes appear to be efficiently removed in the treatment plant since none were detected in the effluent.

Key Words: dyes, analysis, HPLC, diode array spectrophotometry

RÉSUMÉ

Une technique faisant appel à la chromatographie liquide haute performance par formation de paires d'ions et à la spectrophotométrie à réseau de diodes a été utilisée pour caractériser 17 colorants de textile dans les catégories des colorants acides, basiques, directs, plasmosolubles, à mordant et réactifs. L'analyse a révélé que bon nombre des formulations de colorants renferment plus d'un colorant et dans certains cas, plus de six. L'analyse des échantillons d'influent d'une usine de traitement des eaux usées d'une usine de textile a révélé la présence de quatre colorants rouge orangé, dont l'un a été identifié provisoirement au Rouge acide 73 grâce à son temps de rétention et à ses données spectrales. Le procédé de traitement utilisé par l'usine de traitement des eaux usées semble être efficace car aucun des quatres colorants n'a été détecté dans l'effluent.

Mots clés : colorants, analyse, HPLC, spectrophotométrie à réseau de diodes

INTRODUCTION

Dyes and pigments are high volume chemicals which may enter aquatic environments in significant quantities. The distinction between dyes and pigments is largely made on the basis of solubility. Pigments have extremely low solubilities in water, and are not very soluble in organic solvents either, generally being applied in the solid state. Dyes are generally more soluble either in water or organic solvents. Global production of dyes in 1978 was estimated to be 6.4×10^8 kg of active ingredient.[1] About 10^8 kg were produced in the United States in 1986.[2] It has been estimated that 90% of dyes applied end up in fabrics, with the remaining 10% discharged to waste streams.[3] In 1982 about 6.5×10^6 kg of dyes and 2.5×10^6 of pigments were imported into Canada.[4] Most dyes were imported into Quebec and most pigments were imported into Ontario.

Interest in the environmental behaviour of dyes arose largely from concerns about carcinogenicity. Some dyes are carcinogens[5] and others after transformation or degradation yield compounds such as aromatic amines which may be carcinogenic or otherwise toxic.[6] Very little is known of the environmental occurrence, persistence and fate of dyes, largely because of difficulties in their determination at trace levels in environmental media. Due to the great number of dyes and their widely varying nature, there is no single method which would suffice for their chromatographic separation from each other and from co-eluting natural extractives. Gas-liquid chromatography is in general unsuitable because of the low vapour pressures of dyes and their thermal lability at high temperatures. High performance liquid chromatographic

(HPLC) techniques are currently preferred, with determination by single wavelength ultraviolet-visible (UV-VIS) spectrophotometry,[7-19] diode array spectrophotometry (DAD),[20-24] fluorometry[25] or mass spectrometry (MS).[18,26-38] Supercritical fluid column chromatography[39] and capillary zone electrophoresis[40] have also been used.

There is a good deal of interest in determining the occurrence, persistence and toxicity of dyes in rivers in the Eastern Townships of Quebec because much of Canada's textile industry is concentrated in the Eastern Townships, and there are anecdotal accounts of widespread contamination of rivers in the area by dyes. This article describes a HPLC-DAD technique for some classes of textile dyes used in the area and its use in characterizing the purity of some of the dyes. In addition, this article describes the presence of some dyes in influents and effluents of the water treatment plant of a textile mill at Magog on the St. Francois River.

EXPERIMENTAL

Materials

Twenty dyes representative of various classes used in Canada's textile industry were supplied by the Ecological and Toxicological Association of the Dyestuffs Manufacturing Industry. Their names and other information are given in Table I and their structures are given in Figs. 1 and 2. These dyes were used as received. Spectral and HPLC-DAD determinations of these dyes were done with HPLC grade solvents obtained from various suppliers. Organic-

free water was distilled water purified by carbon adsorption and reverse osmosis. Gas chromatography with electron capture detection (GC-ECD) of hexane extracts of this water revealed no contamination, even with 1000-fold concentration. All inorganic salts were analytical grade and were used without further purification.

For the extraction and cleanup of wastewater samples, pesticide grade dichloromethane and other solvents were obtained from different suppliers and their purity (at 1000-fold concentration) was checked before use by GC-ECD. The sodium sulfate and disposable pipets were heated to 500 °C for 24 h before use. Hydrochloric acid and sodium hydroxide solutions used for pH adjustment were prepared from reagent grade chemicals, but were extracted with hexane before use to eliminate contamination. ACRO LC 13 disposable filters with Luer inlets for removing particulate material from samples before HPLC analysis were obtained from Gelman Sciences (Montreal).

Methods

(a) dye standards

Preliminary characterization of dye spectra at room temperature in pH 7 buffer was done with a Shimadzu UV-260 spectrophotometer. In some cases methanol was used to solubilize the dyes. The wavelength of maximal absorption for each dye is shown in Table I.

HPLC analyses of the dye standards were done with a Waters Associates HPLC (Millipore-Waters, Mississauga, Ontario) and 990+ DAD. Samples of 50 μL volume were injected with a Waters WISP 712 autosampler onto a 15 cm x 4.6 mm i.d. reverse-phase Supelcosil LC-8-DB column preceded by a Supelguard LC-8-DB guard column (Supelco, Oakville, Ontario). Both guard and analytical columns were thermostatted at 30°C. The flow rate was 0.25 mL/min. Potassium dihydrogen phosphate was the ion-pair reagent. The following gradient program was employed using solvents A (acetonitrile) and B (0.01 M KH_2PO_4): 70% A - 30% B for 9 min, then a linear gradient to 100% A at 25 min, with a hold at 100% A until 45 min. The DAD recorded 200 - 800 nm spectra of the HPLC effluent every 200 ms. For each dye standard tested, the concentration was adjusted to yield an absorbance of 2.0 units at its wavelength of maximal absorbance as determined with the spectrophotometer.

(b) textile mill wastewater treatment plant influent and effluent

Four-litre samples of influent and effluent were collected from the wastewater treatment plant of a textile mill at Magog on June 12, 1985. The whole water samples were acidified with concentrated HCl to pH 1 and preserved with 200 mL of dichloromethane for transport to the laboratory. The dichloromethane extract of the influent sample was deep orange-pink, while that of the effluent sample was only faintly orange-pink. The samples were extracted in triplicate with dichloromethane within one week of collection. After extraction at acidic pH, the aqueous

phase was made basic to pH 12 by the addition of 10 N NaoH and the water was extracted three more times with dichloromethane in order to isolate any basic dyes which might have been present. The dichloromethane extracts were dried and concentrated to 10 mL. The 10 mL dichloromethane extracts were solvent-exchanged with hexane by addition of hexane and careful evaporation to 0.5 mL with a gentle flow of nitrogen. This procedure was done three times to remove traces of dichloromethane. The extract was made up to 1 mL. At this point there was a small amount of precipitation in the test tube, but all material was transferred to the cleanup column as described below.

The 1 mL hexane extracts were cleaned up with a technique developed for pesticides, PCBs and polynuclear aromatic hydrocarbons.[41] The extracts were applied to activated silica gel columns of length 20 cm and i.d. 1 cm, with a layer of sodium sulfate for drying. Four 100 mL fractions were eluted from the columns. Fraction 1 was hexane; fraction 2 was dichloromethane/hexane (20/80, v/v); fraction 3 was dichloromethane/hexane (60/40, v/v); fraction 4 was 50 mL of dichloromethane followed by 50 mL of methanol. At each solvent change, a little solvent was used to rinse the test tubes containing the original extract, and in this way even the precipitated material was transferred to the cleanup column. All four fractions were solvent-exchanged and reduced to 1 mL of isooctane "keeper". These cleaned up extracts were analyzed by HPLC-DAD as described above.

RESULTS AND DISCUSSION

The ion-pairing technique used is a modification of an earlier method [13], and although

the technique will not have universal applicability, it is suitable for 17 of the 20 dyes listed in Table I. Most importantly, it is suitable for disperse dyes, which are currently of interest because of their extensive use. The data will be useful in setting up retention time - spectral libraries for analyses of environmental samples. However, in the case of reactive dyes it is recognized that in most situations short of accidental discharges of large volumes, it is the hydrolysis products rather than the parent compounds which may be present in effluents. The method was not suitable for Basic Green 4, Basic Violet 1 and Basic Violet 3, which did not elute from the HPLC column over 45 min under the solvent conditions employed.

As expected, many of the dyes were not pure compounds, but were mixtures of dyes having similar spectra but different retention times. Some of the dye formulations had as many as six components, although there was usually only one major compound in the formulation. Table II shows for each dye formulation the retention times and visible wavelength maxima of its components. The absorbance value at each wavelength maximum is given to indicate the relative size of each dye component. For those dye formulations which showed only one peak in the chromatogram (i.e., Direct Yellow 4, Disperse Blue 27 and Reactive Orange 13), peak purity analyses done by taking spectra on the leading edge, top and tailing edge of the peaks indicated single components.

In the analysis of the textile mill wastewater treatment plant samples, four dyes were found in the influent, and none were found in the effluent. Three of the dyes found in the

influent were in the fourth cleanup fraction of the dichloromethane extract obtained under acidic conditions, which indicates that they were relatively polar. Figs. 3a-c show spectra of these dyes which had retention times of 5.55, 6.08, 6.20 min, respectively. All three dyes had absorbance maxima in the range expected for orange or red dyes. The fourth dye was found in the first cleanup fraction of the dichloromethane extract obtained under basic conditions. Its retention time was 4.51 min. Its spectrum, shown in Fig. 3d, is also consistent with those of orange or red dyes. The fact that it was found in the first cleanup fraction indicated that it was relatively non-polar. Unambiguous identification of these dyes would be extremely difficult without particle beam and thermospray mass spectra. However, the peak of retention time 6.20 in the acidic extract has been tentatively identified as Acid Red 73 based on retention time and spectral overlay matching (cf. Fig. 3c).

Within the limits of detection of the HPLC-DAD technique, the four dyes appear to be efficiently removed in the wastewater treatment plant of the textile mill since none were detected in the effluent. Further work is planned on the identification of dyes in rivers in the Eastern Townships by HPLC-MS, and their persistence and fate in aquatic environments.

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REFERENCES

1. E.A. Clarke and R. Anliker, in O. Hutzinger (Editor), Handbook of Environmental Chemistry, Vol. 3, Part A, Springer-Verlag, New York, 1980, pp. 181-215.

2. M.S. Reisch, Chem. Eng. News **66**, 7 (1988).

3. J.J. Porter, A study of the photodegradation of commercial dyes. U.S. Environmental Protection Agency Report EPA-R2-73-058, U.S. EPA Office of Research and Monitoring, Washington, 1973, 94+ pp.

4. Statistics Canada, Items 4160520-4274990, Ottawa, Ontario, Canada (1982).

5. C.E. Searle (Editor), Chemical Carcinogens, American Chemical Society Monograph Series 173, Washington, D.C., U.S.A., 1976, 788+ pp.

6. E.J. Weber, Proc. 196th Natl. Mtg. Amer. Chem. Soc., Los Angeles, Sept. 25-30, Division of Environmental Chemistry **28** (1988), 177-180.

7. W.A. Peeples, II and J.R. Heitz, J. Liq. Chromatogr. **4**, 51 (1981).

8. F.E. Lancaster and J.F. Lawrence, J. Chromatogr. **368**, 248 (1987).

9. A. Shan, D. Harbin and C.W. Jameson, J. Chromatogr. Sci. **26**, 439 (1988).

10. W.C. Tincher, Survey of the Coosa Basin for organic contaminants from carpet processing. Report E-27-630, Environmental Protection Division, Department of Natural Resources, State of Georgia, Oct. 1978, 86 pp.

11. J. Chudy, N.T. Crosby and I. Patel, J. Chromatogr. **154**, 306 (1978).

12. J.F. Lawrence, F.E. Lancaster and H.B.S. Conacher, J. Chromatogr. **210**, 168 (1981).

13. G.E. Martin, M. Tenenbaum, F. Alfonso and R.H. Dyer, J. Assoc. Off. Anal. Chem. **61**, 908 (1978).

14. M.C. Puttemans, L. Dryon and D.L. Massart, J. Assoc. Off. Anal. Chem. **64**, 1 (1981).

15. J.W.M. Wegener, H.J.M. Grunbauer, R.J. Fordham and W. Karcher, J. Liq. Chromatogr. **7**, 809 (1984).

16. H. Grossenbacher, T. Thurnheer, D. Zurrer and A.M. Cook, J. Chromatogr. **360**, 219 (1986).

17. J.P. Chaytor and R.L. Heal, J. Chromatogr. **368**, 450 (1986).

18. A.P. Bruins, L.O.G. Weidolf, J.D. Henion and W.L. Budde, Anal. Chem. **59**, 2647 (1987).

19. L. Gagliardi, A. Amata, G. Cavazzutti, D. Tonelli and L. Montanarella, J. Chromatogr. **448**, 296 (1988).

20. J.W. Wegener, J.C. Klamer, H. Govers, H. and U.A.Th. Brinkman, Chromatographia **24**, 865 (1987).

21. D.K. Laing, R. Gill, C. Blacklaws and H.M. Bickley, J. Chromatogr. **442**, 187 (1988).

22. R.M.E. Griffin, T.G. Kee and R.W. Adams, J. Chromatogr. **445**, 441 (1988).

23. P.C. White and A.-M. Harbin, Analyst **114**, 877 (1989).

24. B.M. Van Liedekerke and A.P. De Leenheer, J. Chromatogr. **528**, 155 (1990).

25. T.W. Sweatman, R.I. Larussa, R. Seshadri and M. Israel, J. Liq. Chromatogr. **10**, 1417 (1987).

26. T. Covey and J. Henion, Anal. Chem. **55**, 2275 (1983).

27. L.D. Betowski and J.M. Ballard, Anal. Chem. **56**, 2604 (1984).

28. D.A. Flory, M.M. McLean, M.L. Vestal and L.D. Betowski, Rapid Commun. Mass Spectrom. **1**, 48 (1987).

29. R.D. Voyksner, Anal. Chem. **57**, 2600 (1985).

30. L.D. Betowski and J.M. Ballard, Anal. Chem. **56**, 2604 (1984).

31. L.D. Betowski, S.M. Pyle, J.M. Ballard and G.M. Shaul, Biomed. Environ. Mass Spectrom. **14**, 343 (1987).

32. J. Yinon, T.L. Jones and L.D. Betowski, J. Chromatogr. **482**, 75 (1989).

33. J. Yinon, T.L. Jones and L.D. Betowski, Biomed. Environ. Mass Spectrom. **18**, 445 (1989).

34. J. Yinon, T.L. Jones and L.D. Betowski, Rapid Commun. Mass Spectrom. **4**, 245 (1990).

35. P.O. Edlund, E.D. Lee, J.D. Henion and W.L. Budde, Biomed. Environ. Mass Spectrom. **18**, 233 (1990).

36. W.C. Brumley, G.M. Brilis, R.J. Calvey and J.A. Sphon, Biomed. Environ. Mass Spectrom. **18**, 394 (1989).

37. M.A. McLean and R.B. Freas, Anal. Chem. **61**, 2054 (1989).

38. R.D. Voyksner, C.S. Smith and P.C. Knox, Biomed. Environ. Mass Spectrom. **19**, 523 (1990).

39. W.P. Jackson and D.W. Later, J. High Resol. Chromatogr. Chromatogr. Commun. **9**, 175 (1986).

40. E.D. Lee, W. Muck, J.D. Henion and T.R. Covey, Biomed. Environ. Mass Spectrom. **18**, 253 (1989).

41. R.J. Maguire, J.H. Carey, J.H. Hart, R.J. Tkacz and H.-B. Lee, J. Agric. Food Chem. **37**, 1153 (1989).

TABLE I

SOME DYES USED BY THE CANADIAN TEXTILE INDUSTRY*

No.	Name	CI No.	CAS No.	Chemical Formula	MW	λ_{max}, nm
1	Acid Black 52	15711	5610-64-0	$C_{48}H_{22}N_6O_{14}S_2Cr$	1022.87	568
2	Acid Orange 60	18732	30112-70-0	$C_{32}H_{30}N_{10}O_6S_2Cu$	778.33	480
3	Acid Red 73	27290	5413-75-2	$C_{22}H_{14}N_4O_7S_2Na_2$	556.49	507
4	Basic Green 4	42000	569-64-2	$C_{23}H_{25}N_2Cl$	364.92	615
5	Basic Orange 2	11270	532-82-1	$C_{12}H_{13}N_4Cl$	248.72	433
6	Basic Violet 1	42535	8004-87-3	$C_{24}H_{28}N_3Cl$	393.96	586
7	Basic Violet 3	42555	548-62-9	$C_{25}H_{30}N_3Cl$	407.99	590
8	Direct Blue 86	74180	1330-38-7	$C_{32}H_{16}N_8O_6S_2Cu$	736.20	613
9	Direct Blue 218	24401	10401-50-0	$C_{36}H_{20}N_6O_{10}S_2Cu_2$	887.80	615
10	Direct Yellow 4	24890	3051-11-4	$C_{28}H_{20}N_4O_8S_2$	580.60	402
11	Direct Yellow 11	40000	1325-37-7	undefined	n/a	419
12	Disperse Blue 26	63305	3860-63-7	$C_{16}H_{14}N_2O_4$	298.30	584
13	Disperse Blue 27	60767	15791-78-3	$C_{22}H_{16}N_2O_7$	420.38	632
14	Disperse Blue 56	63285	12217-79-7	$C_{14}H_9ClN_2O_4$	304.69	562
15	Disperse Blue 79	11345	12239-34-8	$C_{23}H_{25}BrN_6O_{10}$	639.43	546
16	Disperse Red 60	60756	17418-58-5	$C_{20}H_{13}NO_4$	331.33	541,589
17	Mordant Black 11	14645	25747-08-4	$C_{20}H_{13}N_3O_7S$	439.41	620
18	Reactive Black 5	20505	17095-24-8	$C_{26}H_{21}N_5O_{19}S_6Na_4$	995.85	595
19	Reactive Blue 19	61200	2580-78-1	$C_{22}H_{16}N_2O_{11}S_3Na_2$	628.57	592
20	Reactive Orange 13	18270	70616-89-6	$C_{24}H_{18}ClN_7O_{10}S_3Na_3$	667.02	489

*CI, CAS and MW mean Colour Index, Chemical Abstracts Service and molecular weight. The wavelength of maximal absorbance is for the region 400 - 800 nm only. Spectra were obtained in pH 7.0 phosphate buffer (I = 0.05 M) for dyes 1, 3, 9 - 12 and 18 - 20. Methanol cosolvent was required to the extent of 2% for dyes 4, 6 and 7; 10% for dyes 2, 5, 8, 13, 14 and 17; 20% for dye 16; and 40% for dye 15. Direct Yellow 11 is a stilbene self-condensation product with 2-methyl-5-nitrobenzenesulfonic acid.

TABLE II

HPLC-DAD CHARACTERIZATION OF DYES*

Dye	Retention time, min	λ_{max}, nm (absorb.)
Acid Black 52	6.00	440 (0.27)
		574 (0.38)
	6.60	580 (0.08)
	7.85	560 (0.04)
		595 (0.07)
Acid Orange 60	6.60	485 (1.20)
	8.08	485 (0.03)
	8.44	485 (0.01)
	9.00	485 (0.04)
	9.35	480 (0.04)
	10.38	485 (0.007)
Acid Red 73	6.23	518 (0.64)
	8.13	510 (0.01)
Basic Orange 2	12.90	415 (0.35)
	13.36	415 (0.35)

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TABLE II cont'd

Dye	Retention time, min	λ_{max}, nm (absorb.)
Direct Blue 86	6.56	600 (0.55)
		657 (1.70)
		680 (1.55)
	7.50	600 (0.065)
		670 (0.27)
	7.99	600 (0.02)
		670 (0.10)
	10.16	600 (0.01)
		670 (0.15)
	10.45	600 (0.01)
		670 (0.07)
Direct Blue 218	6.49	600 (0.03)
	10.00	600 (0.10)
Direct Yellow 4	6.63	405 (0.65)

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TABLE II cont'd

Dye	Retention time, min	λ_{max}, nm (absorb.)
Direct Yellow 11	5.68	415 (0.18)
	9.70	420 (0.035)
	10.58	415 (0.07)
	10.72	420 (0.10)
	14.52	410 (0.07)
	15.88	420 (0.04)
Disperse Blue 26	9.22	620 (0.03)
	9.80	575 (0.20)
		615 (0.25)
	10.40	595 (0.12)
		630 (0.12)
	13.72	605 (0.003)
Disperse Blue 27	10.40	600 (0.45)

cont'd next page

TABLE II cont'd

Dye	Retention time, min	λ_{max}, nm (absorb.)
Disperse Blue 56	7.56	590 (0.07)
		630 (0.08)
	13.46	570 (0.055)
		615 (0.065)
	14.94	575 (0.065)
		620 (0.075)
	17.14	575 (0.21)
		620 (0.23)
	19.06	585 (0.10)
		625 (0.12)
	21.68	585 (0.042)
		625 (0.043)
Disperse Blue 79	13.20	585 (0.04)
	15.38	578 (0.70)
Disperse Red 60	25.06	513 (0.14)
		550 (0.12)
	27.06	512 (0.17)
		552 (0.14)

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TABLE II cont'd

Dye	Retention time, min	λ_{max}, nm (absorb.)
Mordant Black 11	<u>7.24</u>	630 (1.75)
		650 (1.90)
		685 (1.65)
	8.13	525 (0.10)
		575 (0.11)
		680 (0.08)
	10.63	565 (0.12)
Reactive Black 5	<u>6.59</u>	480 (0.11)
		590 (0.34)
	8.72	600 (0.002)
Reactive Blue 19	<u>6.96</u>	580 (0.70)
		620 (0.70)
	7.62	580 (0.15)
		625 (0.10)
Reactive Orange 13	<u>6.92</u>	485 (0.78)
		510 (0.76)

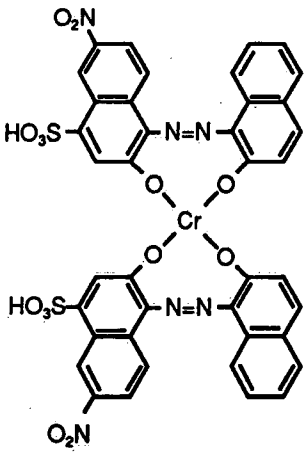
*The retention time of the major component(s) is (are) underlined. Only absorption maxima above 400 nm are shown.

FIGURE CAPTIONS

Fig. 1. Structures of dyes listed in Table I.

Fig. 2. Additional structures of dyes in Table I.

Fig. 3. Spectra of dyes of different retention times in extracts of treatment plant influent under acidic (a-c) and basic (d) conditions: (a) 5.55 min, (b) 6.08 min, (c) 6.20 min and (d) 4.51 min. Fig. 3c shows a spectral overlay with Acid Red 73, with autoscale to match at 500 nm.



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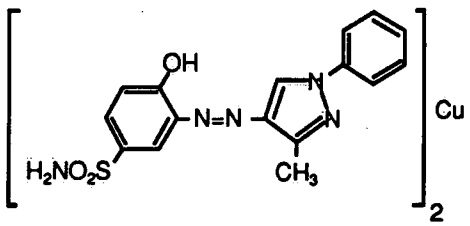

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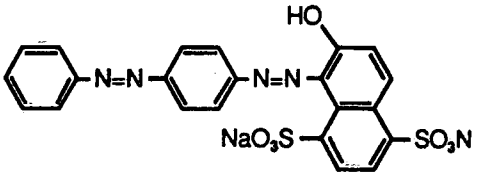
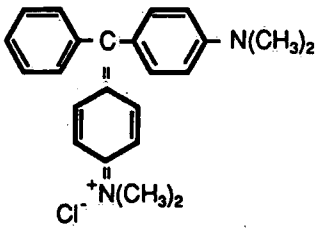
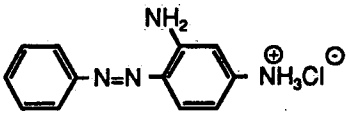


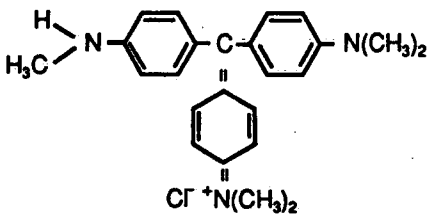
ACID RED 73



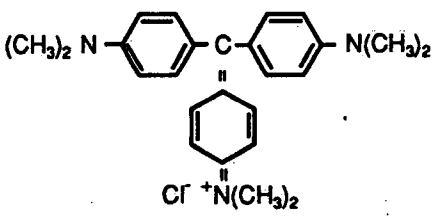
BASIC GREEN 4



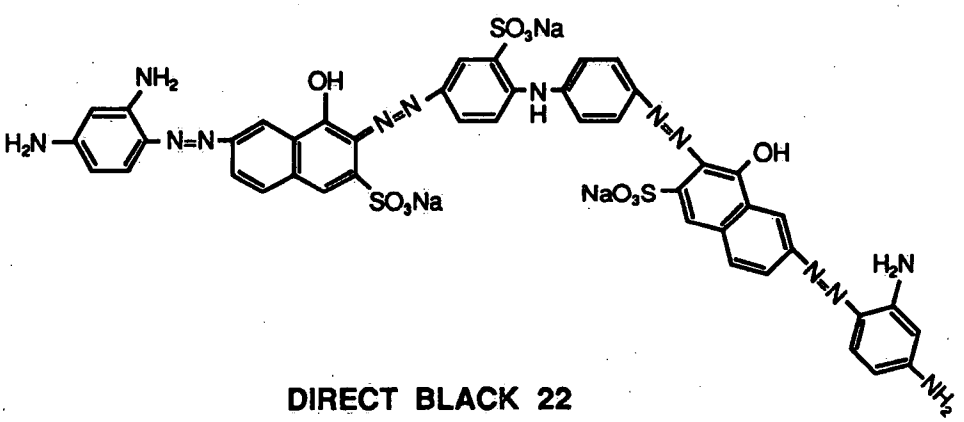
BASIC ORANGE 2



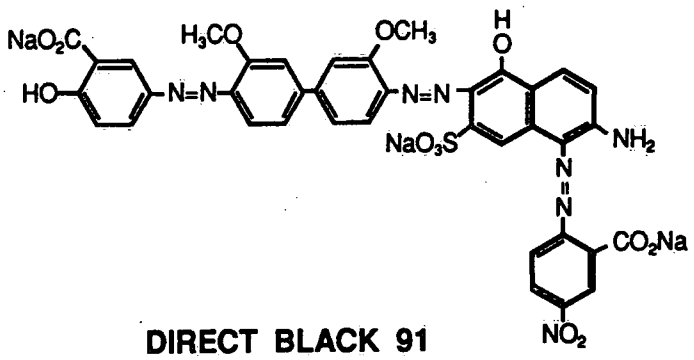
BASIC VIOLET 1



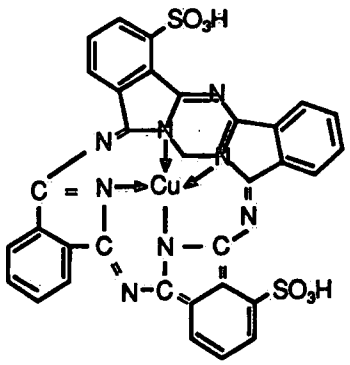
BASIC VIOLET 3



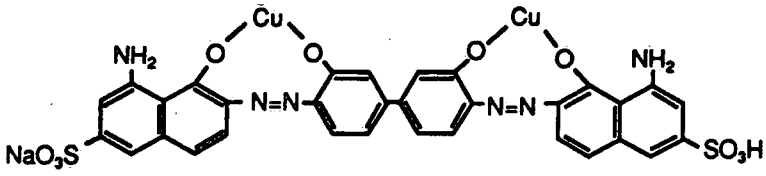
DIRECT BLACK 22



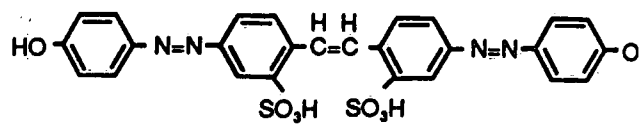
DIRECT BLACK 91



DIRECT BLUE 86

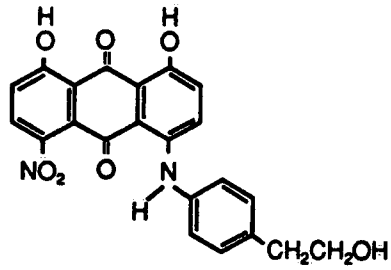


DIRECT BLUE 218

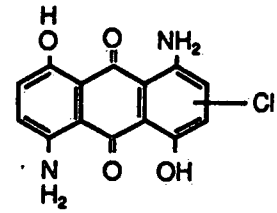


DIRECT YELLOW 4

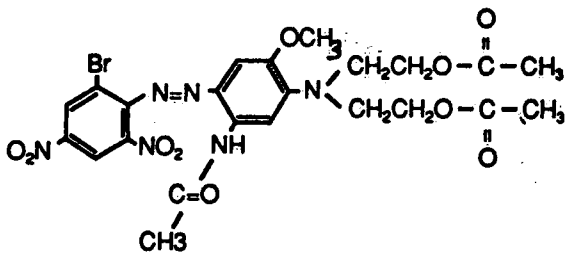
DISPERSE BLUE 26



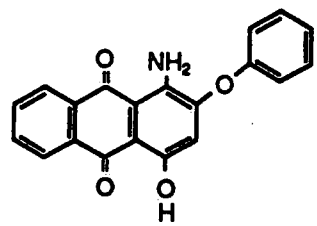
DISPERSE BLUE 27



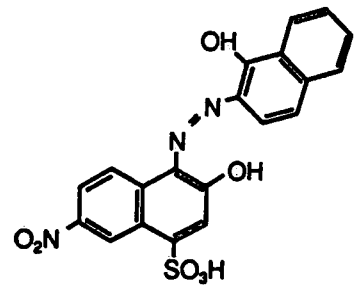
DISPERSE BLUE 56



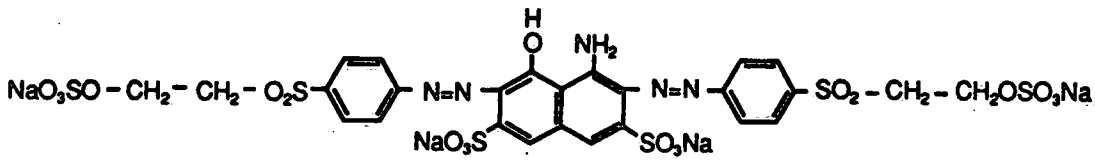
DISPERSE BLUE 79



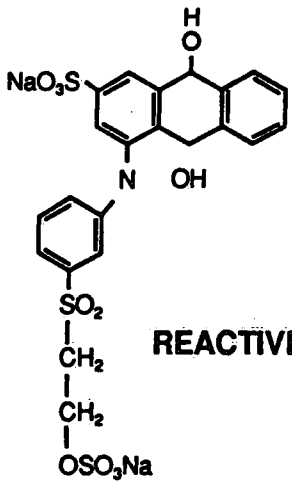
DISPERSE RED 60



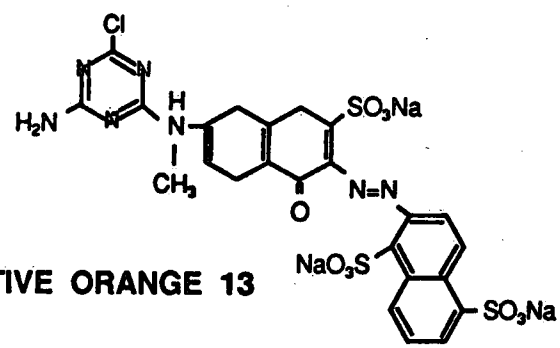
MORDANT BLACK 11



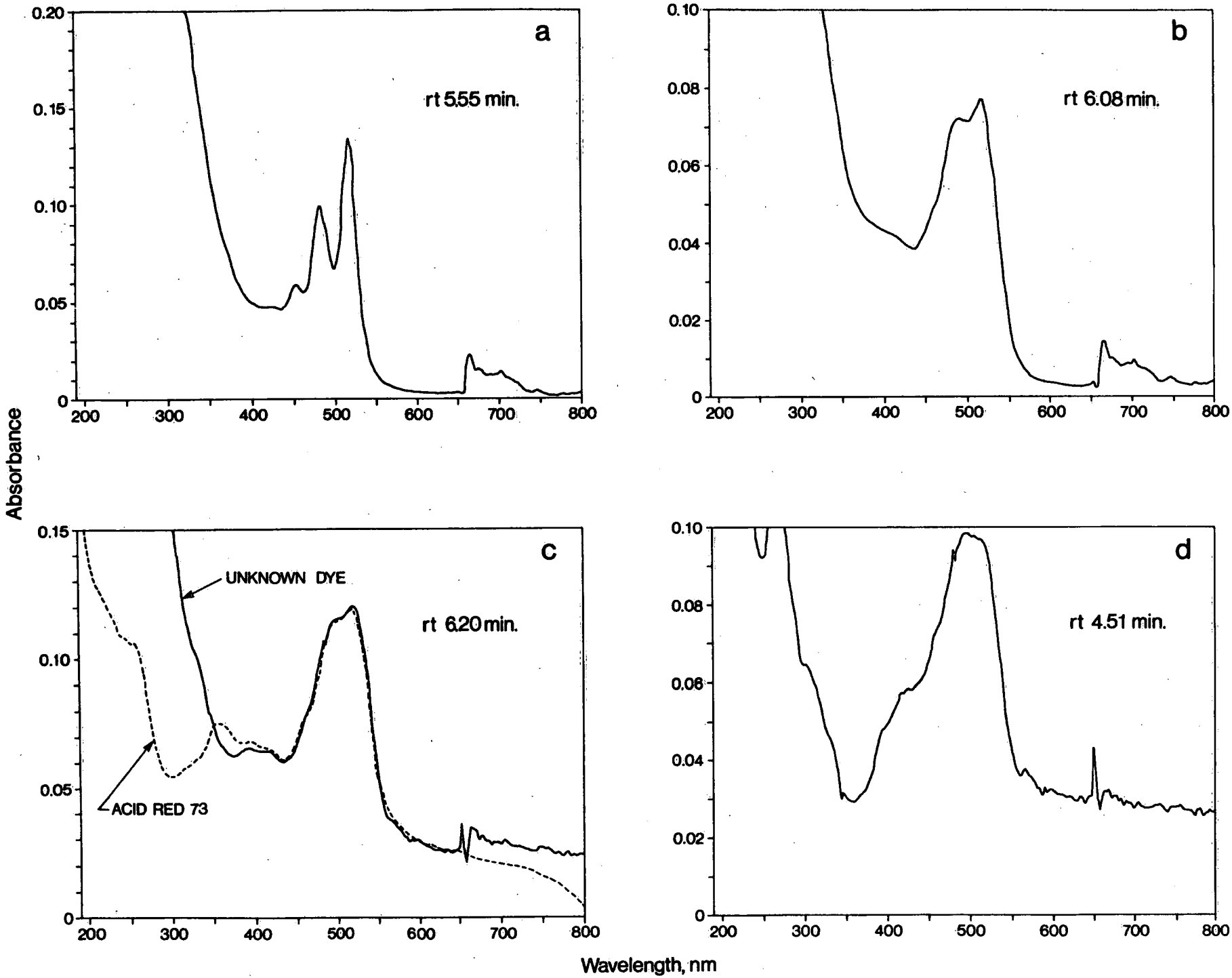
REACTIVE BLACK 5

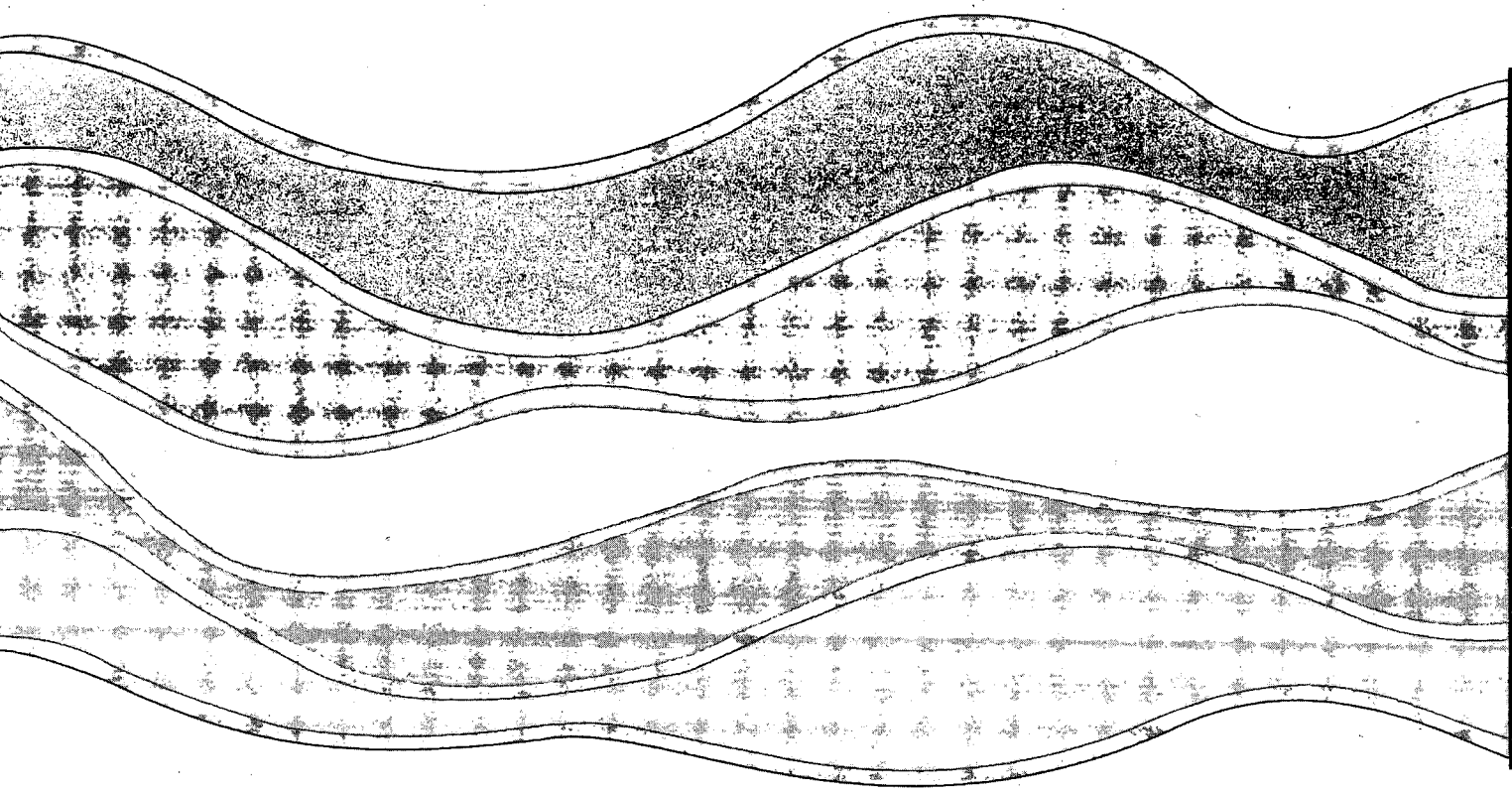


REACTIVE BLUE 19



REACTIVE ORANGE 13


a
rt 5.55 min.
b
rt 6.08 min.
c
UNKNOWN DYE
rt 6.20 min.
ACID RED 73
d
rt 4.51 min.
Absorbance
Wavelength, nm



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