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Purification Procedures for Synthetic Dyes: Part 4—Flash Chromatography

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ABSTRACT

The suitability of flash chromatography for the purification of synthetic dyestuffs has been investigated. The results of this study indicate that this technique offers a rapid procedure for the generation of gram quantities of a disperse dye, provided the dye to be purified is reasonably soluble in ordinary organic solvents such as toluene, ethyl acetate, or hexane. It is also apparent that the water-containing eluents commonly used to develop hydrophilic dyestuffs do not give satisfactory results when silica gel is used, due to strong eluent–adsorbent interactions. As a consequence, the purification of hydrophilic dyes required deactivated alumina. The speed with which a purification is accomplished by flash chromatography often offsets the higher amount of solvent required compared with the amounts used in dry column chromatography and countercurrent chromatography.

INTRODUCTION

In previous papers from these laboratories, the authors reported results from the evaluation of dry column chromatography,[1] countercurrent chromatography[2,3] and analytical HPLC[4] in the purification of both hydrophobic and hydrophilic synthetic dyestuffs. More recently, flash chromatography for the purification of disperse dyes, some hydrophilic dyes, and some novel dye intermediates was examined.

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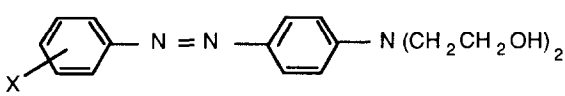
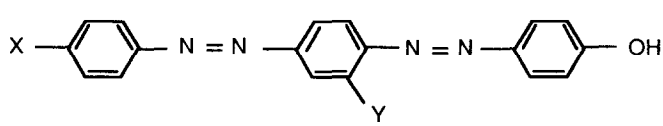
Flash chromatography (FC)[5] is a low- to medium-pressure column chromatography procedure that can be conducted using silica gel, fluorisil, or alumina as the adsorbent. Silica gel expressly for FC and special columns and accessories for FC are readily available (silica gel grade 643, 230–425 mesh, type 150A, Cat. No. S743-1, Fisher Scientific Company, Fair Lawn, NJ 07410, USA; flash chromatography column, Cat. No. Z10, 412-4, Aldrich Chemical Company, Milwaukee, WI 53233, USA). Either nitrogen or compressed air can be used to drive the eluent through a packed column. It is suggested that for optimum resolution, the solvent head above the adsorbent bed should drop 2 inches per minute (5 cm min^{-1}) during the chromatography. This means that the rate at which components are collected from the column is usually too fast to employ a fraction collector. For preparative separation of most compounds, a collection of Erlenmeyer flasks is necessary. Optimum results are also achieved when silica gel (40–63 μm) is used.

RESULTS AND DISCUSSION

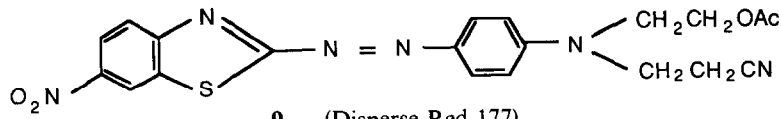
The utility of FC in the purification of synthetic dyes was determined with the aid of the disperse dyes **1–13**, the cationic dye **14**, the direct dyes **15** and **16**, and the dye intermediates **17–22**.



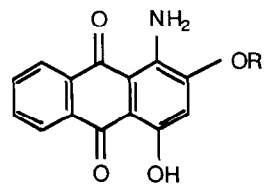
	X		*X*
1	H	**4**	2′-CN
2	2′-Me	**5**	2′-NO_2
3	2′-OMe	**6**	4′-NH_2 (Disperse Black 9 precursor)



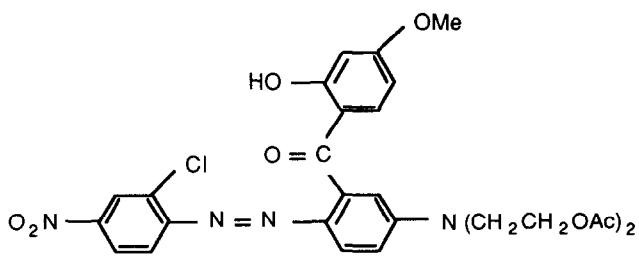
	X	*Y*
7	H	H (Disperse Yellow 23)
8	NO_2	OMe (Disperse Orange 29)



9 (Disperse Red 177)



	R
10	C_6H_5 (Disperse Red 60)
11	$(CH_2)_6OH$ (Disperse Red 91)



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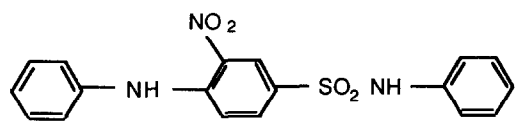
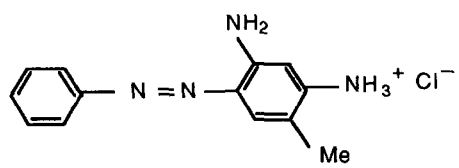
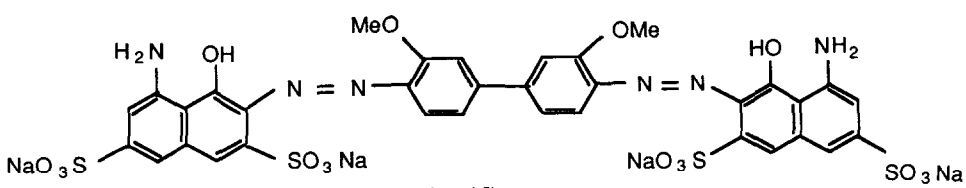


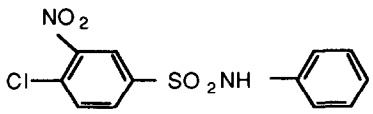
13 (Disperse Yellow 42)



14 (Basic Orange 3)



15 (Direct Blue 15)
16 (Copper complex, Direct Blue 218)



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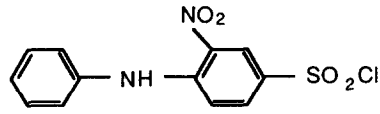

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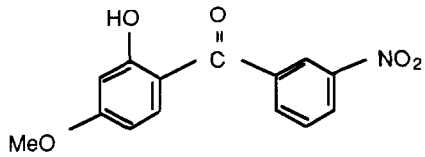

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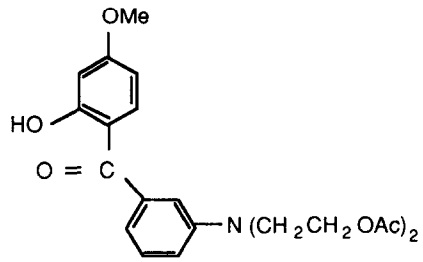

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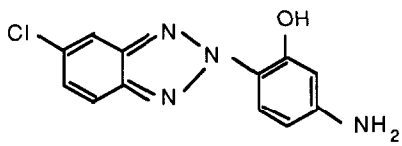

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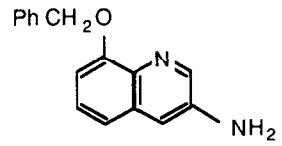

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These dyes and intermediates had been synthesized for research projects involving various aspects of the photochemistry and the genotoxicity of synthetic dyes and were found to contain up to five components by TLC, with the major component always being the desired compound.

All of the disperse dyes (**1–13**) and dye intermediates (**17–22**) were purified readily using FC. Typically, each of the crude compounds was chromatographed, using a 40:1 ratio of silica gel to compound, from start to finish in 1–2 h. In the process, 1·5–2 liters of solvent (normally a mixture of toluene and EtOAc) were required. An extreme case was the purification of dye **12** (an analog of the lightfast dye Disperse Red 167) which required 16 h and 8 liters of solvent to collect 1 g of pure dye from 2 g of a crude sample, due to both the low solubility of the dye in the eluent used and the difficulty of the separation. On the other hand, the purification of 3 g of dye **2** was complete in 30 min and required 1·5 liters of solvent to isolate 1·7 g of pure compound.

Attempts to purify the basic dye (**14**) afforded results that were unsatisfactory until this dye was first converted to the free base. The resulting dye molecule then chromatographed readily like the disperse dyes. The basic dye molecule was then regenerated upon treatment of the pure free base with anhydrous HCl. The other hydrophilic dyes in this study (dyes **15**, **16**) could be purified only in 150 mg amounts in a single pass but required a ratio of about 500:1 of adsorbent to dye. In both cases the chromatography

was quite slow, as was characteristic of purifications that employed an H_2O-containing eluent (in this case *n*-PrOH:H_2O:aq. NH_3:pyridine, 4:1:3:2 by vol.) and silica gel as the initial adsorbent. Also, the authors found that the use of a vehicle containing NH_3 led to the introduction of a small amount of silica in the solutions collected from the column. It was later found that the use of alumina deactivated to Brockman grade V allows one to avoid this contamination problem but at the expense of the superior resolution observed using silica gel.

As in cases involving other relatively simple organic molecules, the purification of the dye intermediates **17–21** proceeded quite smoothly by FC. For instance, compound **17** (10·3 g) was purified on 200 g of silica gel using 2 liters of solvent to give an 85% recovery of analytically pure product. In another case 5·5 g of analytically pure **18** were obtained from 7·0 g of a five-component crude mixture.

The authors also examined the merits of loading the crude sample onto the column as a dry silica gel powder containing the adsorbed compound ('dry loading') in comparison with loading a concentrated solution of the crude compound ('wet loading'). It was found that only those compounds which were very soluble in the eluent employed and which were devoid of impurities having $\Delta R_f \leq 0{\cdot}10$ gave satisfactory results using the 'dry loading' procedure. The 'wet loading' procedure worked better for all other compounds. The key weaknesses in the 'wet loading' procedure are: (1) the smaller quantity of purified compound produced, due to limitations in the volume of sample that could be loaded on the top of the column and give good results; and (2) if the compound was much less soluble in the eluent than in the loading solvent employed, precipitation of the compound could occur to cause blockage of the solvent flow.

Finally, to facilitate the determination of the quantity of adsorbent to use when changing the size of the chromatography column employed, the following simple formula was developed:

$$\text{Weight of adsorbent (g)} = 13{\cdot}3\ [\text{Column inner diameter (cm)}]^2$$

Thus, for a 3-cm inner-diameter column, 120 g of adsorbent was used for a relatively easy separation. This minimum amount of adsorbent was adjusted upwards as the difficulty of the separation increased.

EXPERIMENTAL

General

The ^{1}H-NMR and ^{13}C-NMR spectra were recorded on a Bruker 250 MHz spectrometer, the elemental analyses were performed by Atlantic Microlabs,

Norcross, GA, USA, and the mass spectra were recorded on either a Hewlett–Packard 5985B or JEOL HX110HF double-focusing mass spectrometer. The melting point data were recorded on a Mel-Temp melting point apparatus and are uncorrected.

Purification of the dyes and intermediates

Dye 1

A slurry of 120 g of silica gel in PhMe:EtOAc (1:2, v/v) was poured into a 3-cm inner-diameter flash chromatography column and then allowed to settle over the next 5 min. A solvent 'head' of about 1 inch (2·5 cm) was generated, and onto the top of the column was poured 10 g of silica gel onto which 3·0 g of **1** had been adsorbed. A 1-inch (2·5 cm) layer of sand was added followed by a 2-inch (5 cm) 'head' of the eluent. The solvent head was drained three to four times or until free of color (from the sample). At this point a 500 ml solvent reservoir was placed at the top of the column and filled with the eluent (PhMe:EtOAc, 1:2, v/v). Nitrogen gas was then used to produce a solvent flow rate of 50 ml min^{-1}, and 100-ml fractions were collected until TLC showed that the separation was complete. The combined fractions of pure **1** were evaporated to dryness to give 1·3 g, m.p. 134–135°C; ^{1}H-NMR ($CDCl_3$), δ (ppm): 7·76 (d,3H), 7·49 (m,3H), 6·85 (d,3H), 4·85 (t,2H), 3·57 (m,8H). Analysis: Calcd for $C_{16}H_{19}N_3O_2$: C, 67·35; H, 6·71; N, 14·73. Found C, 67·39; H, 6·76; N, 14·66%.

The procedure outlined above for dye **1** was used to purify 3 g of dyes **2–6**. The data collected for the pure compounds are reported below.

Dye 2

The yield of pure dye was 1·7 g, m.p. 106–108°C; ^{1}H-NMR ($CDCl_3$), δ (ppm): 7·76 (d,2H), 7·52 (d,1H), 7·30 (m,3H), 6·85 (d,2H), 4·82 (br s,2H), 3·58 (m,8H), 2·62 (s,3H). Analysis: Calcd for $C_{17}H_{21}N_3O_2$: C, 68·23; H, 7·02; N, 14·05. Found: C, 68·46; H, 7·15; N, 13·86%.

Dye 3

The yield of pure dye was 1·4 g, m.p. 129°C; ^{1}H-NMR ($CDCl_3$), δ (ppm): 7·19 (d,1H), 7·72 (d,2H), 7·47 (dd,1H), 7·39 (dt,1H), 6·98 (t,1H), 6·84 (d,2H), 4·83 (t,2H), 3·92 (s,3H), 3·59 (m,8H). Analysis: Calcd for $C_{17}H_{21}N_3O_3$: C, 64·75; H, 6·71; N, 13·32. Found: C, 64·53; H, 6·81; N, 13·15%.

Dye 4

The yield of pure dye was 1·2 g, m.p. 120°C; ^{1}H-NMR ($CDCl_3$), δ (ppm): 7·97 (d,1H), 7·80 (m,4H), 7·56 (dt,1H), 6·92 (d,2H), 4·87 (br s, 2H), 3·60 (br s, 8H). Analysis: Calcd for $C_{17}H_{18}N_4O_2$: C, 65·79; H, 5·85; N, 18·05. Found: C, 65·55; H, 5·89; N, 17·95%.

Dye 5
The yield of pure dye was 1·6 g, m.p. 137°C; ^{1}H-NMR ($CDCl_3$), δ (ppm): 7·98 (dd,1H), 7·72 (m,4H), 7·58 (dt,1H), 6·88 (d,2H), 4·86 (t,2H), 3·60 (m,8H). Analysis: Calcd for $C_{16}H_{18}N_4O_4$: C, 58·18; H, 5·45; N, 16·97. Found: C, 58·24; H, 5·52; N, 16·89%.

Dye 6
The yield of pure dye was 0·35 g, m.p. 156°C; ^{1}H-NMR ($CDCl_3$), δ (ppm): 7·60 (d,2H), 7·50 (d,2H), 6·80 (d,2H), 6·60 (d,2H), 5·75 (s,2H), 4·81 (t,2H), 3·57 (m,8H). The CI mass spectrum showed the $M^{\pm} - 31$ ($m/e = 269$) to be the base peak of the spectrum. The molecular ion peak ($m/e = 300$) was 39% relative intensity.

Dye 7
This dye was purified via the 'wet loading' (standard) procedure[5] in which 25 ml of a PhMe:EtOAc (10:1, v/v) solution containing 0·043 g of **7** per ml. The separation was then conducted using the same solvent system to give 0·79 g of pure **7**, m.p. 186°C; ^{1}H-NMR (DMSO-d_6), δ (ppm): 10·5 (br s,1H), 8·10 (d,2H), 8·00 (m,3H), 7·90 (d,2H), 7·70 (m,2H), 7·70 (d,2H). The CI mass spectrum of **7** showed the molecular ion to be the base peak.

Dye 8
This dye was purified according to the procedure outlined above for dye **7**. The pure compound had m.p. 232°C; ^{1}H-NMR (DMSO-d_6), δ (ppm): 8·40 (d,2H), 8·10 (d,2H), 7·85 (d,2H), 7·80 (m,3H), 6·95 (d,2H), 4·10 (d,3H). The CI mass spectrum of **8** showed the molecular ion to be the base peak.

Dye 9
The three-component mixture containing the dye (1 g) was purified according to the procedure described for **7**, using PhMe: EtOAc (1:1, v/v) as the eluent, to give 0·2 g dye having m.p. 172°C; ^{1}H-NMR (DMSO-d_6), δ (ppm): 9·08 (d, $J = 2·2$ Hz,1H), 8·33 (dd, 1H), 8·18 (d,1H), 7·92 (d,2H), 7·14 (d,2H), 4·28 (t,2H), 3·92 (dt,4H), 2·89 (t,2H), 2·00 (s,3H). The pure compound had $R_f = 0·4$ on silica gel using PhMe:EtOAc (1:1, v/v).

Dye 10
This dye was purified via the 'dry loading' technique described for the purification of dyes **1**–**6**. In this specific example, 2·24 g of crude **10** were adsorbed onto 11 g of silica gel, loaded on top of a PhMe/silica gel bed, and the resulting sample was chromatographed using PhMe to give 1·8 g of pure dye, m.p. 182°C; ^{1}H-NMR (DMSO-d_6), δ (ppm): 8·22 (m,2H), 7·86 (m,2H), 7·56 (m,2H), 7·35 (m,3H), 6·19 (s,1H). ^{13}C-NMR (DMSO-d_6), δ (ppm): 184·6,

182·0, 158·6, 155·5, 153·3, 140·0, 134·4, 134·0, 133·0, 132·5, 130·6, 126·6, 126·1, 125·8, 120·9, 108·5, 108·0, 107·6.

Dye 11

This dye (0·3 g) was chromatographed on a 2-inch (5 cm) diameter column using PhMe:EtOAc (2:1, v/v) to give 0·16 g of pure dye, m.p. 155°C (dec.), $R_f = 0{\cdot}27$; ^{1}H-NMR (DMSO-d_6), δ (ppm): 8·25 (m,2H), 6·82 (s,1H), 4·40 (t,1H), 4·15 (m,4H), 3·40 (m,4H), 1·45 (m,4H). ^{13}C-NMR (DMSO-d_6), δ (ppm): 183·2, 181·1, 160·7, 155·8, 140·0, 134·4, 133·6, 132·7, 132·6, 126·2, 125·6, 106·2, 106·1, 104·8, 69·4, 60·6, 32·4, 29·0, 28·2, 25·2.

Dye 12

Crude **12** (2·0 g) was purified on a 3-cm diameter column, using the procedure described above for dye **10** and PhMe:EtOAc (4:1, v/v), to give 0·98 g of pure dye, m.p. 160–163°C, $R_f = 0{\cdot}35$; ^{1}H-NMR (DMSO-d_6), δ (ppm): 12·36 (s,1H), 8·33 (d, $J = 2{\cdot}3$ Hz,1H), 8·13 (dd,1H), 7·98 (d,1H), 7·37 (d,1H), 7·18 (m,1H), 7·08 (d,1H), 7·00 (d, $J = 2{\cdot}5$Hz,1H), 6·54 (d, $J =$ 2·4 Hz,1H), 6·38 (dd,1H).

Dye 13 and Compound 18

A mixture (8 g) containing **13**, **18**, and two other components was separated using a 3-inch (7·5 cm) diameter column, and the 'dry loading' procedure. The use of cyclohexane:EtOAc (10:1, v/v) afforded 2·5 g of pure **18**, m.p. 93–94°C, $R_f = 0{\cdot}52$; ^{1}H-NMR ($CDCl_3$), δ (ppm): 8·40 (d, $J = 2{\cdot}0$ Hz,1H), 7·70 (dd,1H), 7·39 (m,4H), 7·24 (m,2H). When the eluent was changed to cyclohexane:EtOAc (4:1, v/v), 1·3 g of pure **13** were obtained, m.p. 154–156°C (lit.[6] 157°C); $R_f = 0{\cdot}28$; ^{1}H-NMR ($CDCl_3$), δ (ppm): 10·34 (s,1H), 9·85 (s,1H), 8·49 (d, $J = 2{\cdot}2$ Hz,1H), 7·73 (dd,1H), 7·43 (t,2H), 7·28 (m,5H), 7·16 (dd,3H), 7·13 (t,1H).

Dye 14

The free base of this dye was liberated into EtOAc with the aid of 10% $NaHCO_3$. The crude free base (3·8 g) was chromatographed using the 'dry loading' procedure to give 2·45 g of pure compound; ^{1}H-NMR (DMSO-d_6), δ (ppm): 8·26 (d,1H), 7·92 (d,2H), 7·44 (t,2H), 7·15 (t,1H), 6·52 (d,1H), 5·92 (s,1H). This material was converted to the HCl salt using anhydrous HCl to give pure **14** which decomposed without melting at 200°C and had $R_f = 0{\cdot}59$.

Dye 16

When 1 g of crude **16** was chromatographed on 150 g of deactivated alumina (Brockman activity V) using the 'dry loading' procedure and *n*-PrOH:H_2O:NH_4OH:pyridine (4:1:3:2, by vol.) there was obtained 0·16 g of dye ($R_f =$

0·63) that contained a trace impurity at the origin. Repeated attempts to get a purer sample were unsuccessful.

Compound 17
This intermediate was purified using PhMe: cyclohexane (1:1, v/v) and the procedure outlined for dye **1**. From 10 g of crude material were obtained 7·5 g of pure **17**, m.p. 80–82°C, $R_f = 0{\cdot}45$ [PhMe:EtOAc, 4:1 (v/v)]; ^{1}H-NMR ($CDCl_3$), δ (ppm): 8·25 (d, $J = 2{\cdot}0$ Hz,1H), 7·94 (dd,1H), 7·74 (d,1H), 6·96 (t,2H), 6·84 (d,2H), 6·59 (t,1H). Analysis: Calcd for $C_{12}H_9N_2O_4SCl$: C, 46·08; H, 2·92; N, 8·90. Found: C, 45·96; H, 2·92; N, 8·90%.

Compound 19
Using the procedure outlined for dye **1**, a 3-inch (7·5 cm) column, and PhMe as the eluent, 7·0 g of crude sample were purified to give 5·5 g of pure compound, m.p. 96–98°C, $R_f = 0{\cdot}75$ [PhMe:EtOAc, 4:1 (v/v)]: ^{1}H-NMR ($CDCl_3$), δ (ppm): 12·34 (s,1H), 8·44 (d, $J = 1{\cdot}7$ Hz,1H), 8·37 (dd,1H), 7·93 (dd,1H), 7·68 (t,1H), 7·35 (d,1H), 6·5 (d, $J = 2{\cdot}5$ Hz,1H), 6·41 (dd,1H), 3·85 (s,3H).

Compound 20
Using a 2-inch (5 cm) diameter column and PhMe:EtOAc, 1:1 (v/v), 4·5 g of crude **20**, an extremely viscous oil was obtained, $R_f = 0{\cdot}20$ [PhMe:EtOAc, 4:1 (v/v)]. ^{1}H-NMR ($CDCl_3$), δ (ppm): 12·32 (br s,1H), 7·51 (d,1H), 7·33 (m,2H), 7·02 (d,1H), 6·85 (d,1H), 6·58 (d, $J = 2{\cdot}3$ Hz,1H), 6·51 (dd,1H), 4·19 (t,4H), 3·84 (s,3H), 3·65 (t,4H), 1·98 (s,3H).

Compound 21
This compound (7·0 g) was purified on a 3-inch (7·5 cm) diameter column using the 'dry loading' procedure and PhMe:EtOAc, 10:1 (v/v), initially and then [PhMe:EtOAc, 4:1 (v/v)] to complete the isolation of product. This process afforded 5·0 g of pure **21**, m.p. 186–188°C, $R_f = 0{\cdot}42$; ^{1}H-NMR (DMSO-d_6), δ (ppm): 8·07 (d, $J = 2{\cdot}1$ Hz,1H), 7·98 (d,1H), 7·54 (d,1H), 7·42 (dd,1H), 6·30 (d, $J = 2{\cdot}1$ Hz,1H), 6·25 (dd,1H). CI mass spectrometry afforded the molecular ion as the base peak of the spectrum.

Compound 22
This quinoline amine (0·4 g) was purified on a 2-inch (5 cm) diameter column using the 'wet loading' procedure described for the purification of dye **1** and using PhMe:EtOAc, 2:3 (v/v) to give 0·28 g of **22**, m.p. 145–147°C, $R_f = 0{\cdot}28$; ^{1}H-NMR (DMSO-d_6), δ (ppm): 8·38 (s,1H), 7·54–7·12 (m,8H), 6·84 (d,1H), 5·67 (s,2H), 5·21 (s,2H). The molecular ion was the base peak of the CI mass spectrum.

CONCLUSIONS

Like dry column chromatography, flash chromatography works best when disperse dyes are undergoing purification. The main advantage of this procedure over dry column chromatography and other techniques such as preparative layer chromatography is the speed with which gram quantities of pure dyestuffs are obtained. The main disadvantages of this procedure are (1) the cost of obtaining multiple column set ups, and (2) the requirement for eluents that allow a high eluent flow ratio (i.e. low-viscosity solvent systems). The latter requirement makes the purification of most hydrophilic dyes a difficult task on silica gel, in that the eluents often preferred for such separations normally do not afford an acceptable flow rate.

REFERENCES

1. Freeman, H. S., Williard, C. S. & Hsu, W. N., *Dyes and Pigments*, **7**(6) (1986) 397.
2. Freeman, H. S. & Williard, C. S., *Dyes and Pigments*, **7**(6) (1986) 407.
3. Freeman, H. S., Hao, Z., Mills, K. P. & McIntosh, S. A., *J. Liq. Chromatogr.*, **11**(1) (1988) 251.
4. Mills, K. P., Freeman, H. S., Whaley, W. M. & Carroll, F. I., *Dyes and Pigments*, **8**(5) (1987) 389.
5. Still, W. C., Kahn, M. & Mitro, A., *J. Org. Chem.*, **43** (1978) 2923.
6. Fischer, P., *Ber.*, **24** (1891) 3794.