The Identification of EEC Food Colours

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R_f values (using paper chromatography) of colours included in the EEC Directive and due to be added to the U.K. permitted list in July 1974 are tabulated with respect to 12 solvents. The principal spectrophotometric peaks and other properties of the colours are also considered.

On 27th September, 1972 the Ministry of Agriculture, Fisheries and Food announced proposals designed to harmonise United Kingdom regulations[1] and the European Economic Community (EEC) Council Directive on colours for human food. Accordingly it was proposed—

(*a*) to remove 7 colouring matters from the list of permitted colours, namely, Red 10B, Red 6B, Fast Red E, Oil Yellow XP, Oil Yellow GG, Red FB and Violet BNP;

(*b*) to add 9 new dyes to the list of permitted colours, namely, Indanthrene Blue, Patent Blue V, Quinoline Yellow, Fast Yellow AB, Brilliant Blue FCF, Violet 6B, Pigment Rubine and Burnt Umber (for certain cheese rinds only), and Methyl Violet (only for marking the skins of citrus fruit and for marking raw and unprocessed meat). Definitions of each of these colours are given in Table I. Subsequently it was decided to defer the introduction of Violet 6B pending further studies on its safety in use.

The R_f values obtained using paper chromatography for the currently-permitted dyes (using 12 solvents) and for some of the EEC colours (using 7 solvents) have been previously tabulated[2,3]. It would now be useful to have available a Table covering spectrophotometric data and R_f values employing all 12 solvents for the colours to be added to the U.K. permitted list from July 1, 1974[4] together with Violet 6B. New samples of the EEC colours were supplied by two manufacturers for the tests.

Experimental

PAPER CHROMATOGRAPHY

The paper chromatography was carried out in one litre beakers as previously described[5]. The various solvents used were as follows:

These should be freshly prepared.

1. Ammonia (S.G. 0·880)/water (1:99).
2. Aqueous sodium chloride (2·5 per cent. w/v).
3. Sodium chloride in 50 per cent. ethanol (2 per cent. w/v).
4. *iso*-Butanol/ethanol/water (1:2:1).

TABLE I

DEFINITIONS OF EEC COLOURS PROPOSED FOR ADDITION TO THE U.K. LIST

Serial No.	Name of Colour	Colour Index Number	Scientific Name or Description
E 104	Quinoline Yellow	47005	sodium salt of a mixture of the mono- and disulphonic acids (mainly the latter) of quinophthalone or 2-(2-quinolyl)indane-dione
E 105	Fast Yellow AB	13015	disodium 2-amino-5-(4-sulphophenylazo) benzenesulphonate
E 130	Solanthrene blue RS or Anthragen Blue, or Indanthrene Blue	69800	6;15-dihydro-5;9;14;18-anthrazinetetrone (Indanthrone)
E 131	Patent Blue V	42051	calcium di-4-[4-diethylammoniocyclohexa-2;5-dienylidene-(4-diethylaminophenyl) methyl]-6-hydroxybenzene-1;3-disulphonate
—	Brilliant Blue FCF	42090	disodium 4′;4″-di(*N*-ethyl-3-sulphonatoben-zylamino)triphenylmethylium-2-sulphonate
—	Violet 6B*	42640	monosodium salt of *N,N′*-(4″ dimethyl-aminotriphenyl-methylio)-4;4′-di-(*N*-ethylaminomethylbenzene-4-sulphonic acid)
E 180	Pigment Rubine or Lithol Rubine BK (for colouring rinds of hard cheese only)	15850	only the calcium and aluminium salts of 3-hydroxy-4-(2-sulpho-*p*-tolylazo)-2-naphthoic acid
E 181	Burnt umber (for colouring rinds of hard cheese only)	—	product obtained by roasting in air a mixture consisting essentially of iron and manganese oxides, and calcium and aluminium silicates, carbonates and sulphates
—	Methyl violet (for marking citrus fruit and raw and unprocessed meat only)	42535	mixture of the hydrochlorides of the more highly methylated pararosanilines containing principally the *N*-tetra, penta-, and hexa-methyl derivatives

* Introduction of Violet 6B deferred pending further study of its safety in use.

5. *n*-Butanol/water/glacial acetic acid (20:12:5).
6. *iso*-Butanol/ethanol/water (3:2:2). Then to 99 ml of the solution add 1 ml of ammonia solution (S.G. 0·880).
7. 80 g of Phenol + 20 g of water.
8. Ethyl methyl ketone/acetone/water/ammonia (S.G. 0·880) (350:150:150:1).
9. Ethyl methyl ketone/acetone/water (7:3:3).
10. Ethyl acetate/pyridine/water (11:5:4).
11. Dilute 5 ml of ammonia (S.G. 0·880) to 100 ml with water and dissolve 2 g of trisodium citrate in the solution.
12. Concentrated hydrochloric acid (S.G. 1·18)/water (6·5:30).

With the more compact spots the R_f value was taken as the bisector, or mean values of the leading and trailing edges. With long streaks the values for both the leading and trailing edges were recorded.

TABLE II

R_f VALUES AND MAIN SPECTROPHOTOMETRIC PEAKS OF VARIOUS EEC COLOURS

	Solvent Numbers (*see text*)												
	1	2	3	4	5	6	7	8	9	10	11	12	Main spectrophotometric peaks *nm*
Quinoline Yellow	0·00–1·00	0·00–0·76	0·00–0·64	0·29,0·50	0·20,0·42	0·20,0·54	0·42,0·72	0·24,0·64	0·29,0·69	0·18,0·51	0·00–0·72	0·00–0·67	410 in acid
Fast Yellow AB	0·90	0·56	0·74	0·40	0·25	0·42	0·24	0·37	0·34	0·22	0·50	0·61	385 in alkali
Solanthrene Blue RS, or Anthragen Blue, or Indanthrene Blue	0·00	0·00	0·00	0·00	0·00	0·00	0·00	0·00	0·00	0·00	0·00	0·00	—
Patent Blue V	0·95	0·79	1·00	0·77	0·57	0·60	0·85	0·62	0·65	0·41	0·97	1·00	625 in alkali
Brilliant Blue FCF	1·00	1·00	1·00	0·54	0·44	0·48	0·66	0·35	0·35	0·18	0·95	1·00	625 in alkali
Violet 6B	0·90	0·00–0·83	1·00	0·76	0·54	0·63	1·00	0·60	0·57	0·44	0·00–0·82	1·00	540 in neutral
Methyl Violet	0·11	0·00 (0·00–0·22)	1·00	1·00	1·00	1·00	1·00	1·00	1·00	0·67	0·14	1·00	590 in neutral
Pigment Rubine, or Lithol Rubine BK	0·00 (0·00–0·47)	0·00 (0·00–0·15)	0·00 (0·00–065)	0·00–0·86	0·00–0·70	0·00 (0·00–0·62)	0·00 (0·00–0·79)	0·00 (0·00–0·67)	0·00	0·00 (0·62)	0·14	0·00	—

SPECTROPHOTOMETRY

For the spectrophotometry the dyes were examined (as appropriate) in neutral solution (0·02 per cent. ammonium acetate), in acid (0·1 N HCl) and in alkaline solution (0·1 N NaOH).

FLUORESCENCE AND EFFECT OF ADDING ACID AND ALKALI TO SPOTS

The separated dried spots were also examined for fluorescence under an ultra-violet lamp and for any colour changes on addition of 10 per cent. NaOH and conc. HCl.

Results and Discussion

The R_f values and spectrophotometric data obtained with the EEC colours examined are given in Table II. It should be borne in mind that R_f values are influenced by factors such as the exact composition, pH and the age of the solvent, the quality of the paper, concentration of the dye and the temperature. It is important therefore always to compare any unknown dye with a set of known dyes on the same paper and to rely on tabulated R_f values only for selecting the best solvent to give a good separation between the expected spots.

On the basis of the R_f values in Table II and those previously tabulated[2], the following solvents are apparently the most suitable for distinguishing between the various groups of colours (including those currently permitted in the U.K.):

Colour Group	Recommended Solvents
Reds	3, 6, 11
Yellows/Oranges	8, 11, 12
Blues/Violets/Greens	6, 9, 11
Browns	4, 8, 9
Blacks	9

TABLE III

EFFECT ON SPOTS OF EEC COLOURS OF UV LIGHT AND THE ADDITION OF ACID AND ALKALI

		Spot Tests		
	Fluorescence	Dry	Sodium hydroxide (10 per cent.)	Hydrochloric acid (concentrated)
Quinoline Yellow	—	Yellow	Pale brownish yellow	No change
Fast Yellow AB	—	Bright yellow	No change	Bright red
Patent Blue V	—	Turquoise blue	Blue	Very pale yellow
Brilliant Blue FCF	—	Turquoise blue	Pink	Pale yellow
Violet 6B	—	Violet	Almost colourless	Colourless
Methyl Violet	—	Violet	No change	No change
Pigment Rubine	Pink (with HCl)	Pinky red	Brick red	No change

The spectrophotometric peaks of the water-soluble organic dyes in aqueous solution are also given in Table II. The corresponding curves from which they are derived are shown in Figs. 1–6.

The effects of u.v. light and the addition of acid and alkali are given in Table III.

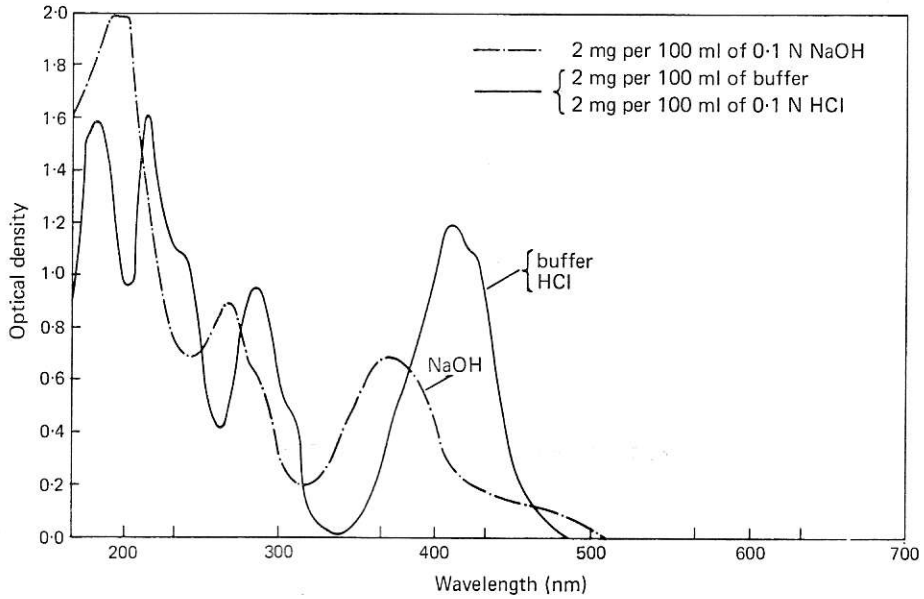

FIG. 1. Absorption curve for Quinoline Yellow.

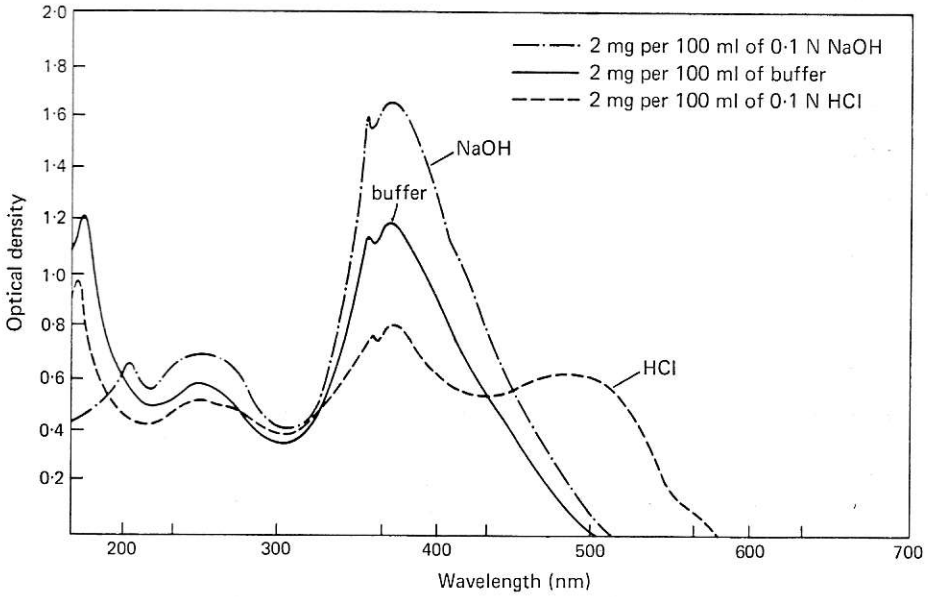

FIG. 2. Absorption curve for Fast Yellow AB.

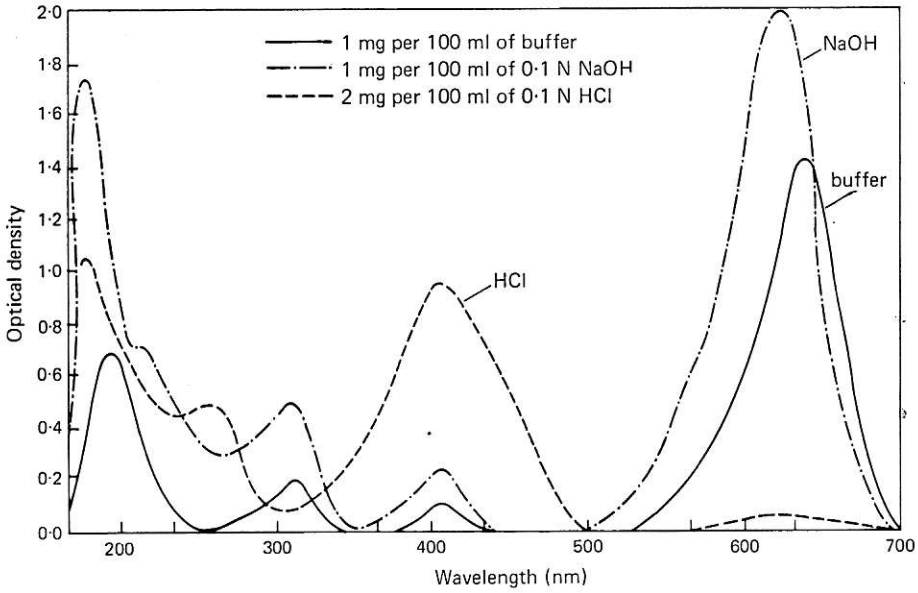

FIG. 3. Absorption curve for Patent Blue V.

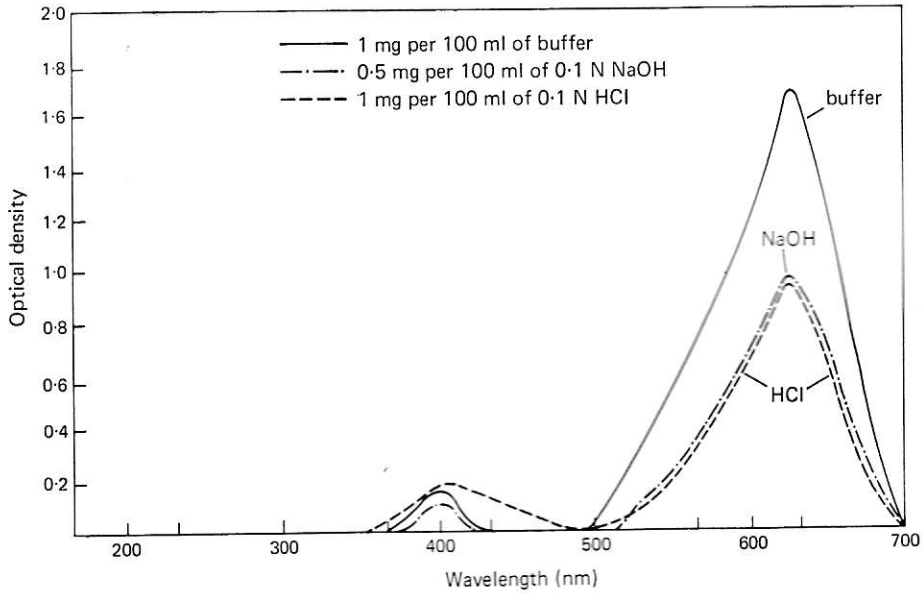

FIG. 4. Absorption curve for Brilliant Blue FCF.

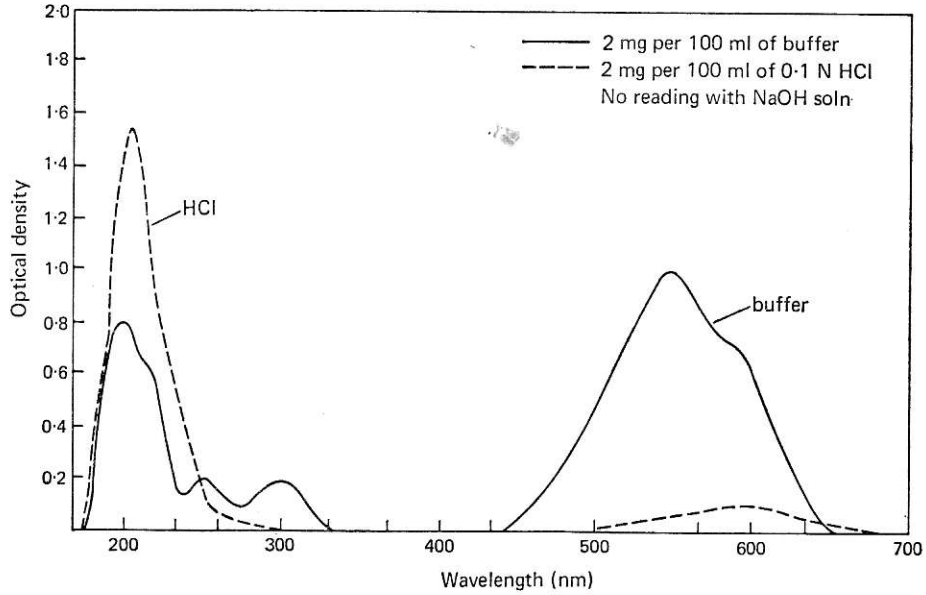

FIG. 5. Absorption curve for Violet 6B.

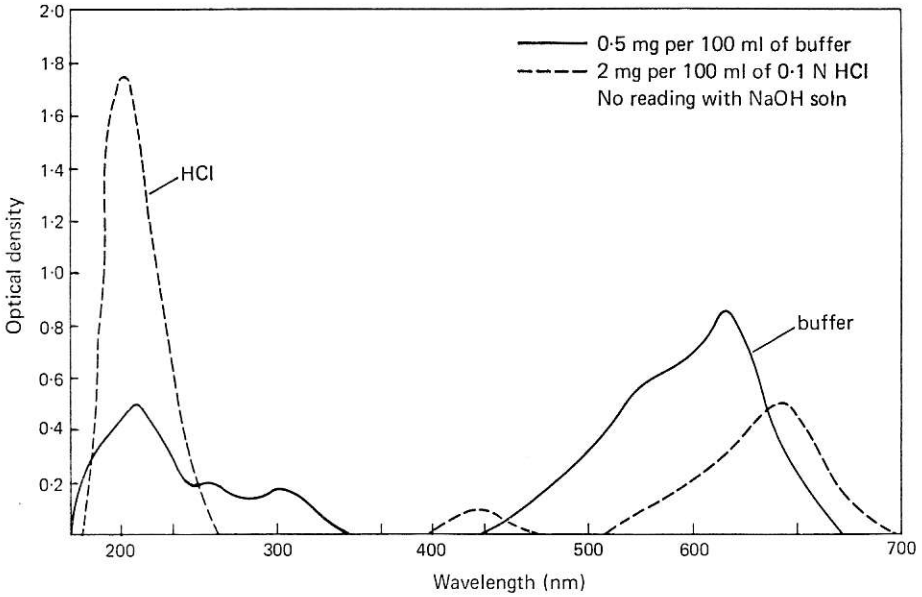

FIG. 6. Absorption curve for Methyl Violet.

Although the tests were performed in most cases on duplicate samples from different sources some doubt must exist regarding the complete authenticity and purity of some of the dyes used for the tests. For example, the 6 separate samples of quinoline yellow examined appeared to contain markedly differing proportions of the mono- and disulphonates (see Table I). Also, difficulties arise in identifying indanthrene blue due to its general insolubility.

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References

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