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LIGHT ABSORPTION PATTERNS OF INTACT *ROSA* FLOWERS IN RELATION TO THE FLOWER COLOUR*

MASATO YOKOI

Faculty of Horticulture, Chiba University, Matsudo, Chiba Prefecture, Japan

and

NORIO SAITÔ

Chemical Laboratory, Meiji-Gakuin University, Tokyo, Japan

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Abstract—Absorption curves of fresh, intact petals from 18 rose cultivars and 2 species were measured and compared with visual evaluations of their colours and there was a reasonable correlation. The *in vivo* maxima of anthocyanin absorption were in the range of 520–560 nm. Five patterns of absorption spectrum in the visible region were recognized: (a) maximum range *ca.* 520–535 nm (red roses); (b) as (a) but low absorbance (pink roses); (c) absorption pattern varying with age of flowers; (d) absorption at long wavelengths in blue roses due to co-pigmentation of cyanin, flavonols; (e) absorption of carotenoids and anthocyanins together in yellow, orange or orange red flowers.

INTRODUCTION

IN AN earlier paper,[1] it was demonstrated that the pattern of absorption spectra of intact, anthocyanin-containing flowers fell into 3 groups, confirming the isolation studies of Hayashi *et al.* of genuine anthocyanins.[2–10] More recently, similar observations were noted in the case of intact petals of roses.[11]

It is well known that there are great many colour variations in rose cultivars, although there are only a few anthocyanins, flavonols, and carotenoids present.[12–18] To explain this

* Part I in the projected series "Colour and Pigment Distribution in Ornamental Plants".

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paradox, the relationships between the absorption curves of the intact tissues of rose cultivars and the visual evaluations of the colour quality were determined with the help of a colour chart and colorimetric designation.

RESULTS AND DISCUSSION

The absorption curves of fresh petals of 18 rose cultivars and 2 species were measured and the results are given in Table 1 together with colour and pigment data. There was a reasonable correlation, except between the purity obtained from the C.I.E. diagram and the absorbance of the fresh petals, especially for flowers having strong spectral absorption.

By measuring anthocyanin content in intact petals, Stewart *et al.*[11] showed that there was a good correlation between absorbance and anthocyanin content. In this paper, similar results were obtained, using visual estimation of anthocyanin content from chromatograms.

The variation in colour of the petals from orange to red (code number of RHS Colour Chart 24–56)[19,20] was dependent on the anthocyanidins present. For example, code number of RHS Colour Chart increased with the increase in amount of cyanidin. On the other hand, the code number decreased with the increase in pelargonidin concentration. However, in the case of the cultivars, Charleston, Brilliant Light, Tzigane and Wiener Charm which all lacked pelargonidin pigments, the carotenoids affected their colours instead (Table 1).

TABLE 1. COLOUR AND PIGMENT DATA

Rose cultivars and species	Colour*	Colour group in RHS Colour Chart†	RHS Colour Chart code number†	Dominant wavelength (nm)‡	Purity (%)‡
Wiener Charme	Coppery orange	Orange	25 A	588	79
Charleston (young)	Yellow flushed crimson, becoming crimson	Orange, red	25 C, 43 B	585, 606	63, 72
Zorina	Grenadine-red	Red	41 A	614	70
Duftwolke	Coral red becoming geranium red	Red	43 B, C	607·5, 611·5	70, 52
Brilliant Light	Bright red	Red	45 B	616	77
Pharaon	Geranium-red to salmon coral	Red	46 B, A	612–620	62–74
Charleston (old)	Crimson	Red	46 B	612	74
Tzigane	Rose-red, reverse yellow	Red, yellow–orange	46 D, 21 C	494, 591	64, 62
Sea Pearl	Soft pink	Red	48 C	611	30·5
South Seas	Coral-pink	Red	50 A	621	52
Pink Star	Deep pink	Red	52 A	642	44
Americana	Bright red	Red	53 A	633	50
Josephine Bruce	Crimson	Red	53 A	636	52
Spanish Beauty	Delicate pink	Red	55 A	−495	35
Haru-no-brûsu	Pink	Red	55 C	−495	14
François Juranville	Bright salmon-pink	Red	56 A	−496	8
Isabel de Ortiz	Deep pink	Red–purple	57 D	−497	44
Rosa gallica var. *versicolor*	Pink and red	Red–purple	66 A, C	−497·2, −498·0	65, 35
Rosa rugosa	Purple	Red–purple	67 C	−501·5	44
Blue Girl (old)	Silvery lilac-blue	Purple	75 C	−548	13
Sterling Silver	Lilac	Purple	75 C, D	−499	5

* Modern Roses 6 and 7 (1965, 1969).
† R.H.S. Colour Chart (1966).
‡ Obtained from C.I.E. diagram.
¶ + = trace; ++ = low; +++ = intermediate; +++++ = high.

[19] *Royal Horticultural Society Colour Chart* (1966) London.
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In the cultivar Charleston, the absorption pattern of the petals changed with age (Table 1). In young flowers, carotenoid absorption was greater than anthocyanin absorption, whereas the reverse was true in the older, crimson flowers.

In cultivars with red to red–purple flowers, chrysanthemin occurred in the flowers with relatively smaller code numbers of the RHS Colour Chart, and it was not found in the flowers with the larger code numbers. The anthocyanidins are cyanidin and peonidin. The major anthocyanin in the wild rose, *Rosa rugosa*, is peonin[13,15,16] and its λ_{max} is at a slightly longer wavelength than that of a typical red rose containing cyanidin glycosides.

The pigment of the 'so-called' blue roses, Blue Girl and Sterling Silver, is cyanin and yet their λ_{max} are at 548 and 563 nm, instead of at 532 nm, as in the case of the cultivar Americana which also contains cyanin. Moreover, there is an intense absorption band near 380 nm in the blue roses. Thus, large amounts of the flavonols kaempferol and quercetin are present in the petals, a fact confirmed by PC and TLC. Harborne[13,14] earlier suggested the presence of co-pigmentation in mauve or purple roses. Yazaki *et al.*,[21] and Asen *et al.*[12,27] have also shown that the co-occurrence of anthocyanins and flavonols produces a shift of the λ_{max} to longer region, due to co-pigmentation. Indeed, the absorption curve of the blue roses exhibits a distinct shoulder at 600–650 nm.

FOR 18 ROSE CULTIVARS AND 2 SPECIES

Absorbance from intact fresh petal			Visua lrating of anthocyanin concentration on chromatogram					Occurrence of carotenoids
max (nm)	Absorbance	Half width band at 535 nm	Cal	Pel	Peo	Chr	Cy§	
447–8, 480, 535	1·16, 1·05, 0·48	50					+++¶	C
422, 445, 478, 499, 520	0·96, 0·96, 0·80, 0·88, 0·85	58				+++	+++	C
510	1·20	45	+++	+++			++	
520	1·31	36	+++	+++		+++	+++	
422, 532	1·20, 2·11	36				++++	++++	C
527	2·77	42	++	++++		+	++++	
445, 528	1·40, 1·91	56				++++	+++	C
456, 490, 545	0·68, 0·75, 0·80	35					+++	C
525	0·20	35	++	++		++	++	
525	1·82	40	+++	+++			++	
531	1·55	37				+	+++	
532	3·89	35					++++	
535	2·85	42	+			++	++++	
540	0·54	41				++	++	
535	0·29	50		+			++	
540	0·14	50					+	
535	1·39	36					+++	
545	1·13	47					+++	
543	0·94	49			+++		++	
548	0·53	80					++	
563	0·62	83					++	

§ Cal = callistephin (pelargonidin 3-glucoside), pel = pelargonin (pelargonidin 3,5-diglucoside), peo = peonin (peonidin 3,5-diglucoside), chr = chrysanthemin (cyanidin 3-glucoside), cy = cyanin (cyanidin 3,5-diglucoside).

‖ Carotenoids.

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EXPERIMENTAL

The petal samples of roses were taken from full-coloured, fresh flowers, supplied by Keisei Rose Institute, Chiba, Japan. Their anthocyanins and flavonols were extracted from the same flowers used for absorption determinations of petals.

Absorption curves of the intact tissues of roses. Spectral absorption was directly measured on the intact petals using a recording spectrophotometer operated as double-beam instrument (Shimadzu Multi-purpose Recording Spectrophotometer, Type MPS-50L).[22-24] The band width (Table 1) was measured as the distance from the wavelength of maximum absorption (λ_{max}) to the long wavelength side of the absorption curve at one-half the maximum A.

Visual and colorimetric evaluations of the flower colour. Code numbers of RHS Colour Chart matched with the flower colour tested were recorded by comparing directly with the flower petals of roses and C.I.E. values[25] were obtained using Colour Measuring and Difference Counting Meter (Colour Studio), Model CS-K 5 (Nihon-Denshoku Co., Tokyo).

Analyses of anthocyanins, flavonols, and carotenoids. After obtaining the absorption spectra of the intact petals and colour data, flower anthocyanins were immediately extracted with cold MeOH (0·1% HCl). The extract was filtered through Celite and concentrated to *ca.* 1/10 volume under reduced pressure at 30–40°. Flavonol glucosides in the petals were extracted with hot 80% EtOH for 30 min and aglycones were hydrolysed with 10% HCl for 2 hr and the extracts were taken into Et_2O. The concentrated extracts were directly spotted on Tôyô-Roshi paper No. 51 or glass plates coated with Avicel microcrystalline cellulose powder and chromatographed with authentic samples using the standard solvents. The relative amounts of anthocyanins were estimated by the visual comparison on chromatograms. The presence of carotenoids was determined by their absorption at 400–500 nm after extraction with 80% MeOH, C_6H_6, or light petrol.

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