STUDIES ON *STREPTOCARPUS*

I. GENETICS AND CHEMISTRY OF FLOWER COLOUR IN THE GARDEN STRAINS

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(With One Text-figure)

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Introduction

The present paper is intended to be the first of a series making a comprehensive survey of the genetics and chemistry of flower colour in the genus *Streptocarpus*, with particular reference to the action of the genes concerned in pigment production. The identity of each species mentioned has been confirmed by Mr B. L. Burtt of the Royal Botanic Gardens, Kew. The garden forms used in the experiments were selected from British nurserymen's stocks. For the chromosome counts, buds were fixed in Carnoy for half a minute followed by La Cour 2 BE, sectioned at 14 μ and stained by the iodine gentian violet method. Root tips were fixed in La Cour 2 BE, sectioned at 6 μ and stained with gentian violet. We are indebted to Mr L. La Cour for the preparation of the cytological material.

Species

The genus *Streptocarpus* was named by Lindley in 1828 and was included by Bentham and Hooker in the division Cyrtandreae of the family Gesneriaceae. The genus comprises some fifty to sixty species which fall naturally into two distinct groups, the caulescent species

found mainly in Central Africa and Madagascar and the acaulescent forms in South Africa. The acaulescent species may be further divided into the monophyllous and polyphyllous sections. The distinction is somewhat arbitrary, since monophyllous species may occasionally produce two or even three leaves.

(*a*) *Chromosome numbers*. The acaulescent and caulescent groups differ not only in their morphology and geographical distribution, but also in chromosome numbers (Table I). The acaulescent species examined have 32 chromosomes; the caulescent 30 chromosomes, somewhat smaller than those in the acaulescent forms. In all species a number of bivalents show secondary pairing. This, together with the high haploid numbers, $n=15$ and 16, suggests that *Streptocarpus* is allopolyploid, probably allotetraploid.

(*b*) *Flower colours*. Flower colour in the species varies considerably in intensity and to a lesser extent in the degree of blueness, but broadly speaking it is always bluish except in the case of *S. lutea* (ivory white) and *S. Dunnii* (brick red). The anthocyanin of the latter is cyanidin 3-pentoseglycoside together with *ca.* 5–10 % of cyanidin 3-bioside.[1] (For formulae see Scott-Moncrieff (1936).) The flower colour is influenced to some extent by the presence of relatively large amounts of an orange-red β-naphthaquinone colouring matter (Price & Robinson, 1938).

The remainder of the species contain anthocyanins derived from delphinidin. In the majority of these species, the anthocyanin is malvidin (3′ : 5′-0-dimethyl delphinidin) 3 : 5-dimonoside, occasionally contaminated with traces of cyanin. In *S. polyanthus* and *S. gracilis* however the delphinidin is incompletely methylated and the anthocyanins are mixtures of malvidin, petunidin and delphinidin 3 : 5-dimonosides.

S. lutea is not now in cultivation, but it is clear from the records that, though recessive for anthocyanin, it is a member of the malvin series.

(*c*) *Species hybrids*. The history of the genus under cultivation is fully recorded in the literature. The dates of introduction of the various species are given in Table I. The first hybrid, *S. bifloro-polyanthus*, was raised in 1859 from *S. Gardeni* (syn. *biflorus*) × *S. polyanthus*. This was followed by *S.* × *Greenii* about 1876, from *S. Saundersii* × *S. Rexii*. Next, about 1883 *S. Rexii* (blue) was crossed with *S. luteus* (ivory white) and blue and white forms were derived from back-crossing the F_1 to *luteus*. These were the only hybrids made within the acaulescent group before 1886, and

[1] The term bioside is used here to indicate a biose residue made up of two hexose units as compared with pentoseglycoside which indicates a biose residue made up of a hexose unit and a pentose unit.

until that time all the streptocarpuses in cultivation had bluish or ivory-white flowers.

In 1886 *S. Dunnii* flowered at the Royal Botanic Gardens, Kew, where William Watson, realizing its value as a potential means of obtaining new colours, used it for pollinating *S. Rexii* and *S. lutea* and obtained progenies

TABLE I

Acaulescent species

First flowered afte introduction	Species	Chromosome no.	Anthocyanin pigment
1826	*Rexii* (Hook.) Lind.	32	Malvidin 3 : 5-dimonoside + trace of cyanidin dimonoside
1855	*polyanthus* Hook.	32	Malvidin 3 : 5-dimonoside containing petunidin and/or delphinidin
1855	*Gardeni* Hook.	32*	Malvidin 3 : 5-dimonoside
1861	*Saundersii* Hook.	—	—
1882	*luteus* N. E. Brown	—	—
1886	*Dunnii* Mast.	32	Cyanidin 3-pentose glycoside + 5-10 % cyan. 3-bioside
1889	*parviflorus* E. Meyer	—	—
1889	*Wendlandii* Sprenger	32	Malvidin 3 : 5-dimonoside
1890	*Woodii* C. B. Clarke (syn. Fannineae)	—	—
1891	*Galpinii* Hook. fil.	32	Malvidin 3 : 5-dimonoside
1900	*Mahoni* Hook. fil.	—	—
1905	*grandis* N. E. Brown	32*	Malvidin 3 : 5-dimonoside
1907	*cyaneus* S. Moore	32*	Malvidin 3 : 5-dimonoside
1915	*denticulatus* Turrill	—	—
1935	*Michelmorei* Burtt	32	Malvidin 3 : 5-dimonoside
1936	*gracilis* Burtt	32	Malvidin 3 : 5-dimonoside containing petunidin and/or delphinidin
1936	*Comptonii* Mansf.	32	—
1936	*Haygarthii* N. E. Brown	32	Malvidin 3 : 5-dimonoside containing some petunidin and/or delphinidin
	Caulescent species		
1884	*Kirkii* Hook. fil.	30	Malvidin 3 : 5-dimonoside containing some petunidin and/or delphinidin
1885	*caulescens* Vatke	30	Malvidin 3 : 5 dimonoside, pure
1906	*Holstii* Engl.	30	Malvidin 3 : 5-dimonoside, pure
1913	*orientalis* Craib	—	—
1932	*saxorum*	30*	Mainly delphinidin 3 : 5-dimonoside, probably containing some petunidin and malvidin

* Root-tip counts.

from both crosses. The *Rexii-Dunnii* hybrids bore "mauve-purple" flowers and were given the name *S.* × *kewensis*. The *lutea-Dunnii* offspring had flowers of a "bright rosy crimson" and were called *S.* × *Watsonii*; they were found to be completely self-sterile.

In 1887, *Rexii*, *lutea*, × *kewensis* and × *Watsonii* were crossed in all possible combinations. The flowers of the resulting seedlings showed con-

siderable variation in colour, and some twenty of the most distinct forms, including × *kewensis* and × *Watsonii*, were passed on to Messrs Veitch, who were largely responsible for the development of the early strain. In later years *Saundersii*, *polyanthus*, *Wendlandii*, *Woodii* and *cyaneus* were crossed into the strain originated by Watson and by further selection the large-flowered garden hybrids as we know them to-day were gradually developed.

GARDEN STRAIN

(*a*) *Flower colours*. The garden hybrids may be divided into seven distinct classes for flower colour—blue, mauve, magenta, rose, pink,

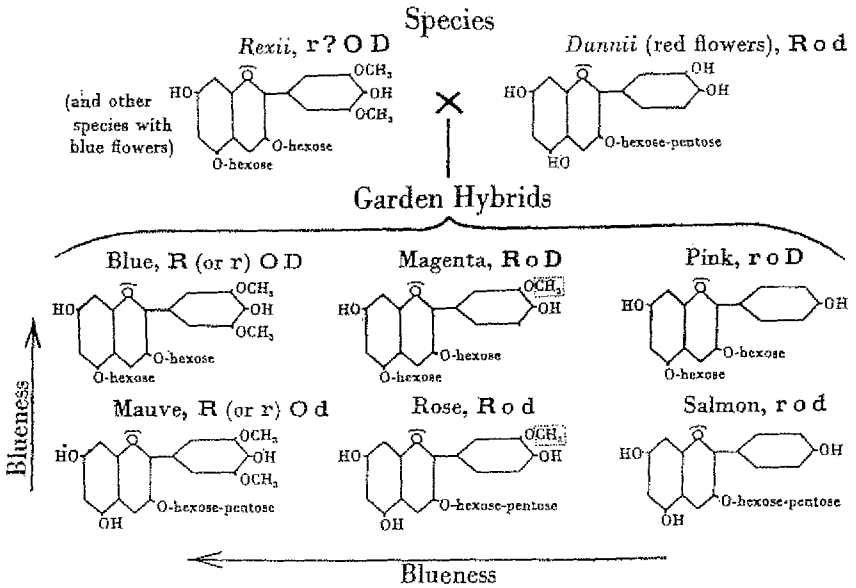

Fig. 1. Chemical and genetical scheme showing the inheritance of anthocyanins in the garden strain of *Streptocarpus*.

salmon and ivory (acyanic) (Table II). Corresponding to the first six classes are six types of anthocyanin comprising derivatives of the three main classes of anthocyanidins, delphinidin, cyanidin and pelargonidin (for formulae, see Beale *et al.* (1939)). Each of these occurs in one or other of two subgroups according to the nature and position of the glycosidal attachments. In one subgroup the anthocyanins are pure 3 : 5-dimonosides; in the other they are mixtures in varying proportions of 3-pentoseglycosides and 3 : 5-dimonosides (Fig. 1). The 3 : 5-dimonosides are a class of glycosides with one hexose molecule substituted at position 3 and another at position 5 on the pigment molecule. The

3-pentoseglycosides have both a hexose and a pentose molecule substituted at position 3, the hydroxyl at 5 being unsubstituted (for nomenclature see Robinson & Robinson, 1932). Thus the blue, magenta and pink varieties contain, respectively, the 3 : 5-dimonosides of each of the three anthocyanidin types, while the mauve, rose and salmon forms contain mixtures of the 3-pentoseglycosides and 3 : 5-dimonosides of each of these types (see Fig. 1).

The anthocyanins in pink and salmon flowers are derived from pelargonidin, and those in the magenta and rose from cyanidin, but in the latter case the anthocyanidin is methylated; that is to say, the hydrogen atom of the 3′ hydroxyl group is replaced by a CH_3 radicle. Actually the methylation is never complete, so that magenta and rose flowers are mixtures of anthocyanins derived from peonidin and cyanidin. Methylation does not occur in the pelargonidin series.

In the delphinidin series (blue and mauve) the anthocyanins are derivatives of malvidin, methylation at both positions 3′ and 5′ being practically complete. The seven colour classes are shown in Table II.

TABLE II

Flower colour	Genotype	Anthocyanins
Ivory	All a forms	Nil
Salmon	**A r o d**	Pelargonidin 3-pentose glycoside + 3 : 5-dimonoside
Pink	**A r o D**	Pelargonidin 3 : 5-dimonoside
Rose	**A R o d**	Peonidin + cyanidin 3-pentoseglycosides + 3 : 5-dimonosides
Magenta	**A R o D**	Peonidin + cyanidin 3 : 5-dimonosides
Mauve	**A r O d** **A R O d**	Malvidin 3-pentoseglycoside + 3 : 5-dimonoside
Blue	**A r O D** **A R O D**	Malvidin 3 : 5-dimonoside

It will be seen from the above that flower colour variation in *Streptocarpus* depends, in the main, upon two kinds of changes in the anthocyanin molecule, (1) change in the state of oxidation of the anthocyanidin, (2) change in the number and position of attachment of sugar residues. These results fall into line with those found in the majority of flowers which have been studied (see Scott-Moncrieff (1939)).

(*b*) *Genetics.* (i) *Breeding Results.* Flower colour in the garden strain is determined mainly by four gene pairs **A**-**a**, **R**-**r**, **O**-**o** and **D**-**d**, the combinations of which give rise to the seven pigment types (Table II). The inheritance of these genes is disomic and independent (Tables III and IV). Deviation and heterogeneity χ^2s (Mather, 1938, §§ 5–8) for back-cross and F_2 generations are shown in Table V. It will be seen that for **R**, **O** and **D** there is good agreement with the expected ratios and the data are homogeneous except in the case of **Oo** back-crossed. This

discrepancy, is due to bad segregation in four families related in pairs. If these four families are omitted then the remaining 20 families show deviation $\chi^2 = 1{\cdot}145$, D.F. = 1, $P = 0{\cdot}3$–$0{\cdot}2$; heterogeneity $\chi^2 = 20{\cdot}197$, D.F. = 19, $P = 0{\cdot}5$–$0{\cdot}3$ and the data are homogeneous. The shortage of dominants (49 **O** : 123o) observed in the four aberrant families could not have been due to faulty scoring and at present remains obscure. The inheritance of **A** is curious in that nine of the sixteen back-cross and forty of the fifty-one F_2 families showed an excess of recessives. Since the observed results often approximated to the 3 : 5 and 9 : 7 ratios expected from complementary genes, crosses were made between twenty-two different **a** plants derived from eleven different families, but coloured forms were never found. Heterogeneity χ^2 shows that the families are very heterogeneous. In the back-cross families χ^2 for one degree of freedom indicates a significant difference between the segregations obtained from **Aa** × **aa** and **aa** × **Aa**. Whether this is generally true for reciprocal crosses of **A** and **a** or whether it results from the particular selection of the plants used as parents, is not yet clear. Although the ratios for **A**–**a** are unsatisfactory, the breeding results are consistent in regard to the expected genotypes.

(ii) *Gene action.* **A** is necessary for anthocyanin production; in its absence the flower colour is ivory white. Its action is quantitative as well as qualitative, the heterozygote producing less anthocyanin than the homozygous dominant. In the absence of **R** and **O** the anthocyanin produced by **A** is derived from pelargonidin. **R** introduces an additional substituent in the anthocyanin molecule at position 3′, giving rise to cyanidin derivatives. **O**, which is epistatic to **R**, introduces two substituents, giving rise to delphinidin derivatives. Wit (1937) in his work on *Callistephus*, claims that the three genes controlling the production of pelargonidin, cyanidin and delphinidin derivatives constitute a multiple allelomorph series. Beale *et al*, 1939, working on *Lathyrus odoratus*, find the state of oxidation is controlled by two pairs of independent genes, S_m and E. As will be seen from Tables III and IV, the genes **R** and **O** in *Streptocarpus* agree with those of *Lathyrus* in their independence.

If the degree of methylation of the anthocyanins in *Streptocarpus* is controlled genetically, then the garden strain must be homozygous for the gene or genes responsible, since **R** produces, not cyanidin, but peonidin-cyanidin mixtures and **O** produces, not delphinidin or petunidin but malvidin.

The gene **D** is completely dominant and gives rise to the 3 : 5-dimonosides found in the blue, magenta and pink classes. The production of

mixtures of 3-pentoseglycosides and 3 : 5-dimonosides in the absence of **D** is determined by at least two other pairs of genes not yet identified.

TABLE III

Genes	F_2 No. of families	XY	Xy	xY	xy	Back-cross No. of families	XY	Xy	xY	xy
O–D	8	233	105	93	39	2	42	60	39	57
R–D	3	53	23	18	6	—	—	—	—	—
A–R	13	394	133	306		1	11	7	32	
A–D	5	131	43	104		—	—	—	—	
A–O	15	479	156	322		—	—	—	—	

TABLE IV

3/33 (deep blue) **AARROODD** × 1/33 (ivory) **aaRROodd**

15/34 (salmon) **AArroodd** × 14¼/34 (blue) **AaRROoDd**

4^{118}/36 (mauve) **AARrOodd** → 66/38	Obs.	Calc.	4^{143}/36 (mauve) **AARrOodd** → 67/38	Obs.	Calc.
Mauve {**AROd**, **ArOd**}	59	65·26	Mauve	63	60·75
Rose **ARod**	21	16·31	Rose	16	15·19
Salmon **Arod**	7	5·44	Salmon	2	5·06

TABLE V

Genes	Back-cross No. of families	$X : x$	χ^2 Deviation from 1 : 1	χ^2 Heterogeneity	Degrees of freedom	P
Aa	16	473 : 594	13·722		1	Very small
				49·282	15	Very small
Rr	3	66 : 51	1·923		1	0·2–0·1
				0·197	2	0·95–0·9
Oo	24	571 : 611	1·354		1	0·3–0·2
				52·704	23	Very small
Dd	16	397 : 445	2·736		1	0·1
				21·703	15	0·2–0·1

Genes	F_2 No. of families	$X : x$	χ^2 Deviation from 3 : 1	χ^2 Heterogeneity	Degrees of freedom	P
Aa	51	2077 : 1187	224·904		1	Very small
				246·021	50	Very small
Rr	28	1012 : 312	1·454		1	0·3–0·2
				23·502	27	0·7–0·5
Oo	27	891 : 332	3·005		1	0·1–0·05
				28·357	26	0·5–0·3
Dd	18	574 : 202	0·435		1	0·7–0·5
				16·847	17	0·5–0·3

It is clear from these and unpublished data that the colours blue, mauve, magenta and rose result essentially from the inheritance of the

genes **O**, **D** from *Rexii* (or other blue-flowered species) and **od** from *Dunnii*. Blue and rose correspond genetically to the parental classes, while the recombination classes **Od** and **oD** are the mauve and magenta types. The origin of the pink and salmon forms cannot be traced with certainty, but it seems that they first appeared in the garden strain about the end of the first decade of this century.

SUMMARY

An account is given of the flower colours, chromosome numbers and flower pigments of cultivated species and hybrids of *Streptocarpus*.

The origin of the garden strain is recounted and the results of breeding experiments within the strain are shown to agree with the known origin of the garden forms.

The action of the flower colour genes is as follows. **A** is necessary for the production of anthocyanin, and alone, produces anthocyanins derived from pelargonidin. **R** in the presence of **A** produces anthocyanins derived from cyanidin. **O**, is epistatic to **R** and in the presence of **A** gives anthocyanins derived from delphinidin. Thus **R** adds one substituent hydroxyl (or methoxyl) group to the anthocyanidin molecule and **O** adds two. **D** produces 3 : 5-dimonoside in place of pentoseglycoside-dimonoside (**d**) mixtures. Seven distinct colour classes result from the combination of these genes. Methylation is complete with the delphinidin derivatives but incomplete with the cyanidin derivatives.

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