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Relationship between species in the genus *Rosa*, section *Pimpinellifoliae*

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The morphology of twelve species of *Rosa* is described and similarities between these species are assessed. Possible origins of the tetraploid species from diploid species are indicated on grounds of comparative morphology.

The wild origins of living and herbarium specimens are given in order to supplement published data on geographical distribution.

Meiosis in pollen mother cells, viability of pollen grains at anthesis and ability to set seed was studied in several F_1 hybrids: no indication of complete or even partial sterility was found. Reproductive isolation is therefore unlikely to be maintained by reduced fertility of interspecific hybrids.

Three species are reduced to synonymy with three other species, being retained as subspecific taxa. Two species are transferred from section *Pimpinellifoliae* to section *Cinnamomeae*.

KEY WORDS:—*Rosa*—phenetic classification—cladistic classification—hybrids.

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INTRODUCTION

The genus *Rosa* includes over 100 species, distributed throughout the temperate and subtropical regions of the Northern Hemisphere. Their nomenclature is extremely confused (Rowley, 1959), but according to the popularly accepted classification of Rehder (1940), the genus has 4 subgenera: 3 are monotypic and the fourth, *Rosa*, includes all the remaining species grouped

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into 10 sections. The genus exhibits a typical polyploid series with a basic chromosome number of 7. Euploids range from $2n = 2x = 14$, to $2n = 8x = 56$ (Darlington & Wylie, 1955) and aneuploids are rare (Rowley, 1960a).

The present investigation was initiated in order to obtain data on the comparative morphology and geographical distribution of species in the section *Pimpinellifoliae* of the subgenus *Rosa*, and to identify pre- or post-zygotic sterility barriers which might reproductively isolate the species from one another. It was hoped that the information obtained would provide a fresh perspective for taxonomic revision of this group. The investigation was based on fresh material at the University of Reading, and herbarium specimens which were examined at the Royal Botanic Gardens, Kew, and the British Museum (Natural History).

The section *Pimpinellifoliae* includes nine diploid species (*R. omeiensis* Rolfe, *R. sericea* Lindl., *R. hugonis* Hemsl., *R. xanthina* Lindl., *R. graciliflora* Rehd. & Wils., *R. primula* Boulenger, *R. ecae* Aitch., *R. koreana* Kom., and *R. farreri* Stapf.) and three tetraploid species (*R. spinosissima* L., *R. hemisphaerica* Herrm., and *R. foetida* Herrm.). Boulenger (1934) considered the tetraploid species *R. nanothamnus* Boulenger, to have affiliations with the *Pimpinellifoliae*, but Roberts (1975) showed that this species should be placed in the section *Caninae*.

No living accession of *R. graciliflora* was obtainable, and since reference to living material was considered essential it was decided not to consider this species further.

R. hemisphaerica is merely a double-flowered cultivar of *R. rapinii* Boiss., and no other character has been described by which they can be distinguished (Rowley, 1959). Its flowers have no functional anthers or carpels as these parts have been replaced by petals and populations of this mutant could not perpetuate themselves by seed in the wild. For this reason these two taxa have here been treated as one species, under the earlier name of *R. hemisphaerica*.

Rehder (1940) provides the following criteria to distinguish between *R. hugonis* and *R. xanthina*:

Character	<i>R. hugonis</i>	<i>R. xanthina</i>
(a) Bristles on main shoot	present	absent
(b) No. of leaflets per leaf	5 to 13	7 to 13
(c) Shape of leaflets	oval, obovate, or elliptical	oval, or suborbicular, rarely elliptical
(d) Diameter of flowers	5 cm	4 cm

Herbarium specimens of the two species were virtually indistinguishable: they were usually represented by lateral branches so the character of 'main shoots with or without bristles' could not be used. Most flowers were imperfectly preserved, and it was not always possible to distinguish them on the basis of their diameter. The two species could not be distinguished on number and shape of leaflets alone, as they overlap considerably in both characters. Although living specimens can be distinguished, only one plant of each species was available in the collection at Reading. It was therefore decided, for the

purposes of morphological comparison, to treat the two species together, designating them as *R. hugonis*/*R. xanthina*.

A diploid species which Rehder placed in the *Cinnamomeae*, *R. forrestiana* Boulenger, is also included in this study as it has much in common with some of the *Pimpinellifoliae*.

PLANT MATERIAL USED, GEOGRAPHICAL DISTRIBUTIONS AND CYTOLOGICAL TECHNIQUE

Living material in the Rose Collection of the Plant Science Laboratories, University of Reading was used.

Four accessions are known to be of wild origin, but the others were received from Botanical Gardens or private collections, and may be one or more generations removed from the wild. In all cases the specimens match the published descriptions and herbarium specimens.

Hybrids raised by G. D. Rowley, who kindly allowed them to be used in this investigation were: *R. primula* ♀ × *R. hugonis* ♂ (13.57.1) *R. primula* ♀ × *R. ecae* ♂ (40.57.A. and 40.57.B.) *R. sericea* ♀ × *R. primula* ♂ (7.56) *R. sericea* ♀ × *R. xanthina* ♂ (13.59.) *R. xanthina* ♀ × *R. sericea* ♂ (18.57.E.) *R. hugonis* ♀ × *R. xanthina* ♂ (8B.56.) *R. spinosissima* ♀ × *R. foetida* ♂ (25.59.)

Although it is well established that the *Pimpinellifoliae* occur in Europe and Asia, published statements (Rehder, 1940; Rehder & Wilson, 1915) on their distributions have always appeared in terms of broad geographical areas, with no indication as to the number of records on which they were based. The origins and accession numbers of living plants which were examined in the course of this investigation and a summary of areas in which specimens have been collected is listed below for each species.

R. omeiensis (accession number: AB978 and AA73)

Bhutan, China (Hupeh, Kansu, Shansi, Shensi, Szechwan, East and South East Tibet, and Yunnan), India (Assam), and Sikkim.

R. sericea (accession numbers: V192E (raised from seed from Nepal), K904, OMP and G938)

Burma, China (South, South-East and South-West Tibet, and Yunnan), India (Assam, Punjab and Uttar Pradesh), and Nepal

R. ecae (accession number: SN)

Afghanistan, China (Shansi), Pakistan (Chitral), and U.S.S.R. (Turkestan).

R. primula (accession numbers: G179 (cuttings from Turkestan) and M3728) U.S.S.R. (Turkestan).

R. hugonis/*R. xanthina* (accession numbers respectively A207 and V551)

China (Hopch, Kansu, Shansi, Shantung, Southern Sinkiang, Szechwan, and South Manchuria), Korea, and Mongolia.

R. koreana (accession number: M87)

Korea.

R. forrestiana (accession number W693)

China (Yunnan), and 'Tibet-China border area'.

R. foetida (accession numbers: SG and M481 (*R. foetida* var. *bicolor*))

Afghanistan, Afghanistan/Pakistan (Kunar Valley) Pakistan (Buluchistan and

Chitral), India (Punjab), Iran, Iraq, Jammu and Kashmir, Syria, Turkey, and U.S.S.R. (Armenia, Turmenistan and Turkestan).

R. hemisphaerica:

India (West Himalaya) and Turkey.

R. farreri (accession number: W479)

N.W. China (Rehder, 1940).

R. spinosissima (accession number: S403 (seed from the Crimean Peninsula) and K1451 (cuttings from Scotland))

China (Szechwan and Manchuria), Europe (Austria, Belgium, Bulgaria, Czechoslovakia, France, Greece, Holland, Scandanavia, Switzerland, and the United Kingdom), Korea, Mongolia (Altai Mountains), Turkey, and U.S.S.R. (Caucasus Mountains, Krymskiy Poluostrov, and East Turkestan).

Distribution maps were compiled on the basis of the above records. The distributions of *R. omeiensis*, *R. sericea*, *R. ecae* and *R. primula* are shown in Fig. 1, those of *R. hugonis/R. xanthina*, *R. koreana* and *R. forrestiana* are shown in Fig. 2, and those of *R. foetida*, *R. hemisphaerica* and *R. spinosissima* are shown in Fig. 3.

The distributions of *R. sericea*, *R. omeiensis*, and *R. spinosissima* are each based on over 100 records, and those of both *R. foetida* and *R. hugonis/R. xanthina* are based on over 50 records. As there were only nine records of *R. ecae*, two each of *R. hemisphaerica*, *R. koreana* and *R. forrestiana*, and one of *R. primula*, they may well be more widely distributed than the above statements imply.

All the specimens which were examined of *R. farreri*, had been obtained from plants in cultivation whose wild origins were unknown.

Tackholm (1922) and Hurst (1925) drew attention to a pattern of distribution of species of *Rosa* which is related to levels of polypoidy. They showed that in both the new world and the old world, diploid species are nearest to the equator, tetraploid species generally have a more northerly distribution, while hexaploids and octoploids are nearest to the North Pole. The ranges of each of the three tetraploid species which fall into the *Pimpinellifoliae* extend further West than the diploid species, but only that of *R. spinosissima* extends significantly further North.

In order to study meiosis in pollen mother cells, young flower buds were first fixed in 3 : 1, ethanol : acetic acid, hydrolysed in N HCl for 10 minutes at 60°C , and then stained in Feulgen Reagent. Acetocarmine squashes of anthers were then viewed by phase contrast microscopy.

TAXONOMIC DISCRIMINANTS AND THE ASSESSMENT OF SIMILARITY

The characters which are discussed are those which provide effective discriminants between the species under consideration. Fortuitously all the character states were amenable to qualitative definition and this enabled an assessment of similarity between the species to be based on Coefficients of Association.

Taxonomic discriminants are listed in Table 1, and the character states assumed by each species are presented in Table 2 for purposes of comparison. In Table 1, characters are identified by numbers, and the various states of each

Figure 1. Distribution map of the diploid species: *R. omeiensis*, *R. sericea*, *R. ecae* and *R. primula*.

Figure 2. Distribution map of the diploid species: *R. hugonis/R. xanthia*, *R. koreana* and *R. forrestiana*.

Figure 3. Distribution map of the tetraploid species: *R. foetida*, *R. hemisphaerica* and *R. spinosissima*.

Table 1. List of taxonomic discriminants

Number assigned to character	o	Character state	+	x
	HABIT			
1	markedly stoloniferous	not markedly stoloniferous		
2	branching sarmentose	branching not sarmentose		
	STEMS			
3*	reddish	brownish		
	LATERAL SHOOTS			
4	phyllotaxy distichous	phyllotaxy polystichous (usually pentastichous)		
	ARMATURE			
5	dense	sparse		
6	heteracanthous	homoacanthous		
7	prickles hooked	prickles straight		
8	some alate prickles	no alate prickles		
9	prickles laterally compressed	prickles terete		
	STIPULES			
10	margins sinuous and tightly inrolled	margins entire and not inrolled		margins sinuous and loosely inrolled
	LEAFLETS			
11†	number per leaf—distribution 1	number per leaf—distribution 2		number per leaf—distribution 3
12	serrate at apex only	serrate all round		
13	serration double	serration single		
14	serration obtuse	serration acute		
15	glaucous above	not glaucous above		
16	villous below	pubescent below		
17	densely glandular below	glands on main nerves only		
	INFLORESCENCE			
18	flowers in corymbs	flowers solitary		
19	bracts absent	bracts present		
20	pedicels thick and fleshy in fruit	pedicels thin in fruit		
21	pedicels eglandular	pedicels glandular		
	CALYX			
22	sepals not leafy	sepals leafy		
23	sepals spinose	sepals not spinose		
24	sepals strongly reflexed	sepals hardly reflexed		
	CALYX AND COROLLA			
25	tetramerous	pentamerous		
	COROLLA			
26	petals yellow	petals red		petals white
27	cup shaped	saucer shaped		
28	petals broad based, apex curled	petals narrow based, apex not curled		
	DISC			
29	broad	narrow		
	ANDROECIUM			
30	stamens fused to disc	stamens quite free below		
31‡	stamens not inflexing	stamens inflexing		
	GYNOECIUM			
32**	styles short, stigmas bunched into a tight cap	styles long, stigmas not bunched		
	AROMA			
33	not foetid	foetid		

Number assigned to character	Character state		
	o	+	x
	HIPS		
34**	globose	ovoid	
35	mahogany in colour	red	black
	CHROMOSOME NUMBER		
36	28	14	

* The red colour of the main stems and branches of *R. primula* and *R. ecae* can be distinguished, even on herbarium sheets, from the dull brown colour in the other species investigated. Newly formed branches of these other species are sometimes red in early spring, but darken after 2 or 3 weeks.

† The number of leaflets per leaf was counted on each of ten leaves, which were arbitrarily chosen on each herbarium specimen. The highest and lowest figure for each herbarium specimen was then recorded. In 4 species and the *R. hugonis/R. xanthina* assemblage, the number of specimens examined was large enough for meaningful histograms to be drawn (Fig. 4) showing numbers of specimens in each class of leaflet number. Modal classes for highest numbers of leaflets were the same in *R. sericea*, *R. spinosissima*, and *R. hugonis/R. xanthina*, as also were the modal classes for the lowest numbers. In *R. omeiensis*, the divergence between the modal classes of highest and lowest numbers of leaflets is greater than in any of the other species. This might be a consequence of the generally higher values exhibited allowing more flexibility in numbers of leaflets. The arbitrary designations of distributions 1, 2 and 3 are used in Table 1 to distinguish the three types of distribution which were found.

‡ Only one living plant of *R. forrestiana* was available for examination. Its flowers had broad discs, to which the filaments were partly fused, and its stamens were consequently incapable of inflexion. As this character could only be satisfactorily studied on living material and in the absence of further information, an entry of 'no comparison' was made against this species in Table 2.

** Oval hips and short styles were found together in the same species, so usually were globose hips and long styles. Style length was recorded shortly after the flowers opened, while the receptacle was at an early stage of development, and the hip was not recognisably globose or oval. Therefore the 'length of style' is not geometrically dependent on hip shape, i.e. they are not 'necessary correlates' in the sense of Sokal & Sneath (1963). They were therefore recorded as separate characters.

character are fully defined and entered in a column which is headed by symbols (o, + or x) to facilitate the construction of Table 2.

Coefficients of Association were calculated between all pairs of species, and are shown in Table 3, where they are expressed as percentages, to the nearest whole number. Each coefficient is the ratio of the number of character states two species have in common to the total number of characters on which they were compared, and is therefore an estimate of the degree of similarity between the species. For a full discussion and presentation of the different possible coefficients, see Sokal & Sneath (1963: 128 et seq.).

Where the symbol 'nc' (no comparison) was entered in Table 1 against a particular species, the denominator (total number of characters compared) was correspondingly reduced when coefficients of association were calculated. When two or three states of the same character were found in a single species, coefficients of association were calculated separately for minimum and maximum similarity and the mean value was taken.

When two species showed the same states of a particular character on which they both varied, they were said not to differ, and to have maximum similarity on that character.

The similarity between species and groups of species is expressed in the form of a dendrogram of affinities in Fig. 5. In constructing this dendrogram, the

Table 2. Character states found in the species. (See Table 1 for the character states referred to by the symbols ○, + and ×. The symbol nc (no comparison) is entered where information was missing

Character numbers																																			
1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25	26	27	28	29	30	31	32	33	34	35	36
<i>R. omeiensis</i>	+	+	+	+	+	+	+	○	○	○	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
<i>R. sericea</i>	+	+	+	○	○	+	○	○	○	○	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
<i>R. hugonis/R. xanthina</i>	+	+	+	+	+	+	+	○	○	○	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
<i>R. eciae</i>	+	+	○	+	+	+	+	○	○	nc	+	○	○	○	○	○	○	○	○	○	○	○	○	○	○	○	○	○	○	○	○	○	○	○	+
<i>R. primula</i>	+	+	○	+	+	+	+	○	○	nc	+	○	○	○	○	○	○	○	○	○	○	○	○	○	○	○	○	○	○	○	○	○	○	○	+
<i>R. koreana</i>	+	+	+	○	+	+	+	+	+	nc	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
<i>R. forrestiana</i>	+	+	+	+	+	+	+	+	+	nc	+	+	+	+	+	+	○	+	+	+	+	+	+	+	+	+	+	+	+	+	nc	+	+	+	+
<i>R. farreri</i>	+	+	+	+	+	+	+	+	+	nc	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
<i>R. spinosissima</i>	○	+	+	+	○	+	+	+	○	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	○	○
<i>R. foetida</i>	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
<i>R. hemisphaerica</i>	+	+	○	+	+	+	○	+	+	nc	+	+	+	○	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	nc	+	○

Figure 4. Highest and lowest numbers of leaflets per leaf in samples of ten leaves from each herbarium specimen.

species with the greatest similarity were the first to be united. The group or groups so formed were linked progressively with other species or groups of species with which they showed the greatest similarity.

The similarity between a species and a group is given as its average similarity with each member of the group. The similarity between two groups was calculated as the average similarity of each member of one group with each number of the other group. This method is described and discussed by Sokal & Sneath (1963: 180 et seq.), where it is called the 'unweighted pair group

Table 3. Matrix of coefficients of association. (Figures expressed in percentages to the nearest whole number)

<i>R. omeiensis</i>	
<i>R. sericea</i>	92
<i>R. hugonis/R. xanthina</i>	79 82
<i>R. ecae</i>	67 67 83
<i>R. primula</i>	70 70 86 97
<i>R. koreana</i>	53 53 69 54 57
<i>R. forrestiana</i>	49 49 65 47 50 85
<i>R. farreri</i>	54 54 70 53 56 90 87
<i>R. spinosissima</i>	65 68 81 69 71 69 56 64
<i>R. foetida</i>	53 53 68 64 67 60 62 61 63
<i>R. hemisphaerica</i>	60 60 79 62 65 65 64 69 68 79

R. omeiensis
R. sericea
R. hugonis/R. xanthina
R. ecae
R. primula
R. koreana
R. forrestiana
R. farreri
R. spinosissima
R. foetida
R. hemisphaerica

Table 4. Viability of pollen at anthesis and number of seeds per hip in species and hybrid accessions

Species or hybrid accession	% viable pollen at anthesis*	No. seed per hip**
DIPLOID SPECIES		
<i>R. omeiensis</i> AB978	81.8	7.8
<i>R. sericea</i> G938	95.7	10.4
<i>R. hugonis</i> A207	62.2	7.9
<i>R. xanthina</i> V551	81.1	10.6
<i>R. koreana</i> M87	94.5	0.0
<i>R. forrestiana</i> W693	78.0	7.8
<i>R. farreri</i> W479	97.4	6.3
DIPLOID HYBRIDS		
<i>R. sericea</i> × <i>R. primula</i> 7.56.	89.7	8.3
<i>R. sericea</i> × <i>R. xanthina</i> 13.59.	22.2	—***
<i>R. xanthina</i> × <i>R. sericea</i> 18.57.E.	94.4	5.4
<i>R. primula</i> × <i>R. ecae</i> 40.57.A.	95.2	4.7
<i>R. primula</i> × <i>R. ecae</i> 40.57.B.	48.6	6.9
<i>R. primula</i> × <i>R. hugonis</i> 13.57.1.	85.4	1.3
<i>R. hugonis</i> × <i>R. xanthina</i> 8B.56.	89.3	7.4
TETRAPLOID SPECIES		
<i>R. spinosissima</i> S403	50.6	13.9
<i>R. spinosissima</i> K1451	84.5	10.7
<i>R. foetida</i> SG	27.5	0.0
TETRAPLOID HYBRID		
<i>R. spinosissima</i> × <i>R. foetida</i> 25.59.	72.0	3.3

* each value based on a sample of 900 or more pollen grains.

** each value based on seed from a sample of 12 or more hips.

*** no mature hips on the plant.

viability are interpreted with caution and with due regard to this known level of variability.

The average number of seeds which set per hip on open-pollinated plants was assessed on samples of not less than 12 hips (Table 4). There are many environmental factors which might affect the production of seeds and this value cannot be regarded as a sensitive index of a plant's fertility. However, an observation that an F_1 hybrid never, or only rarely produced seed might be taken as evidence that it is incapable of completing its cycle of sexual reproduction. Such failures might be due to irregularities in megasporogenesis, which might be strongly suspected if irregularities had been detected in microsporogenesis. Alternatively, it might be due either to the inability of stylar tissue to support normal growth of a pollen tube, or to an inability to support normal development of a zygote.

Diploid plants

As a result of terminalization, the frequency of chiasmata at metaphase I was lower than at diakinesis in each of the plants which was studied at both stages.

In all species and hybrids, most cells at diakinesis and at metaphase I of meiosis had seven bivalent associations. Univalent chromosomes and/or multivalent associations were found in some species and hybrids, but in relatively low frequencies. As the frequency of univalent chromosomes in the F_1 hybrids was no higher than in species, univalents provide no clear evidence of limited homology between parental genomes; it is more probable that these univalents resulted from random failure in the formation of chiasmata leading to a complete lapse of association between paired chromosomes at diplotene.

Multivalent associations were seen in five of the diploid species and all seven of the diploid hybrids. These took the form of trivalent or quadrivalent associations indicating heterozygosity for at least one translocation, and hexavalent associations indicating heterozygosity for at least two translocations. In only two plants, *R. sericea* and *R. omeiensis*, were no multivalents recorded. Studies in the genus *Oenothera*, reviewed by Cleland (1962), have shown that chromosomes in multivalent associations may be so oriented at metaphase I of meiosis, as to ensure an ordered segregation at anaphase I, leading to the formation of balanced gametes. However, such a mechanism demands a regularity in the formation of multivalents and an alternate configuration of chromosomes at metaphase I, neither of which were evident in any of the plants discussed here. Thus it might be anticipated that in each of the plants with multivalent associations a proportion of segregants would be unbalanced and that this would result in the production of unbalanced gametes. However, this expectation was not borne out by studies of pollen and seed, as several plants in which multivalents were found compare favourably with *R. sericea* and *R. omeiensis* with respect to viability of pollen and/or numbers of seed per hip (Table 4). Furthermore, as multivalents were found in five out of the seven diploid species examined in this investigation and as Erlanson (1933) has reported multivalents in plants of wild origin of *R. blanda*, *R. woodsii* and *R. pisocarpa* it seems that natural selection does not rigorously eliminate translocation mutants in natural populations of roses. The only immediate conclusion that can be drawn from them is entirely negative: the

appearance of multivalent associations in the hybrids constitutes neither proof that the genomes of the parent species have diverged significantly during evolution, nor evidence that gene flow between the parent species would be restricted.

In those plants representing the seven diploid species, the highest percentage of viable pollen was recorded in *R. farreri* (97.4%) and the lowest was recorded in *R. hugonis* (62.2%). In the hybrids *R. sericea* × *R. primula* 7.56., *R. xanthina* × *R. sericea* 18.57.E., *R. primula* × *R. ecae* 40.57.A., *R. primula* × *R. hugonis* 13.57.1., and *R. hugonis* × *R. xanthina* 8B.56., the percentage of viable pollen fell within this range and several seeds per hip were set. However, in the hybrid *R. primula* × *R. ecae* 40.57.B. there was only 48.6% viable pollen and in the hybrid *R. sericea* × *R. xanthina* 13.59., there was only 22.2% viable pollen. In the case of *R. primula* × *R. ecae* 40.57.B., there were, on average, 6.9 seeds per hip and there is thus no question of there being a complete barrier to gene flow between the parental species through F_2 sterility. Its sib. *R. primula* × *R. ecae* 40.57.A., was apparently highly fertile, having 95.2% viable pollen and on average 4.7 seeds per hip, so there is no unequivocal evidence that hybrids between *R. primula* and *R. ecae* suffer even reduced fertility.

The hybrid *R. sericea* ♀ × *R. xanthina* ♂ 13.59., had fewer viable pollen grains (22.2%) than any of the other species or hybrids and no seed developed to maturity during the three year period of the investigation. In the absence of further information it might have been concluded that this was indicative of a sterility barrier between *R. sericea* and *R. xanthina*. However, the reciprocal hybrid *R. xanthina* ♀ × *R. sericea* ♂ 18.57.E., had a high frequency of viable grains (94.4%) and, on average, there were 5.4 seeds per hip. The reason for the sterility of *R. sericea* ♀ × *R. xanthina* ♂ 13.59., is obscure. The frequency of chiasmata at metaphase I was higher than in the other plants examined but otherwise meiosis was unexceptional. The plant seemed healthy and there was no sign of any disease which might have reduced its vigour. Possibly the maternal cytoplasm of *R. sericea* differed in some important respect from that of *R. xanthina*. In order to assess whether or not sterility of F_1 plants might, in some degree, reproductively isolate the parental species, it would be necessary to assess the fertility of a wider selection of F_1 hybrids between them.

Apart from the hybrid *R. sericea* × *R. xanthina* 13.59., only one diploid plant, *R. koreana* M87, failed to produce any seed. This accession is known to be of wild origin, meiosis in pollen mother cells was in no way exceptional and a high proportion (94.5%) of pollen grains were viable at anthesis. It therefore seems that some environmental factor, possibly climatic, is responsible for its failure to set seed. Nevertheless this observation serves to show that caution is needed in assessing the viability of a plant outside its natural environment.

Tetraploid plants

Univalent chromosomes and multivalent associations were found in both accessions of *R. spinosissima*, in *R. foetida* and in the hybrid *R. spinosissima* × *R. foetida*. Whereas multivalent associations in a diploid plant is indicative of heterozygosity for a translocation, in a tetraploid plant it might be due either to heterozygosity for a translocation or to segmental allopolyploidy. In either case, the formation of multivalents might be expected to lead to a loss

of fertility. However, as in the diploid species and hybrids which were studied, there was no clear evidence that appearance of univalents or multivalents was necessarily associated with loss of fertility, because in clone K1451 of *R. spinosissima*, 84.5% of pollen was viable at anthesis and there were, on average, 10.7 seeds per hip. Also, although only 50.6% of pollen was viable in clone S403 of *R. spinosissima* a high average of 13.9 seeds per hip was recorded.

The fertility of clone SG of *R. foetida* was strikingly low. Only 27.5% of pollen was viable at anthesis and no seeds were found in the hips. This species is grown in many gardens in Britain, where it is well known for its low fertility. No successful attempt to use it in crosses as an ovular parent has been recorded in the literature. However, as a pollen parent it is not totally barren; it has been used in breeding several garden roses (Harms, 1956) and it was the pollen parent of the hybrid *R. spinosissima* $\times$ *R. foetida* 25.59. In this hybrid, the viability of pollen grains at anthesis was high (72%) and there were, on average 3.3 seeds per hip. This hybrid is clearly more fertile than *R. foetida* and these observations underline the strangely low level of fertility in that species.

DISCUSSION AND CONCLUSIONS

The dendrogram of affinities (Fig. 5), shows a primary dichotomy between one group formed by *R. forrestiana*, *R. farreri* and *R. koreana* and a second group formed by all the other species. It can also be seen that the three species of the first group are closely similar to one another (united at the 86% level of similarity). The evidence that *R. farreri* and *R. koreana* are phenetically more closely related to *R. forrestiana* than to the other *Pimpinellifoliae* is therefore strong. *Rosa forrestiana*, *R. farreri* and *R. koreana*, each of which is diploid, share 7 character states (represented in Table 2 by the symbol + for characters 9, 10, 19, 22, 26, 32 and 34) which are not found in any of the other diploid species studied here. The simplest interpretation of this phenomenon is that they form a monophyletic group which diverged from the other diploid species before or when differences in these morphological characters arose (Table 6).

As *R. farreri* and *R. koreana* are both phenetically and cladistically closely related to *R. forrestiana*, and since they appear to have no obvious affinity with any of the remaining 11 sections of the genus, it is proposed that they should be placed with *R. forrestiana* in the section *Cinnamomeae*.

Each of the tetraploid species combine character states which are otherwise polarized in one or other of the two diploid groups which were presumed, above, to be monophyletic, and may thus have arisen as amphidiploid hybrids between them. This theory could be tested by attempting to resynthesize the tetraploid species experimentally through hybridization and doubling the number of chromosomes.

Chromosome pairing in *R. spinosissima* was quite regular, which suggests that either the genomes of the putative parental groups were sufficiently distinct for pairing to occur preferentially between genomes of common origin, or that after the initial tetraploid was formed, genomic or genotypic changes took place which stabilized meiosis, ensuring regular formation of bivalents. A study of meiosis in an F_1 hybrid between the presumed monophyletic groups of diploid species would have helped to clarify this issue and it is unfortunate that such a hybrid was not available in this investigation.

Table 5. Analysis of chromosome associations at diakinesis and metaphase I of meiosis in species and hybrids

	Species or hybrid	Diakinesis						Metaphase I								
		average no. chromosome associations of given class per cell						average no. chromosome associations of given class per cell								
		Cells examined	Univalents	Bivalents	Trivalents	Quadrivalents	Hexavalents	Frequency of chiasmata per cell	Cell examined	Univalents	Bivalents	Trivalents	Quadrivalents	Hexavalents	Frequency of chiasmata per cell	
Diploid species	<i>R. omeiensis</i> AB978	32	—	7.00	—	—	—	9.84	25	—	7.00	—	—	—	—	7.80
	<i>R. sericea</i> G938	29	0.28	6.18	—	0.34	—	8.17	54	0.26	6.75	—	0.06	—	—	7.39
	<i>R. hugonis</i> A207	50	0.18	6.14	0.02	0.34	0.02	8.62	50	0.24	6.22	0.04	0.30	—	—	7.48
	<i>R. xanthina</i> V551	60	—	7.00	—	—	—	8.72	60	—	7.00	—	—	—	—	7.18
	<i>R. koreana</i> M87	50	0.20	6.90	—	—	—	8.86	50	0.16	6.88	—	0.02	—	—	7.42
	<i>R. forrestiana</i> W693	37	0.37	6.44	0.05	0.14	—	8.03	36	0.08	6.80	0.03	0.07	—	—	7.67
	<i>R. farreri</i> W479								25	0.08	6.72	—	0.12	—	—	7.24
Diploid hybrids	<i>R. sericea</i> x <i>R. primula</i> 7.56.	50	0.08	6.96	—	—	—	9.58	29	0.20	6.90	—	—	—	—	7.28
	<i>R. sericea</i> x <i>R. xanthina</i> 13.59	81	—	6.92	—	0.04	—	9.26	99	0.12	6.86	—	0.04	—	—	8.30
	<i>R. xanthina</i> x <i>R. sericea</i> 18.57.E.	60	0.07	6.92	—	0.02	—	8.18	15	—	7.00	—	—	—	—	7.53
	<i>R. primula</i> x <i>R. ecae</i> 40.57.A.	50	0.04	6.80	—	0.06	0.02	8.86	50	0.02	6.87	—	0.06	—	—	7.62
	<i>R. primula</i> x <i>R. ecae</i> 40.57.B.	20	0.10	6.95	—	—	—	9.45	50	0.04	6.98	—	—	—	—	7.46
	<i>R. primula</i> x <i>R. hugonis</i> 13.57.1.	87	0.05	6.44	—	0.27	—	10.24	20	0.30	6.65	—	0.1	—	—	7.20
	<i>R. hugonis</i> x <i>R. xanthina</i> 8B.56.	55	—	6.86	—	0.07	—	8.95								
Tetraploid species	<i>R. spinosissima</i> S403	50	0.22	13.64	0.02	0.10	—	19.82	50	0.76	13.62	—	—	—	—	17.44
	<i>R. spinosissima</i> K1451	25	0.32	13.10	—	0.36	—	15.72	25	0.40	13.48	—	0.16	—	—	14.40
	<i>R. foetida</i> SG	25	0.48	10.20	0.16	1.64	—	19.76	39	0.56	13.23	—	0.23	—	—	14.51
Tetraploid hybrid	<i>R. spinosissima</i> x <i>R. foetida</i> 25.59.	4	—	14.00	—	—	—	16.00	19	1.26	13.00	0.11	0.10	—	—	14.11

Table 6. The two main groups of diploid species and the character states which distinguish them

	Group 1	Group 2
Species	<i>R. forrestiana</i> <i>R. farreri</i> <i>R. koreana</i>	<i>R. omeiensis</i> <i>R. sericea</i> <i>R. hugonis/R. xanthina</i> <i>R. ecae</i> <i>R. primula</i>
Character states	Prickles terete Margins of stipules entire and not tightly inrolled Bracts present Sepals leafy Petals red Styles long, stigmas not bunched Hips ovoid	Prickles laterally compressed Margin of stipules sinuous and tightly inrolled Bracts absent Sepals not leafy Petals yellow or white Styles short, stigmas bunched into a tight cap Hips globose

The irregularity of meiosis and consequent sterility of *R. foetida* indicate that neither of the alternative assumptions made for the evolution of *R. spinosissima* applies in this species. In view of its sterility it is pertinent to record comments written on herbarium sheets by collectors of specimens kept at the herbarium of the Royal Botanic Gardens, Kew. Two collectors in Iraq stated, respectively, that this species was grown as a hedge plant and that petals were used in making jam. A collector in Turkey commented on its popularity in gardens and as a hedge plant and another stated that he had not seen it growing wild and doubted if it did. These observations indicate that *R. foetida* is a popular domestic plant and may be either rare or non-existent in the wild condition. It therefore seems possible that its propagation is principally clonal and that there is not a high selective premium on fertility.

R. spinosissima extends further to the north and west than the diploid species, although it partially overlaps the ranges of *R. hugonis/R. xanthina* and *R. koreana*. If, as Vavilov (1951) has postulated, the centre of evolution of roses is in S.E. Asia, where most of the diploid species of the genus are concentrated, *R. spinosissima* may have exploited the more remote and possibly less hospitable territories of Mongolia and Europe and thereby avoided competition with the longer established diploid species. The two other tetraploid species, *R. foetida* and *R. hemisphaerica*, have a more westerly distribution than the diploid species and the same principle may be invoked to explain this.

Although no hybrids between diploid and tetraploid species were studied, it is likely that they would be largely sterile, but it would be wrong to assume that they would necessarily be totally sterile. Wulff (1954) and Rowley (1960b) have reported that although triploid roses are usually relatively infertile, they occasionally give rise to fertile diploid or tetraploid offspring. Work on other genera has indicated the possibility of introgression between diploids and tetraploids through partially fertile triploids. For example, Jones

& Borrill (1962) have shown that triploid hybrids of *Dactylis* bore some seed when they were allowed to outcross to diploids and tetraploids.

Rowley (1959) united *R. sericea* and *R. omeiensis* under the earlier name; *R. sericea*. These species were also found to be closely similar in this investigation (92%), but the similarity between *R. primula* and *R. ecae* is even greater (97%) and *R. hugonis* is so closely similar to *R. xanthina* that it is virtually impossible to distinguish herbarium specimens of the two species. As *R. sericea* and *R. omeiensis* have been united, it would be consistent to unite *R. primula* with *R. ecae* and *R. hugonis* with *R. xanthina*. No evidence of sterility barriers between *R. primula* and *R. ecae* emerged from studies on the two F_1 hybrids *R. primula* $\times$ *R. ecae* 40.57.A. and 40.57.B., or between *R. hugonis* and *R. xanthina* from studies of the F_1 hybrid *R. hugonis* $\times$ *R. xanthina* 8B.56., so no *a priori* argument can be advanced against uniting the species of either pair.

Although the evidence presented accords with Rowley's decision to unite *R. omeiensis* with *R. sericea*, it is now proposed that the two taxa be given rank as subspecies since their distributions represent major geographical groupings, even though they overlap in part, viz.

Rosa sericea Lindley subsp. *sericea*.

R. sericea Lindley in *Rosarum Monographia*, 105 (1820).

R. sericea subsp. *omeiensis* (Rolfe) Roberts comb. nov.

R. omeiensis Rolfe in *Curtis's Botanical Magazine*, 138: t. 8471 (1912).

It is proposed that *R. primula* and *R. ecae* should likewise be united and that the two subordinated taxa should also rank as subspecies for similar reasons, viz.

Rosa ecae Aitch. subsp. *ecae*

R. ecae Aitch. in *Journal of the Linnean Society (Botany)*, 18: 54 (1881).

Rosa ecae subsp. *primula* (Boul.) Roberts comb. nov.

R. primula Boul. in *Bulletin du Jardin botanique de l'Etat Bruxelles*, 14: 121 (1936).

Further, *R. hugonis* and *R. xanthina* do not deserve separate status as species and it is proposed to unite them, retaining the former at the lowest rank of forma, viz.

Rosa xanthina Lindley forma *xanthina*

R. xanthina Lindley f. *spontanea* Rehder in *Journal of the Arnold Arboretum*, 51: 209 (1924).

R. xanthina Lindley f. *normalis* Rehder & Wilson in *Sargent Plantae Wilsonianae*, 2: 342 (1915)

Rosa xanthina forma *hugonis* (Hemsl.) Roberts comb. nov.

R. hugonis Hemsley in *Curtis's Botanical Magazine*, 131: t. 8004 (1905).

Neither the hybrid *R. primula* $\times$ *R. hugonis* 13.57.1., representing a cross between *R. ecae* sensu amplo and *R. hugonis* sensu amplo nor *R. sericea* $\times$ *R. primula* 7.56., representing a cross between *R. sericea* sensu amplo and *R. ecae* sensu amplo, showed any sign of F_1 sterility. Furthermore, it was concluded from a study of *R. xanthina* $\times$ *R. sericea* 18.57.E. and *R. sericea* $\times$ *R. xanthina* 13.59., which represent crosses between *R. sericea* sensu amplo and *R. xanthina* sensu amplo, that, if there are any sterility barriers between the

parental species, they are incomplete. *R. sericea* sensu amplo, *R. ecae* sensu amplo and *R. xanthina* sensu amplo were found to differ consistently in their morphology. If these species were not, by some means, reproductively isolated, it might be anticipated that these differences would be blurred by introgressive hybridization. As no evidence of effective sterility barriers was found in these studies of F_1 hybrids, it is postulated that reproductive isolation may be effected through spatial separation. Distributions of the three species overlap in China, but separation may be achieved in the region of overlap, perhaps because of different ecological preferences.

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REFERENCES

- BOULENGER, G. A., 1934. Roses d'Asie. *Bulletin du Jardin botanique de l'Etat, Bruxelles*, 13: 165-266.
- CLELAND, R. E., 1962. The cytogenetics of *Oenothera*. *Advances in Genetics*, 11: 147-237.
- DARLINGTON, C. D. & WYLIE, A., 1955. *Chromosome atlas of flowering plants*. London: Allen & Unwin.
- ERLANSON, E. W., 1933. Chromosome pairing, structural hybridity and fragments in *Rosa*. *Botanical Gazette*, 94: 551-566.
- HARMS, F., 1956. The origin of *Rosa pernetiana*. *Rosenjahrbuch*, 14: 118-21.
- HURST, C. C., 1925. *Experiments in genetics*. Cambridge.
- JONES, K. & BORRILL, M., 1962. Chromosomal status, gene exchange in *Dactylis*, 3. The role of the inter-ploid hybrids. *Genetica*, 32: 296-322.
- REHDER, A., 1940. *Manual of cultivated trees and shrubs*, 2nd ed. New York: Macmillan.
- REHDER, A. & WILSON, E. H., 1915. *Rosa* L. in C. S. Sargent (Ed.), *Plantae Wilsonianae*: 304-45. Cambridge, Mass: Publics. Arnold Arboretum No. 4.
- ROBERTS, A. V., 1975. The nature and taxonomic significance of the system of inheritance in *Rosa nanothamnus* (Rosaceae). *Botanical Journal of the Linnean Society*, 71: 59-66.
- ROWLEY, G. D., 1959. Some naming problems in *Rosa*. *Bulletin du Jardin botanique de l'Etat Bruxelles*, 29: 205-11.
- ROWLEY, G. D., 1960a. Aneuploidy in the genus *Rosa*. *Journal of Genetics*, 57: 258-68.
- ROWLEY, G. D., 1960b. Triploid Garden Roses. *Rose Annual*, 45: 108-13.
- SOKAL, R. R. & SNEATH, P. H. A., 1963. *Principles of numerical taxonomy*. San Francisco & London: W. H. Freeman.
- TACKHOLM, G., 1922. Zytologische studien uber die gattung *Rosa*. *Acta Horti Bergiani*, 7: 97-381.
- VAVILOV, N. I., 1951. *The origin, variation, immunity and breeding of cultivated plants*. New York: Ronald Press Co.
- WULFF, H. D., 1954. Cytologische Untersuchungen an einer fertilen triploiden Rose. *Planta*, 44: 472-90.