Effects of Ionizing Radiations on Roses II. Meiotic studies on rose cultivar 'Pink Parfait' and its gamma ray induced mutants

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Genus *Rosa* was the first among ornamental plants which attracted the attention of early cytologists (Rowley 1967). Cytological investigations on roses have been made by several workers (Tächholm 1920, 1922, Blackburn and Harrison 1921, Hurst 1925, 1928, 1941, Erlanson 1928, 1930, 1931, 1933, 1938, Blackhurst 1948, Wulff 1951, Wylie 1954, Harrison 1958, Rowley 1961, Shahare and Shastry 1963, Darlington 1965, Salaria 1965, Klasterska 1969, Khanna and Lata 1971 and others). *Rosa* forms a regular polyploid series from $2\times$ to $8\times$ (Tächholm 1922). Besides various ploidy levels, the occurrence of aneuploidy, structural hybridity and irregular meiosis are common phenomenon in garden roses (Shahare and Shastry 1963). Their usual method of propagation by vegetative means has helped in the conservation of these genetic defects. According to Rowley (1967) garden roses are mainly comprised of tetraploids. These have been regarded as crypt-hybrids by Harrison (1958).

Materials and methods

Young flower buds from mutant and control plants were fixed in acetic-alcohol (1:3) to which a few drops of ferric chloride were added. Anthers were squashed in aceto-carmin employing the usual technique. Cytological studies were confined to metaphase I, anaphase I and II. Pollen fertility was determined on the basis of stainability in acetocarmine.

Results

Numerically the chromosomes in 'Pink Parfait' and the two mutants were uniform with $2n=4x=28$. An analysis of metaphase I configurations revealed that the PMCs from control and mutants comprised of quadrivalents, trivalents, bivalents and univalents in various ratios (Table 1). Seven types of chromosome associations were recorded in control and Light pink mutant. Except for one combination, others were common to both. The Deep pink mutant showed only five types of combinations which were, however, not different from the ones observed in the other clones.

The mean number of quadrivalents and univalents per PMC was higher in Light pink mutant (0.9 and 0.74 respectively) as compared with control and Deep

pink flowered mutant. The percentage of cells with two quadrivalents per PMC (Fig. 1) was highest (14.3) in Light pink mutant and lowest (2.44) in control. Similarly the cells with a chain quadrivalent (Fig. 2) and bivalents with or without

Table 1. Chromosome configurations at metaphase I in control and gamma ray induced mutants of 'Pink Parfait'

Clone	Chromosome associations in PMCs				Percentage of PMCs
	IV	III	II	I	
Control	2	—	10	—	2.44
	1	1	10	1	4.88
	1	—	12	—	51.22
	1	—	11	2	2.44
	—	—	14	—	29.27
	—	—	13	2	2.44
	—	—	12	4	7.31
Mean/PMC	0.61	0.05	12.44	0.41	
S.E.	± 0.084	± 0.069	± 0.18	± 0.32	
Deep pink flowered mutant	2	—	10	—	6.82
	1	1	10	1	2.28
	1	—	12	—	54.54
	—	—	14	—	18.18
	—	—	13	2	18.18
Mean/PMC	0.70	0.023	12.36	0.39	
S.E.	± 0.09	± 0.023	± 0.16	± 0.12	
Light pink flowered mutant	2	—	10	—	7.15
	2	—	9	2	7.15
	1	1	10	1	4.76
	1	—	12	—	40.48
	1	—	11	2	16.66
	—	—	14	—	11.66
	—	—	13	2	11.90
Mean/PMC	0.90	0.048	11.74	0.74	
S.E.	± 0.094	± 0.033	± 0.21	± 0.16	

Table 2. Summary of anaphase I and II data in 'Pink Parfait' and its gamma ray induced mutants

Clone	Total no. of PMCs	Anaphase I			Anaphase II				Ab-normal PMCs %	Pollen fertility %
		Percentage of cells with			Percentage of cells with					
		Unequal segregation	Preco-cious dis-junction	Lag-gard	Bridge	Unequal segregation	Lag-gard	Bridge		
Control	94	7.45	27.65	14.9	0	6.38	2.12	3.2	61.7	31.5
Deep pink flowered mutant	135	6.0	8.2	22.2	0.7	4.4	1.5	0.0	43.0	23.5
Light pink flowered mutant	90	2.2	20.0	12.3	1.1	3.3	1.1	1.1	41.1	22.0

Figs. 1-8. 1-2: Metaphase I. 2 IV+9 II+2 I. (1, light pink mutant); 1 IV+11 II+2 I. (2, deep pink mutant). 3-6: Anaphase I. 14:14. (3, light pink mutant); 13: 14+1 laggard univalent. (4, deep pink mutant); 3 laggard univalents. (5, control); 14: 14 with bridge and fragment. (6, light pink mutant). 7, anaphase II with 2 laggards (control). 8, pollen grains (light pink mutant). Fig. 1-7 $\times 1835$ and Fig. 8 $\times 725$.

trivalent and univalents or ring quadrivalent were commonly observed in all the three clones (Figs. 1 and 2). Deep pink mutant showed highest percentage of such associations (54.54). A reduction of a bivalent in 2.44% PMCs of control and 11.9% of Light pink mutant was balanced by two univalents. Combinations like 1 IV+II II+2 I were not observed in Deep pink mutant while such instances were observed in control and Light pink mutant. The frequency of chain quadrivalents was higher than the rings. PMCs with 14 bivalents were fewer (11.9%) in Light pink mutant while a higher number was recorded in the other mutant (18.18%) and control (29.27%). Occurrence of univalents in control was lower as compared to the mutants.

The analysis of anaphase I showed that 61.7% PMCs were abnormal in 'Pink Parfait' (Table 2). The Deep and Light pink mutants gave a lower percentage of aberrant cells as compared to control i.e. 43.0%, 41.1% respectively. Frequency of PMCs with equal segregation of chromosomes (Fig. 3) at anaphase I was higher (97.8%) in Light mutant. Unequal segregation was observed in 7.45% PMCs of control, 6% of Deep and 2.2% of Light pink mutant. Precocious disjunctions (Fig. 2) were highest in control (27.65%) as compared to the mutants. The frequency of laggards (Fig. 4) was found to be highest (22.2%) in Deep pink mutant and lowest (12.3%) in the other mutant. The PMCs of control (Fig. 5) occupied the intermediate position. Bridges (Fig. 6) were observed at anaphase I in both the mutants. At anaphase II, the percentage of PMCs with unequal segregation and laggards (Fig. 7) was highest in control. Laggards were recorded only in 3.3% of PMCs of Light pink mutant. No bridges were observed in Deep pink mutant while in the other mutant 1.1% PMCs had bridges. The frequency of bridges at anaphase II was found to be highest in control. Pollen fertility was highest (31.5%) in control and lowest (22.0%; Fig. 8) in Light pink mutant.

Discussion

The study of the pairing behaviour of chromosomes at metaphase I in 'Pink Parfait' suggests it to be a segmental allotetraploid. Such instances have been recorded in various plants by Stebbins (1947), Celarier (1957) and in roses by Khanna and Lata (1971) and Lata and Gupta (1975). The high incidence of chromosomal aberrations accompanied with high sterility in 'Pink Parfait' further indicates that hybridity has played an important role in the evolution. The pedigree of 'Pink Parfait' also explains the reason for highly heterozygous nature of this cultivar (McFarland 1965).

The number of bivalents per PMC was found to be decreased, with a consequent increase in the quadrivalents. Such a variation has been recorded after irradiation by Celarier (1957), Lata and Gupta (1975) and others. These changes in chromosome configurations can be attributed to the translocations induced by radiations. Heteromorphic bivalents were observed in all the three clones. The frequency of univalents per PMC increased in the mutants as compared to the control. This again, can be due to chromosomal changes brought by irradiation resulting in reduced pairing.

The number of lagging chromosomes varied from 1 to 2 in different clones. Although observed both at anaphase I and II, the highest percentage of these was recorded in anaphase I in the Deep pink mutant. The univalent being the primary source of these laggards is clear from the fact that their frequency was lowest in control which also had the least percentage of univalents. At anaphase I, the bridges were more frequent in mutants as compared to the control. The occurrence of these inversion bridges shows that the structural differences in some cases were not sufficient to prevent preferential pairing (Celarier 1957). Similar structural changes in chromosomes after irradiation in plants have been recorded by Swaminathan (1961) and Nilan (1968).

Irregular male meiosis is known to result in high pollen sterility which is prevalent in roses (Shahare and Shastry 1963). Khanna and Lata (1971) and Lata (1972) have reported high sterility in cultivar 'Pink Parfait'. The reduced fertility in the mutants as compared to the control shows that irradiation had deleterious effects on the pollen fertility. The higher sterility in gamma induced mutants could be due to genetic, chromosomal and both the effects of radiations. Radiation induced sterility has been recorded by a large number of mutation breeders (Beard *et al.* 1958, Vasudevan *et al.* 1968 and Lata and Gupta 1975).

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Summary

A comparative study of the male meiosis in the rose cultivar 'Pink Parfait' and its two, Deep and Light pink flowered mutants was conducted. 'Pink Parfait' was found to be a segmental allotetraploid. The frequency of univalents and quadrivalents at metaphase I was higher in the mutants. Anaphase I revealed a higher incidence of lagging chromosomes in Deep pink mutant and an increased frequency of chromosome bridges in both the mutants. The deleterious effects of radiations were evident from the higher percentage of chromosomal aberrations and pollen sterility found in mutants as compared with the control.

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