

Phenotypic and Cytologic Analyses of Spontaneous Mutations of the 'Montmorency' Cherry (*Prunus cerasus* L.)

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Abstract. Eleven suspected somatic mutations, consisting of 7 trees and 4 individual branches of the 'Montmorency' cherry, were compared with standard 'Montmorency' for phenotypic traits including tree height, trunk circumference, leaf area, crotch angle, resistance of flowers to frost injury, pollen germination, fruit set, individual fruit weight, fruit removal force, and yield. Differences occurred between some variants and 'Montmorency' controls for all characters except fruit removal force. All variants were tetraploid ($2n = 32$), as is standard 'Montmorency', with no evidence of cytochimerism.

Spontaneous somatic variants (sports) have been reported in fruit crops for over 350 years (4). However, only within the last 50 years have horticulturalists fully acknowledged somatic mutation as a means of cultivar improvement. Historically, more sports have been selected in apples than in other deciduous fruit species with many commercially important apple cultivars so originating (2). Variants also occur in other deciduous fruits, including the 'Montmorency' tart cherry, the subject of this investigation.

'Montmorency', which comprises over 95% of all United States commercial tart cherry orchards (1), exhibits considerable somatic variation in both tree and fruit characters (7). Studies to determine the commercial potential of 'Montmorency' variants began at the Michigan Agricultural Experiment Station in 1922 (6). Little quantitative data were obtained to determine the extent that the variants differed from 'Montmorency', nor were causes for observed phenotypic differences investigated. The objectives of our study were to verify that selected 'Montmorency' variants differed from the standard cultivar in quantitative characters, and to determine if they differed in ploidy from standard tetraploid ($4x = 2n = 32$) 'Montmorency'.

Materials and Methods

The 11 variants were grouped as follows: (a) spur-type growth with normal to heavy yield ('AE-408', 'AE-414', 'HN-418' and 'Idaspur') or with very limited yield ('MC-14'); (b) standard-type growth with consistent heavy yields ('MC-5', 'MC-9', and 'AE-415'); (c) prolonged blooming and ripening ('MC-13' and 'RM-409'); and (d) hybrid-type with both *P. cerasus* and *P. avium* L. tree characteristics, vigorous growth, limited cold hardiness, early blooming, self-sterility, and early ripening, dark,

red-juiced fruit ('MC-15'). Ten variants were selections from Michigan orchards and one commercial nursery selection, 'Idaspur', from an Idaho orchard.

Five of the variants ('AE-408', 'AE-414', 'AE-415', 'RM-409', and 'HN-418') are recent discoveries and were therefore compared to standard 'Montmorency' in a commercial nursery for height, trunk circumference, crotch angle, and leaf area. Six ('MC-5', 'MC-9', 'MC-13', 'MC-14', 'MC-15', and 'Idaspur') had been propagated and planted at the Horticultural Research Center in East Lansing, Mich. in 1969. Trunk circumference, number of flowers/cm² branch cross-sectional area, living flowers/100 buds in May, pollen germination, fruits/100 flowers in July, fruit removal force, fruit weight, yield, and leaf area were determined (Table 1). For 'Idaspur' only pollen germination, leaf area, and fruit removal force were determined.

Living flowers/100 flower buds in May (an indication of frost tolerance), fruits/100 living flowers in July (a measurement of fruit set), and fruit weight were obtained from 2 exposed branches: one on the north, the other on the south side of each of 4 trees of each variant and 'Montmorency'. Living flowers/100 flower buds in May was obtained by counting the number of viable flowers in a sample or about 200 flower buds at the distal end of the branches. Fruits were counted on the same branches in July. Flowers per cm² were calculated by counting the number of flowers per branch and dividing this by the cross-sectional area in cm² of the base of the branch. Fruit weight was the average of 20 fruits per tree (10/limb). Yield was determined as the total weight of all fruit on each tree.

Fruit removal force was obtained from 50 randomly selected fruit from 3 trees of each selection. Pull force was measured using a Mechanical Force Gauge, Model L-1000M (8). Measurements were taken at 2 different times during fruit maturation.

For pollen germination, flowers were randomly selected from 3 trees of each selection 1 day prior to full bloom and the anthers were placed in 13 × 100 mm culture tubes in 10 ml of an aqueous solution of 15% sucrose and 10 ppm boric acid. The tubes were shaken, releasing the pollen, and the solution was pipetted onto 1% agar blocks containing the same solution. The blocks were incubated at 25°C and 100% relative humidity for 18 hours after which percent germination was recorded.

For leaf area, 5 trees of each variant (3 for 'Idaspur') and 'Montmorency' were used. The 3rd, 5th, and 7th leaf from the apices of southerly exposed branches, 3 m above soil level, were measured. Trunk circumference was measured 20 cm above the soil level.

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Table 1. Comparison of vegetative and reproductive characteristics of 6 variants of 'Montmorency' sour cherry with those of standard 'Montmorency'. Budded trees planted at E. Lansing in 1969, data recorded in 1979.

Clone	Living flowers/100 buds in May	Fruits/100 living flowers in July	Wt/fruit (g)	No. flowers/cm ² cross-sectional area	Yield/tree (kg)	Fruit removal force (g)		Pollen germin. (%)	Area per leaf (cm ²)	Trunk circumf. (cm)
						July 18–July 23	July 23–July 23			
Montmorency	232 ab	54 b	4.3 b	121 bc	23 c	381 a	276 a	26 a	34 c	36 a'
MC-5	228 ab	35 ab	5.2 c	147 bc	28 c	487 b	254 a	35 bc	31 c	47 ab
MC-9	191 ab	41 b	5.3 c	116 bc	20 bc	314 a	264 a	37 c	30 b	39 a
MC-13	244 b	37 ab	3.4 a	170 c	14 b	---	---	26 a	28 a	40 a
MC-14	233 ab	10 a	4.4 b	7 a	1 a	---	---	27 ab	31 c	40 a
MC-15	154 a	5	---	92 b	0 a	---	---	23 a	48 c	54 c
Idaspur	---	---	---	---	---	460 ab	270 a	56 d	29 b	---

'Mean separation within column by Duncan's multiple range test, 5% level.

In nursery measurements, 10 trees chip-budded onto *P. mahaleb* L. seedlings in 1978 were used for each variant and 'Montmorency'. Uniform sandy loam soil and standard commercial practices were employed. Measurements were made in late Sept., 1979, after growth had ceased. Trunk diameter was measured with calipers at 3 cm above the graft union and then converted to circumference. Height was measured as the distance from the bud union to terminal of the central leader. Crotch angle degrees from vertical were obtained from the 1st, 2nd, and 3rd lateral branches from the apex of the central leader using a protractor. Average leaf area was obtained using the 3rd, 5th, and 7th leaf from the distal end and measured with a portable area meter (Li-Cor) (Table 2).

Ploidy of the histogenic layers was determined from chromosome counts of root tip squashes for L-II and L-III, with ploidy of L-I inferred from guard cell size (10). Root-tip squashes were obtained from self-pollinated seedlings for L-II determination (9) or from adventitious roots of hardwood cuttings for L-III determination (3, 5). Roots were fixed and stored in 3:1 absolute ethanol; glacial acetic acid. For chromosome counting, fixed roots were placed in 1N HCl at 60°C for 35 minutes, rinsed in water, and squashed in 1% acetocarmine. Guard cell size was measured from finger nail polish impressions. In determining ploidy of single limb mutants, both the normal sector and standard 'Montmorency' were used as controls.

Roots for L-II and L-III ploidy determinations were originally removed from 0900 to 1000 HR and from 1400 to 1500 HR. Low mitotic indices were found at these times making chromosome counting difficult. In roots sampled from 1200 to 1300 HR, mitotic index was greater, with many cells in metaphase and anaphase, allowing for easy chromosome counting.

Table 2. Height, diameter, crotch angle, and leaf area of 1-year-old nursery trees of 'AE-408', 'AE-414', 'AE-415', 'HN-418', and 'Montmorency' sour cherry.

Clone	Height (cm)	Trunk circumf. (cm)	Crotch angle (degrees from vertical)	Avg. leaf area (cm ²)
Montmorency	146 e ²	8.0 e	51 d	43 c
AE-415	129 d	7.5 de	48 cd	42 bc
RM-409	111 c	6.8 cd	45 bc	41 bc
HN-418	108 c	5.7 b	44 b	40 a
AE-408	94 b	6.4 bc	44 b	52 e
AE-414	67 a	4.7 a	38 a	50 d

'Mean separation within column by Duncan's multiple range test, 5% level.

Results and Discussion

'MC-5' and 'MC-9' had greater fruit weight and pollen germination but smaller leaf areas than 'Montmorency'. 'MC-5' also had a greater fruit removal force on July 18 and a larger trunk circumference. Neither selection had greater yields than 'Montmorency' in 1979.

'MC-13' had a smaller average fruit weight and leaf area than the standard 'Montmorency'. Since its distinguishing character is uneven ripening, average fruit size was expected to be smaller, because immature as well as mature fruit were included in the averaged sample.

'MC-15' differed from 'Montmorency' in all characters studied except pollen germination and number of flowers/cm² cross-sectional branch area. 'MC-14' differed from 'Montmorency' in number of flowers/cm² cross-sectional branch area, leaf area, fruit/100 living flowers in July yield/100 living flowers in July and yield/tree. These findings were expected since these variants have obvious morphological differences, one with characters more like *P. avium* and the other with unfruitful spurs.

'Idaspur' differed from 'Montmorency' in pollen germination and leaf area. Pollen germination was twice that of 'Montmorency', a possibly important characteristic if it remains consistent.

All of the nursery-grown variants differed from 'Montmorency' in one or more characteristics. 'AE-415' was found to differ only in height; its similarity to 'Montmorency' was not surprising since it was initially selected for consistent and apparently heavier annual yield and not for general tree structure.

'AE-408', 'AE-414', 'HN-418', and 'RM-409' differed significantly from 'Montmorency' in height, trunk circumference, crotch angle, and leaf area. These differences were readily visible in the nursery.

The 'Montmorency' controls in the nursery comparisons had been treated to eliminate known cherry viruses and the variants had not, and therefore the data may only indicate that virus-free nursery trees are more vigorous than infected ones. However, gross morphological differences of the variants, other than 'AE-415', suggest that this is unlikely. In other experiments in progress, 'Montmorency' trees infected with prune dwarf virus (PDV), *Prunus necrotic ringspot virus* (PNRSV), and green ring mottle (GRM) have not exhibited gross morphological differences of the magnitude reported here. PNRSV-infected and noninfected trees of 'AC-404', another spur-type mutant, were grown side-by-side with the virus-free trees continuing to express their spur-type phenotype.

The nursery selections are still under evaluation for spur development, yield, fruit quality, and winterhardiness. If the spur

character persists in grafted trees, their natural dwarfing tendency could prove valuable, since commercially acceptable dwarfing rootstocks have not been developed for cherries.

No ploidy differences were observed in the variants. The normal chromosome number ($2n = 32$) was found in L-II of all variants and in L-III of those variants which produced adventitious roots. Stomatal size was the same in all cases. None of the variants exhibited any traits characteristic of increased ploidy or cytochimerism (5).

L-I does not produce enough tissue to account for the changed phenotypes of the variants; therefore L-II and/or L-III must be involved. Since L-II gives rise to reproductive tissue, changes in this layer may explain changes in fruiting characteristics. Changes involving both spur-type growth and fruit characters ('RM-409' and 'MC-14') probably involve both L-II and L-III.

Based on quantitative measurements and visual observations we conclude that 'MC-13', 'MC-14', 'MC-15', 'AE-408', 'AE-414', 'HN-418', and 'RM-409' are different from the standard commercial strain of 'Montmorency'. The difference, however, is not due to a change in ploidy level but to other genetic changes.

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Unilateral Incompatibility between Red and Black Raspberries¹

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Abstract. The discrepancy in seed set observed in reciprocal crosses of red (*Rubus idaeus* L.) and black (*R. occidentalis* L.) raspberries verifies unilateral incompatibility. The strength of the incompatibility varies between cultivars; some red x black crosses yield a small percentage of seed set by simple cross-pollination, while others produce almost no seed. Although black cultivars have shorter pistils than red cultivars, the role of this factor in incompatibility is unknown. Bud pollination and heat treatments increased seed set in some normally incompatible red x black crosses.

The purple raspberry is an interspecific hybrid of the red raspberry, *Rubus idaeus*, and the black raspberry, *R. occidentalis* (1). Hybrid purple raspberries typically more closely resemble the black raspberry in a number of characteristics including growth habit, tip layering tendency, response to pinching, type of prickles, pubescence on the fruit surface, and hardiness (4). Purple raspberries are produced when the pollen parent is a red raspberry (3, 5, 7, 9, 10). If the black raspberry is used as the pollen parent, seed set usually fails, although a few reports indicated limited success (2, 6, 8). This problem of unilateral incompatibility is confounded in the oldest literature because the researchers often failed to describe the characteristics of their hybrids.

Unilateral incompatibility between raspberries has been investigated histologically by Zych (10), who found that in in-

compatible crosses of red x black raspberries (seed parent is always expressed first) restricted pollen tube growth occurs within the style. Black raspberry pollen tubes never grew more than one-third of the way down the styles of red raspberry flowers. Normal pollen tube growth was observed in the reciprocal crosses.

One of the authors (A. G. Otterbacher) observed that the pistils of black raspberry cultivars appeared to be slightly shorter than those of red raspberry cultivars. It was hypothesized that incompatibility may be due to the inability of pollen tubes of black raspberry to grow the entire length of red styles.

The objective of this investigation was to investigate unilateral incompatibility between red and black raspberry.

Materials and Methods

Flowers for pistil measurement were obtained in 1977 and 1978 by randomly collecting 10 flowers from each of the fall-bearing red raspberry cultivars 'Fallred', 'September', 'Heritage', 'Fallgold', and the black raspberry cultivars 'Bristol', 'New Logan', 'Plum Farmer', 'Allen', 'Black Hawk', 'Lowden', and 'Domack'³. Ten pistils from each flower were excised and measured from the tip of the stigma to the base of the ovary.

Field-grown 'Fallred' and 'Heritage' red raspberry and 'Allen' and 'New Logan' black raspberry plants were dug in the fall

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³Not a commercially released cultivar.