

Meiotic behavior of two grapevine somatic mutants with ornamental potential

Neiva I. PIEROZZI^{1*}, Mara FERNANDES MOURA²

¹Instituto Agronômico de Campinas (IAC), Centro de Recursos Genéticos Vegetais, 1481 Barão de Itapura Ave., Campinas, Brazil; neiva.pierozzi@sp.gov.br (*corresponding author)

²Instituto Agronômico de Campinas (IAC), Centro Avançado de Pesquisas e Desenvolvimento de Frutas, 1500 Luís Pereira dos Santos Ave., Jundiá, Brazil; mara.moura@sp.gov.br

Abstract

'Niagara Steck' and 'Niagara Variegada' are two grapevine varieties that arose spontaneously as somatic mutants of 'Niagara Rosada'. Berries characterize 'Niagara Steck' at young stages exhibiting a remarkable brown reticulated structure on the epidermis which develops into a brown-bronze russet-like structure as the berries ripe. The mature berries have strongly acidic flesh. 'Niagara Variegada' shows notable variegation in leaves and berries. Meiotic studies were carried out to ascertain if the mutant phenotypes could be related to any change in chromosome number, structure or other irregularity. Floral buds of both varieties at different developmental stages were collected and fixed for cytological analyses. Chromosome counts at diakinesis/metaphase I showed $n=19$ bivalents for both varieties. Univalent, trivalent, tetravalent chromosomes or chain configuration were not recorded ruling out the possibility of any alteration in the chromosome number or structure in both mutant varieties. However, low percentages of meiotic irregularities were recorded such as chromosome stickiness, laggards, non-oriented chromosomes, precocious chromosome segregation, tetrads with one microcyte, microspore fusion, and tetrad degeneration. The total percentage of abnormalities was higher in 'Niagara Variegada' (19.68%) than in 'Niagara Steck' (14.40%) which may have contributed to a lower percentage of pollen fertility (79.80%) when compared to 'Steck' (90.74%). The aforementioned varieties can be propagated by hardwood cuttings and constitute an interesting option for ornamentation of home backyards, patios, and gardens with the advantage that 'Niagara Variegada' bearing sweet edible berries.

Keywords: microsporogenesis; 'Niagara Steck'; 'Niagara Variegada'; ornamental grapevines; spontaneous mutants; *Vitis*

Introduction

The improvement of perennial fruits and ornamental plants is very often done following conventional breeding techniques. Grapevine (*Vitis vinifera* L.), an old woody perennial plant has been cultivated for millennia regarding different purposes such as table grapes for fresh consumption, raisins, jams, juice, and winemaking. New grapevine varieties have been developed through breeding programs. However, due to its long juvenile period that precedes the reproductive cycles, the vegetative propagation of grapevines by

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hardwood cuttings has been predominantly employed. By this approach the desirable genotypes are preserved, mostly intended for wine production, over decades. Nevertheless, somatic spontaneous mutations may sometimes appear during the very initial development of a bud producing a shoot with leaves, inflorescences, or berries with a different phenotype, the bud sports. In conformity with the new phenotype, breeders and growers are therefore able to select a whole branch or part of it for propagation via vegetative techniques such as hardwood cuttings thus preserving new morphological traits.

Among the spontaneous somatic mutations, those affecting berries have also attracted the attention of researchers and vine growers. 'Malian', for instance, occurred as a spontaneous somatic mutant of the grapevine 'Cabernet Sauvignon' with rosy berries (Walker *et al.*, 2006; Vezzulli *et al.*, 2012). The table grapes 'Redimeire' (Pires *et al.*, 2003) and 'Victoria LB' (Ferrara *et al.*, 2017) emerged due to a spontaneous mutation in 'Italia' and 'Victoria' grapevines, respectively, that accounted for the unexpected attractive long berries which became a new and profitable option. However, not very lucrative nor interesting, is the leaf variegation. It can appear as a spontaneous somatic mutation in a bud as in the 'Allepo' grapevine variety, early reported by Knight, (1808) or in the variegated Bruce's sport (Antcliff and Webster, 1962). Moreover, it may occur due to the expression of a recessive gene in seedling phase among the offspring of some interspecific hybrids (Reisch and Watson, 1984) or, either in regenerated plants from somatic embryogenesis (Kuksova *et al.*, 1997) or after some chemical and physical treatments (Sharma and Mukherjee, 1972; Sharma and Mukherjee, 1973; Marcotrigiano, 1997).

'Niagara Steck' and 'Niagara Variegada' are varieties that arose as spontaneous somatic mutations in a vineyard of the table grape 'Niagara Rosada' at different times. 'Niagara' grapevine ($2n=38$) is an interspecific hybrid which derived from an artificial crossing conducted in the USA at the end of 1868, in Niagara County, NY. Plants of *Vitis labrusca* 'Concord' were pollinated by 'Cassady' another hybrid (*V. labrusca* × *V. vinifera*), generating the first 'Niagara' grapevines (Hedrick, 1919; Maia, 2012). The first mutation observed in 'Niagara Rosada' was confined to the berry skin color which originated to 'Niagara Steck'. The mutation was observed in a single cane during the bunch and berries development. All the berries of the bunches displayed a brown-bronze reticulated russet-like epidermis with a notable brilliant light green cap on the vestigial pistil extremity (navel) in contrast to the epidermal matte color which resulted into a remarkable ornamental phenotype (Sousa, 1959). The berries, however, hold an uncommon acidity that persists even at the ripening stage. A distinct branch showing an outstanding variegation pattern in the leaves and berries was later detected in another grapevine producing 'Niagara Variegada'. The immature berries bear white and green stripes running along the length of the fruit. Considering these extremely different traits, the mutant shoots were propagated by hardwood cuttings and have been kept in the grape collection as new varieties and have ever since been studied with different approaches (Sousa, 1959; Sousa, 1996; Tofaneli *et al.*, 2011; Angelotti-Mendonça *et al.*, 2018).

Despite these outstanding features, there is hardly any information available about the meiotic process regarding these two mutant varieties that evaluate the regularity of this process and the percentage of pollen fertility. Therefore, meiotic studies were carried out in 'Niagara Steck' and 'Niagara Variegada' mutant varieties to ascertain if the mutant phenotypes could be related to any change in chromosome number, structure or to meiotic irregularity, since cases of meiotic irregularities have been reported in some mutants of other plant species. The presence of an abnormal unsatellited satellite chromosome, a tetravalent chromosome association at diakinesis and more than 50% of pollen sterility were described in unstable races of variegated *Petunia sp.* (Malinowski, 1935). The variegation observed in *Nicotiana* hybrids was related to the occurrence of dicentric chromosomes at meiosis (Ar-Rushdi, 1957). Bud sport mutants bearing fruits of assorted sizes or morphology are often related to the ploidy level that in many cases cause disturbances in the meiotic process producing seedless or almost seedless fruits. Larger ripened and seedless fruits of guava (*Psidium guajava*) were observed on a spontaneous triploid plant (Raman *et al.*, 1971) and in spontaneous autotetraploid grapevine (Staudt and Kassrawi, 1972), for instance. Conversely, smaller ripened fruits other than the diploid ones were noticed in

some aneuploid plants as well (D’Cruz and Rao, 1977). Therefore, the gain of a single extra chromosome in the diploid genome of a plant is enough to affect the fruit size and/or shape and other vegetative traits, as in trisomic tomatoes reported by Rick and Barton (1954) which displayed abnormal fruit shape and size along with meiotic abnormalities and pollen sterility percentage.

Materials and Methods

Plant materials

‘Niagara Steck’ (hereafter ‘Steck’, Figure 1) and ‘Niagara Variegada’ (hereafter ‘Variegada’, Figure 2) varieties are spontaneous somatic mutants of ‘Niagara Rosada’ (hereafter ‘Rosada’) variety, a hybrid table grape (*Vitis labrusca* x *V. vinifera*). ‘Niagara’ varieties belong to IAC (Instituto Agronômico de Campinas) *Vitis* germplasm collection at the Fruit Center (IAC-Centro Avançado de Pesquisa e Desenvolvimento de Frutas) in the city of Jundiaí, state of São Paulo.

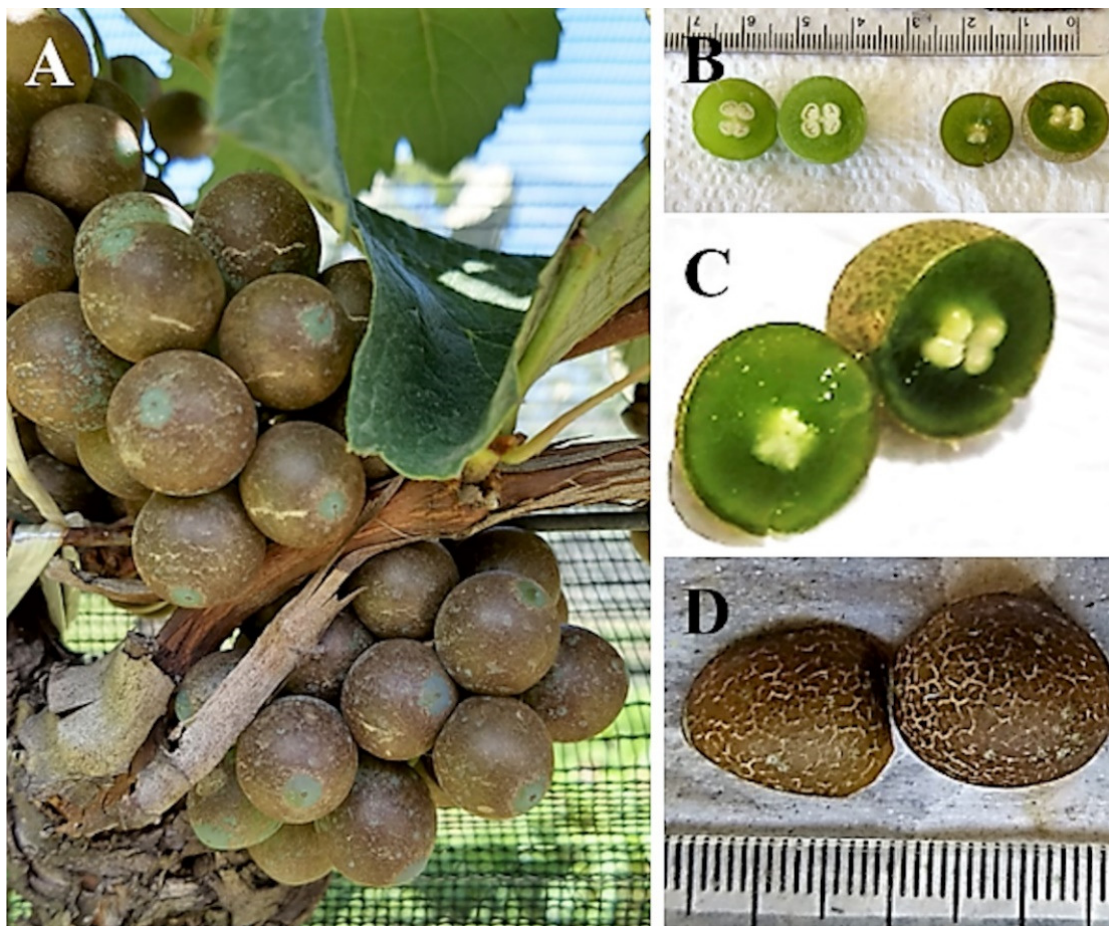


Figure 1. ‘Niagara Steck’ (‘Steck’). (A) Bunch with berries showing the brown-bronze reticulated russeting-like epidermis; (B) Cross section of immature berries; (C) Detail of the flesh and skin; (D) Reticulated russeting-like epidermis (skin) of a ripe berry

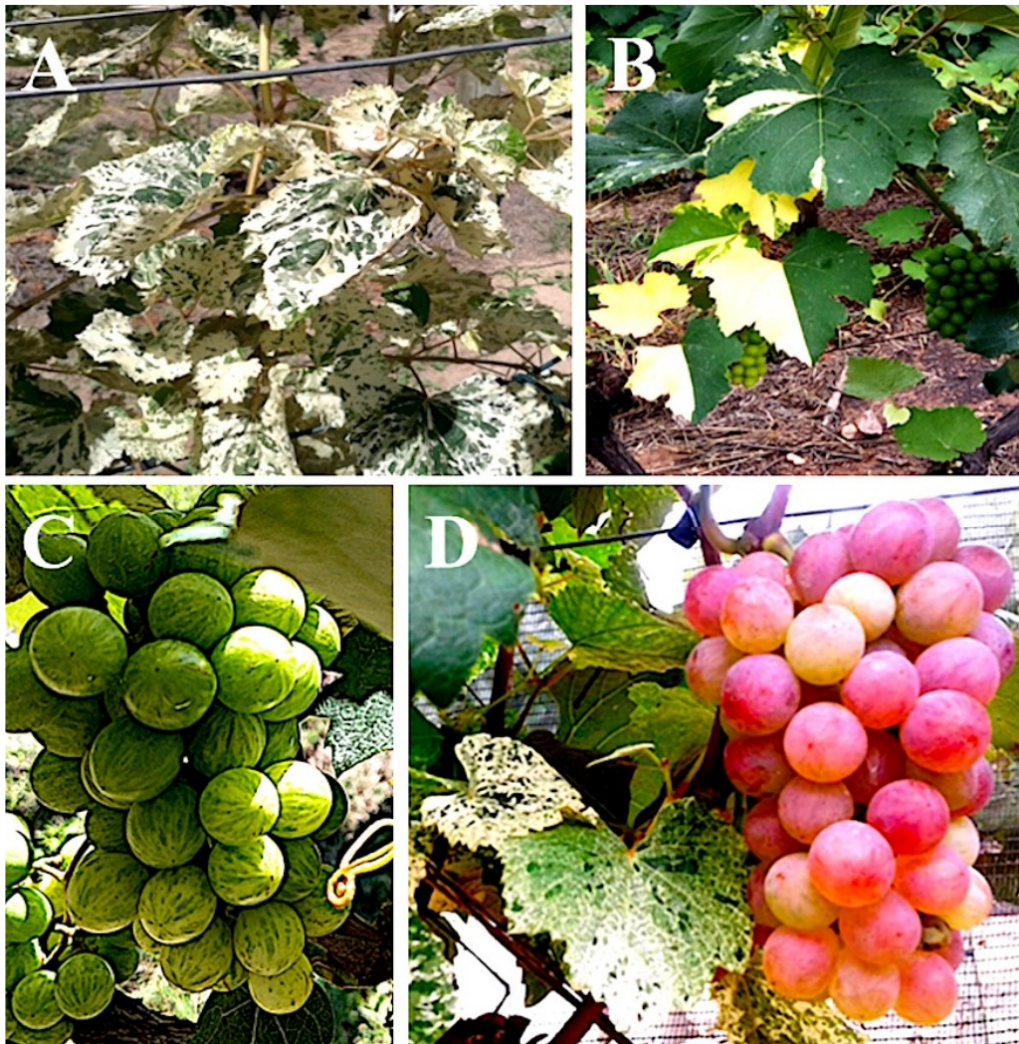


Figure 2. 'Niagara Variegada' ('Variegada'). (A) and (B) Plants with variegated leaves; (C) Immature bunch with stripe variegated berries; (D) Bunch with mature berries showing some light stripes

Cytological procedures

Young inflorescences with flower buds at different stages of development of both 'Steck' and 'Variegada' mutant varieties were collected over two years, 45-50 days after the bud burst which occurred at the end of August. The inflorescences were fixed in 3:1 Carnoy solution (ethanol 99%: acetic acid solution, respectively) and stored at -20°C . The slides for cytological studies were prepared using anthers detached from the buds with the aim of a stereo microscope and squashed in a 1.2% acetic carmine solution. The analyses were carried out from pachytene to telophase II phases and 300 cells were scored in each phase to each variety and the values were expressed as a percentage. The mean percentage value of normal tetrads was calculated regarding 3,000 tetrads for each variety. Similarly, the mean percentage values of pollen grain fertility were estimated by pollen stainability using Alexander A2 dye and 500 grains were counted for each variety. Besides, the mean value of pollen area (μm^2) was also calculated considering 125 fertile grains for each variety. The chiasma average number per cell and per chromosome were calculated considering 100 cells for each variety at metaphase I. The mean percentage values of terminal and subterminal chiasmata per cell were also calculated. The F-test was applied at a 5% level to compare the mean values obtained for each variety seeing: (a) the percentage of normal tetrads; (b) the percentage of pollen fertility and (c) pollen area; (d) chiasma number per chromosome (e) and

chiasma number per cell; (f) the percentage of terminal and subterminal chiasmata per cell. Photomicrographs of some meiotic phases and also tetrads and pollen grains were taken using a black and white Ilford film ASA 125 in Vanox 'Olympus' photomicroscope.

Results

Nineteen bivalents were viewed at diakinesis in both varieties (Figures 3A, 3B, 4A and 4B) ruling out the possibility of any change in chromosome number in these spontaneous somatic mutant varieties. There was no chromosome pairing irregularity observed, no univalent, trivalent nor tetravalent chromosomes at diakinesis or metaphase I either in 'Steck' or 'Variegada' which could, for instance, be related to aneuploidy. Ring or chain configurations between bivalents at diakinesis or metaphase I, characteristic of a chromosome translocation was also not perceived. Besides, no chromosome bridges with fragments at anaphases I or II were observed. However, other abnormalities, though at a low percentage, were noticed during the meiotic phases in both 'Steck' and in 'Variegada' and are summarized in Table 1, and detailed data is provided in Table 2. There were slightly higher percentage values of abnormalities in the 'Steck' and 'Variegada' microsporocytes observed at metaphase I and anaphase I and at in addition to metaphase II and anaphase II (Table 1). Chromosome stickiness at diakinesis, three sets of chromosomes at anaphase I, and five sets of chromosomes at telophase II were only seen only in 'Steck' (Table 2, Figures 3A, 3F, 3N, respectively).

Table 1. Mean percentage of abnormal cells (PMCs) at meiotic phase of 'Steck' and 'Variegada'

% PMCs with abnormalities at some meiotic phases									
Variety	DIA	MI	AI	TI	MII	AII	TII	IV	Total
'Steck'	0.70	4.36	3.96	0.42	1.36	2.29	0.28	1.03	14.40
'Variegada'	0.00	4.34	4.36	0.32	3.43	2.84	0.00	4.39	19.68

DIA (diakinesis), MET I and II (metaphase I and II), ANA I and II (anaphase I and II), TELO I and II (telophase I and II), IV (tetrad).

The varieties also differed significantly with respect to the mean value for pollen area and pollen fertility (Table 3). The results reported that 'Steck' showed the lowest mean value for pollen size area ($299.66 \mu\text{m}^2$) (Figure 3S) when compared to 'Variegada' ($391.13 \mu\text{m}^2$) (Figure 4R) which depicted the highest mean value. Regarding pollen fertility, 'Variegada' displayed the lowest mean value (79.80%) differing significantly from 'Steck' (90.74%) which exhibited the highest mean value.

Despite the small size of chromosomes, it was possible to count the chiasmata on the bivalent arms of the mutant varieties which did not differ regarding the mean value for the percentage of chiasma number per cell and per chromosome (Table 3). Only terminal and subterminal chiasmata were observed in 'Steck' and 'Variegada' with a prevalence of two terminal chiasmata, each one located at the end of opposite arms of the bivalent in 64.63% of the cells scored. Additionally, followed by a lower percentage of two subterminal chiasmata (nearly 18%), by one terminal and one subterminal chiasmata, each one at the end of opposite arms of the bivalent (more than 16%). Two chiasmata on the same chromosome arm were not noticed.

The abnormalities common to both varieties and detailed in Table 2 were (a) chromosome stickiness at metaphase I (Figures 3B and 4B), at anaphase I (Figures 3E and 4F), at anaphase II (Figures 3K and 4J); (b) non-oriented chromosomes at metaphase I (Figures 3C with arrowhead and 4D) and at metaphase II (Figures 3I and 4K); (c) laggard chromosomes at anaphase I (Figures 3D and 4E), at telophase I (Figures 3G and 4G) and at anaphase II (Figures 3J and 4I); (d) precocious chromosome segregation at metaphase I (Figures 3C with arrows and 4C) and at metaphase II (Figures 3L and 4H); (e) five sets of chromosomes at anaphase II (Figures 3M and 4L); (f) tetrads with one microcyte (Figures 3P and 4M); (g) tetrads with microspore fusions (Figures 3Q and 4O); (h) degeneration of fused microspores followed by the rising of a large vacuole (Figures 3R and 4P). In 'Variegada' the process of degeneration began with a decrease in microspore size afterward by the fusion

of the microspores and a subsequent shrinkage (3.64%) (Figures 4O to 4Q) (Table 2) that reflected in a lower mean value of normal tetrads ($95.61 \pm 2.18\%$) (Figure 4N) when compared to 'Steck' ($98.97 \pm 0.47\%$) (Figure 3Q, Table 3). The total percentage of abnormalities observed was higher in 'Variegada' (19.68%) than 'Steck' (14.40%) (Table 1).

Table 2. Mean percentage values of different types of abnormalities observed at meiotic phases in 'Steck' and in 'Variegada'

Phase with abnormalities	Steck %: % chrom. abnorm.	Variegada %: % chrom. abnorm.
Diakinesis:		
. Chromosome stickiness	0.70	0.00
Metaphase I:		
. Chromosome stickiness	0.53	0.31
. Non oriented chromosomes	2.64	3.10
. Precocious chromosome segregation	1.19	0.93
Anaphase I:		
. Chromosome stickiness	2.73	2.17
. Chromosome laggards	1.09	2.19
. 3 sets of chromosomes	0.14	0.00
Telophase I:		
. Chromosome laggards	0.42	0.32
Metaphase II:		
. Non oriented chromosomes	0.69	2.18
. Precocious chromosome segregation	0.67	1.25
Anaphase II:		
. Chromosome stickiness	1.71	0.95
. Chromosome laggards	0.30	0.63
. 5 sets of chromosomes	0.28	1.26
Telophase II:		
. 5 sets of chromosomes	0.28	0.00
Tetrads:		
. With one microcyte	0.77	0.06
. Fusion of microspores	0.13	0.69
. Microspore degeneration	0.13	3.64

Table 3. Mean values for chiasma number per cell and chromosome, and mean percentage values of normal tetrads, pollen grain fertility and pollen area for 'Steck' and 'Variegada'

Chiasma number:	'Steck' cv.	'Variegada' cv.
. per cell	37.89 ± 0.31^a	38.00 ± 0.00^a
. per chromosome	1.99 ± 0.02^a	2.00 ± 0.00^a
Percentage (%) of chiasmata:		
. 2 terminal	64.63 ± 3.69^a	64.63 ± 1.12^a
. 2 subterminal	18.32 ± 2.15^a	18.10 ± 0.92^a
. 1 terminal + 1 subterminal	16.53 ± 2.21^a	17.27 ± 0.84^a
% Normal Tetrads:	99.61 ± 0.47^a	95.61 ± 2.18^b
% Pollen fertility	90.74 ± 6.97^a	79.80 ± 0.84^b
Pollen area	$299.66 \mu\text{m}^2 \pm 37.68^a$	$391.13 \mu\text{m}^2 \pm 28.19^b$

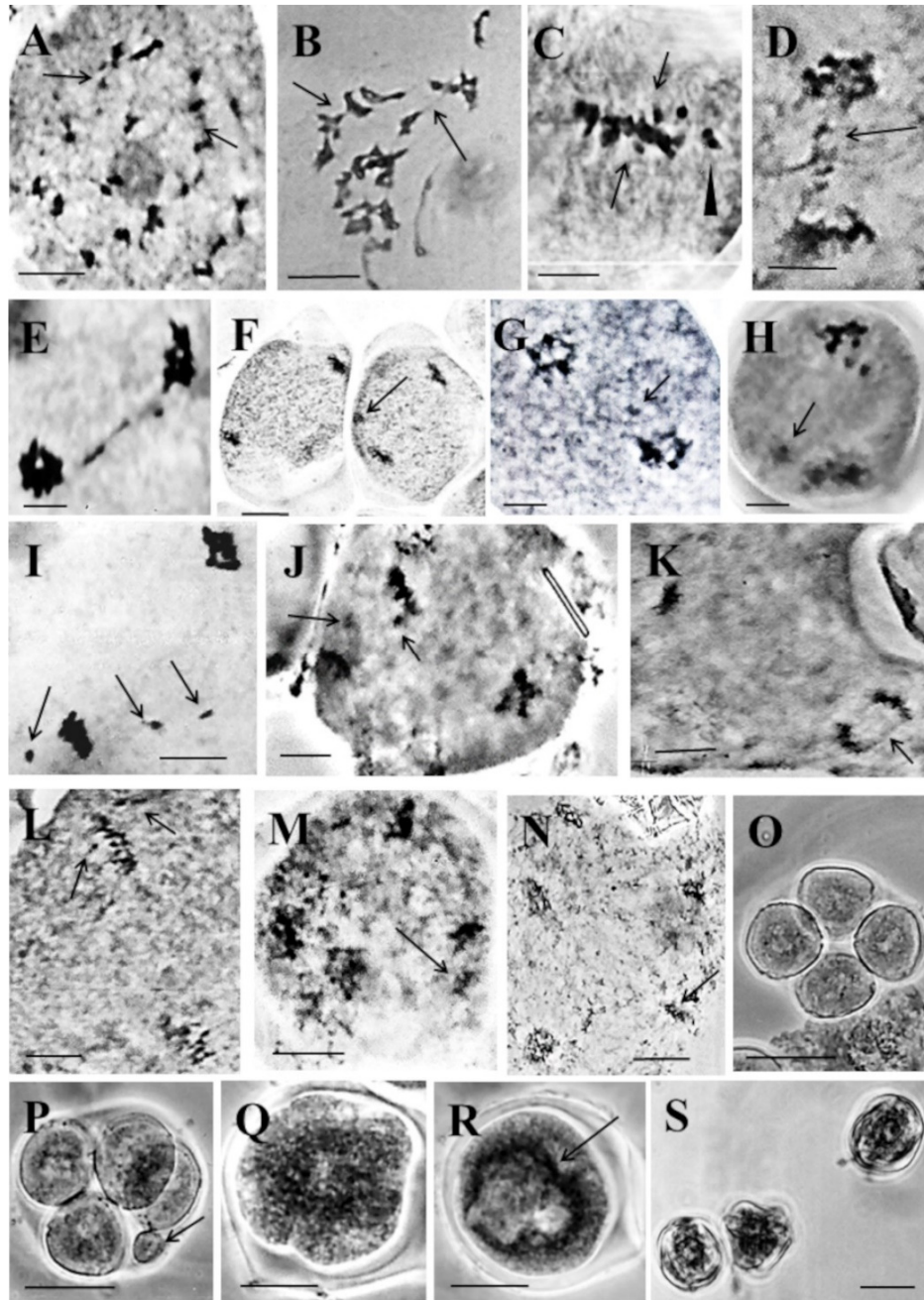


Figure 3. Some meiotic phases of 'Steck'. (A) Chromosome stickiness at diakinesis and (B) at metaphase I; (C) Non-oriented chromosomes (arrowhead) and precocious chromosome segregation (arrows) at metaphase I; (D) Laggard chromosomes at anaphase I; (E) Chromosome stickiness at anaphase I; (F) Three sets of chromosomes at the late anaphase I; (G) Laggard chromosomes at telophase I; (H) Three sets of chromosomes at telophase I; (I) Non-oriented chromosomes at metaphase II; (J) Laggard chromosomes at anaphase II; (K) Chromosome stickiness at anaphase II; (L) Precocious chromosome segregation at metaphase II; (M) Five sets of chromosomes at anaphase II; (N) Five sets of chromosomes at telophase II; (O) Normal tetrad with four micropore; (P) Tetrad with one microcyte; (Q) Fused microspore; (R) Fused microspore with a large vacuole; (S) Pollen grains. Bar: A to N=10 μm ; O to R=40 μm ; S= 50 μm .

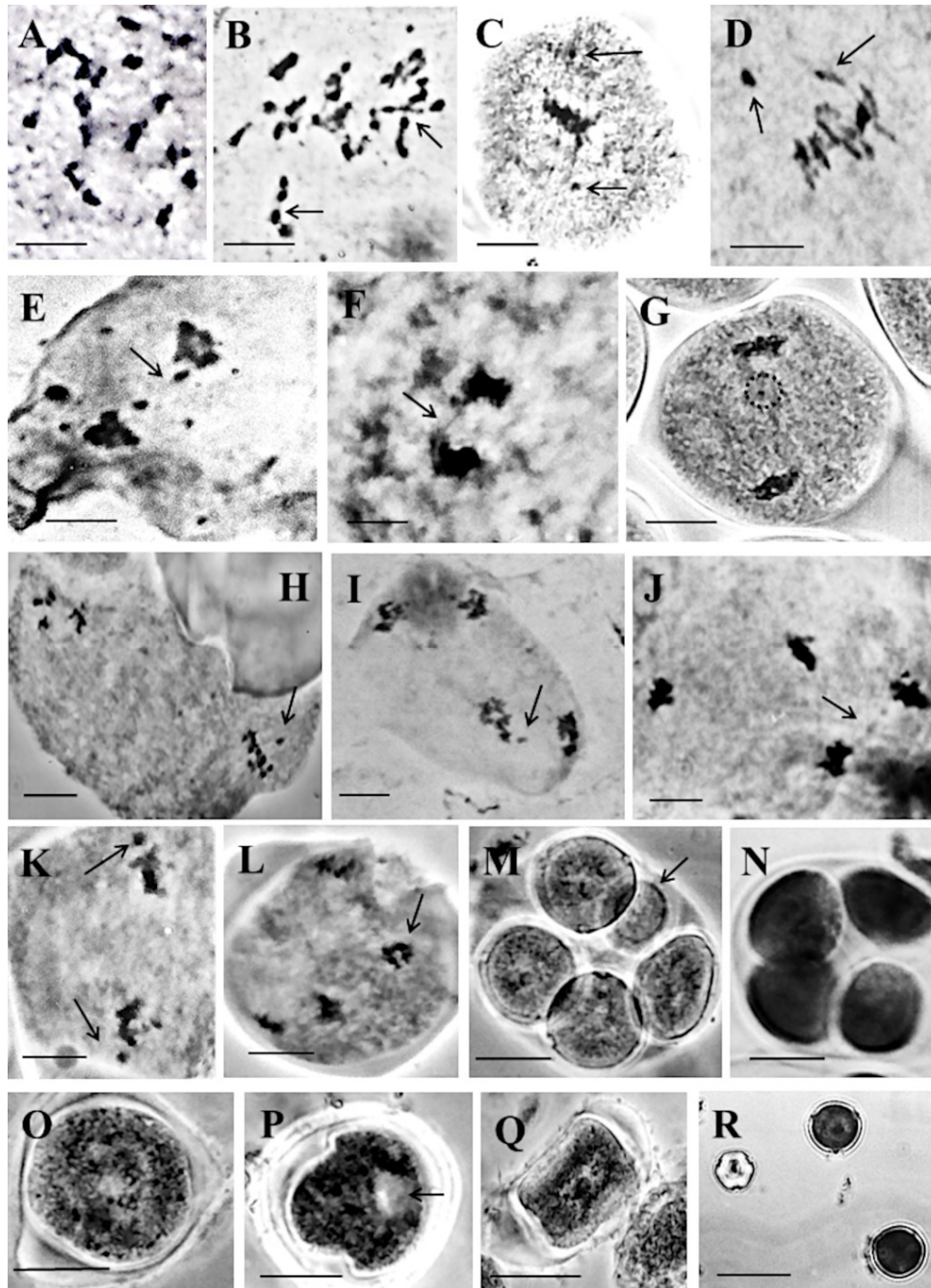


Figure 4. Some meiotic phases of 'Variegada'. (A) Diakinesis; (B) Chromosome stickiness at a polar metaphase I; (C) Precocious chromosome segregation at metaphase I; (D) Non-oriented chromosome at metaphase I; (E) Laggard chromosomes at anaphase I; (F) Chromosome stickiness at anaphase I; (G) Laggard chromosome at telophase I; (H) Precocious chromosome segregation a metaphase II; (I) Laggard chromosome at anaphase II; (J) Chromosome stickiness at anaphase II; (K) Non-oriented chromosome at metaphase II; (L) Five sets of chromosome at anaphase II; (M) Tetrad with one microcyte; (N) Normal tetrad; (O) Fused microspore; (P) Degenerated fused microspore with a large vacuole; (Q) Shrunk fused microspore; (R) Pollen grains. Bar: A to L=10 μ m; M to Q=20 μ m; R=50 μ m

Discussion

Studies carried out in spontaneous or induced variegated grapevine mutants are mainly characterized by the description of the morphological and anatomical characteristics of the mutant plants (Antcliff and Webster, 1962; Sharma and Mukherjee, 1973). However, few of these studies were carried out to understand the genetic basis of the variegation (Reisch and Watson, 1984; Filler *et al.*, 1994, Olson *et al.*, 2021). Even though this information contributed to a better characterization of variegated grapevines, the regularity of the meiotic process or the percentage of pollen fertility were not investigated to evaluate if the mutation producing the variegated trait could have or not affected the meiosis or pollen fertility. The analyses achieved on the meiosis of the spontaneous somatic mutant 'Variegada' revealed that the variegation trait is not associated to a serious disturbance in the meiotic process, which could affect the pollen fertility. Though not a rule since meiotic irregularities have been reported in some variegated plants. A high percentage of meiotic abnormalities such as an expressive percentage of bridges and laggard chromosomes accompanied by a reduction in mean value of chiasma number per cell, as well as by a significant decrease in pollen fertility were observed in the variegated plants of *Trillium grandiflorum* (Chinnappa, 1982). In addition, chromosome instability, chromosome bridges at anaphases I and II were observed in variegated hybrid plants derived from *Nicotiana tabacum* and *N. plumbaginifolia* crossings (Gupta, 1968). Although the occurrence of laggards and non-oriented chromosomes along with precocious chromosome segregation observed in 'Variegada' somatic mutant grapevine had not affected the percentage of normal tetrads, these might have contributed to a decrease in pollen fertility. This mismatch between a higher percentage of normal tetrads and a lower percentage value of pollen fertility has been described in some other plants with meiotic irregularities. Thus, as laggard chromosomes and precocious chromosome segregation in diploid accessions of *Musa spp.* (Adeleke *et al.*, 2004) and in *Mesosetum chasaea* (Silva *et al.*, 2012), or as laggard chromosomes and bridges in *Vicia rigidula* (Kaur and Singhal, 2010) and in *Trigonella pubescens* (Kaur *et al.*, 2013). It is likely that the high percentage of laggard chromosomes and of precocious chromosome segregation observed in 'Variegada', could have resulted of a precocious chiasma terminalization, while a high percentage of non-oriented chromosomes may have been due to a failure in an accurate attachment of the chromosome kinetochore to the spindle fibers. Laggard and non-oriented chromosomes could also have contributed to an uneven chromosomal distribution among some tetrad microspores which in turn may have affected the percentage of pollen fertility. It is interesting that the highest percentage of pollen mother cells with abnormalities and a high percentage of chromosome stickiness observed in the parental 'Rosada' (21.05%) by Pierozzi and Moura (2014) were not strong enough to affect neither the percentage of normal tetrads (99.65%) nor the pollen fertility (93.45%).

Leaf variegation in plants has been associated with the expression of nuclear recessive genes and plastid genes (Zhao *et al.*, 2020) which affect plastid function impairing the photosynthesis. In grapevine, Reisch and Watson (1984) were the first to propose the expression of a nuclear recessive gene *var* to account for the leaf variegation. Further studies in grapevines developed by Olson *et al.* (2022) demonstrated that the variegated phenotype was related to the expression of two nuclear recessive genes *Lvar1* and *Lvar2*. It is possible that the variegation observed in 'Variegada' could be related to the expression of a recessive gene. If so, a mutation that allowed the expression of recessive variegated gene might have taken place in the bud stage just before or during the beginning of bud burst, therefore causing the variegated areas with photosynthesis impairment in leaf and in berry white sectors. However, further studies at molecular level in 'Variegada' grapevine are needed to elucidate what really triggered the somatic mutation which resulted in variegation.

Spontaneous somatic mutations affecting fruit morphology or size are the most studied. Some mutations have brought about a more attractive quality of the fruits while other mutations may have resulted in a harsh, tough, or russeting fruit epidermis (skin) compromising appearance and acceptance in the fruit market. Russeting is considered a fruit surface disorder affecting different varieties such as apples, pears, citrus, and prunes among other (Winkle *et al.*, 2022). The russet trait on the fruit skin may also be triggered by

environmental conditions (Athoo *et al.*, 2020) or could be a result of a gene expression as MdMYB93 responsible for russetting in apples (Legay *et al.*, 2016) or PpRus in Asian pear (Ma *et al.*, 2019). The russetting on the berry skin of 'Concord' grapevine variety described by Goffinet and Pearson (1991) was induced by use of a fungicide. The brown-bronze russetting-like structure observed on the epidermis of the mature berries of 'Steck' is not related to any chemical or physical agents since it spontaneously appeared in all bunches on one cane of 'Rosada' vineyard and was described as a somatic spontaneous mutation by Sousa (1959). Besides, not related to any fungal disease that sometimes affect muscadine grapevine. The brown russetting-like reticulated structure of 'Steck' is very intriguing because it is also not associated to any relevant disturbance in meiosis or pollen sterility. A reticulated brown-bronze structure initially observed in noticeably young immature berries evolved into a plain brown-bronze russetting-like layer covering all the berry skin. It seems that the russetting-like structure that appeared on the berry epidermis may be an abnormal deposition of a ligo-suberified layer on the berry epidermis due to an abnormal expression of gene(s) related to suberin and/or lignin synthesis which is normally expressed on the stem of grapevine canes but not on the berry skin. Interestingly, the berry pulp (flesh) of 'Steck' though not affected, displayed the characteristic light green color of 'Niagara' varieties. However, the berries possess an unexpected very sour flavor, which might be associated to the expression of grapevine *VvPH5* and *VvHP1* genes involved in vacuolar hyper-acidification as reported by Amato *et al.* (2019). Refined studies at molecular level will be necessary to verify if these two mutant traits in 'Steck' berries reflect an abnormal gene(s) expression(s).

'Niagara Rosada' seems to be a variety prone to somatic spontaneous mutations. Besides 'Variegada' and 'Steck' mutants, other mutations have been described (Sousa, 1959; Sousa, 1996; Pires *et al.*, 1988).

Plants exhibiting variegated leaves, or an unexpectedly undesirable fruit shape or skin texture have occasionally been observed in different crops. However, in opposition to these traits, growers have eliminated these plants because they are frequently less vigorous and/or unprofitable. Nevertheless, due to the visual attractiveness of 'Variegada' and 'Steck' as well as their robust growth, these may be used as ornamental grapevines in the same way as the 'Southern Home' hybrid grapevine with its deep-cut sinuses leaf shape resembling maple leaves (Mortensen *et al.*, 1994), the 'Roger's Red' grapevine with scarlet-red fall foliage and edible tasty berries (Dangl *et al.*, 2010) as well as *Vitis coignetiae*, the Crimson Glory grapevine (Lawo *et al.*, 2013).

Conclusions

The spontaneous somatic mutations, which generated 'Steck' and 'Variegada' varieties were not related to any changes in chromosome number or either to a structural modification or to a significant meiotic disturbance strong enough to compromise the pollen fertility. However, a lower percentage of pollen fertility observed in 'Variegada' when compared to 'Steck', may be attributed to a high percentage of laggard chromosomes, precocious chromosome segregation and non-oriented chromosomes.

Authors' Contributions

Both authors read and approved the final manuscript.

Ethical approval (for researches involving animals or humans)

Not applicable.

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Conflict of Interests

The authors declare that there are no conflicts of interest related to this article.

References

- Adeleke MTV, Pillay M, Okoli BE (2004). Relationships between meiotic irregularities and fertility in diploid and triploid *Musa L.* *Cytologia* 69(4):387-393. <https://doi.org/10.1508/cytologia.69.387>
- Amato A, Cavallini E, Walker AR, Pezzotti M, Bliok M, Quattrocchio F, ... Tornielli GB (2019). The MYB-drive MBW complex recruits a WRKY factor to enhance the expression of targets involved in vacuolar hyper-acidification and trafficking in grapevine. *The Plant Journal* 99(6):1220-1241. <https://doi.org/10.1111/tpj.14419>
- Angelotti-Mendonça J, Moura MF, Scarpere Filho JA, Vedoato BTF, Tecchio MA (2018). Rootstock on production and quality of 'Niagara Rosada' grapevine. *Revista Brasileira de Fruticultura* 40(4):e-023 <https://dx.doi.org/10.1590/0100-29452018023>
- Antcliff AJ, Webster WJ (1962). Bruce's sport - a mutant of the Sultana. *Australian Journal of Experimental Agriculture and Animal Husbandry* 2(5):97-100. <https://doi.org/10.1071/EA9620097>
- Ar-Rushdi AH (1957). The cytogenetics of variegation in a species hybrid in *Nicotiana*. *Genetics* 42(3):312-325. <https://doi.org/10.1093/genetics/42.3.312>
- Athoo TO, Winkler A, Knoche M (2020). Russeting in 'Apple' mango: triggers and mechanisms. *Plants* 9:898 <https://www.researchgate.net/publication/343006929>
- Chinnappa CC (1982). Cytology of some variegated forms of *Trillium grandiflorum* (Liliaceae). *Caryologia* 35(1):23-32 <https://doi.org/10.1080/00087114.1982.10796919>
- D'Cruz R, Rao GB (1977). Cytogenetic studies in two guava aneuploids. *Journal of Indian Botanical Society* 41(2):316-321.
- Dangl GS, Raiche R, Sim S, Yang J, Golino DA (2010). Genetic composition of the ornamental grape 'Roger's Red'. *American Journal of Enology and Viticulture* 61(2): 266-271. <http://iv.ucdavis.edu/files/29235.pdf>
- Ferrara G, Gallotta A, Pacucci C, Matarrese MAS, Mazzeo A, Giancaspro A, ... Colelli G (2017). The table grape 'Victoria' with a long-shaped berry: a potential mutation with attractive characteristics for consumers. *Journal of the Science of Food and Agriculture* 97(15):5398-5405. <https://doi.org/10.1002/jsfa.8429>
- Filler DM, Luby JJ, Ascher PD (1994). Incongruity in the interspecific crosses of *Vitis L.* Morphological abnormalities in the F2 progeny. *Euphytica* 78(3):227-237. <https://doi.org/10.1007/BF00027521>
- Goffinet M, Pearson R (1991). Anatomy of russeting induced in Concord grape berries by the fungicide chlorothalonil. *American Journal of Enology and Viticulture* 42(4):281-289.
- Gupta SB (1968). The unstable behavior of a chromosomal fragment of *Nicotiana plumbaginifolia* responsible for chlorophyll variegation in *N. tabacum*. *Genetics* 59(4):453-63. <https://doi.org/10.1093/genetics/59.4.453>
- Hedrick UP (1919). *Manual of American grape-growing*. MacMillan Co., New York.
- Kaur D, Singhal VK (2010). Chromosome number, meiosis and pollen fertility in *Vicia rigidula* Royle and *V. tenera* Grah. from cold desert regions of India. *Cytologia* 75(1):9-14. <https://doi.org/10.1508/cytologia.75.9>
- Kaur D, Kumar P, Singhal VK (2013). Chromosome counts and cytotoxicity in two species of *Trigonella L.* *Cytologia* 78(3):235-242. <https://doi.org/10.1508/cytologia.78.235>
- Knight TA (1808). On the variegation of plants. *Transactions of the Linnean Society of London* 9(1):268-271. <https://doi.org/10.1111/j.1096-3642.1818.tb00344.x>

- Kuksova VB, Piven NM, Gleba YY (1997). Somaclonal variation and *in vitro* induced mutagenesis in grapevine. Plant Cell, Tissue and Organ Culture 49(1):17-27. <https://doi.org/10.1023/A:1005830305206>
- Lawo NC, Lawo J-P, Plenck S, Schrank E, Forneck A (2013). *Vitis coignetiae* (Pulliat) shows partial resistance against leaf-feeding phylloxera and may serve to preserve abandoned vineyard habitats. Mitteilungen Klosterneuburg, Rebe und Wein, Obstbau und Früchterewertung 63(3):132-138.
- Legay S, Guerriero G, André C, Guignard C, Cocco E, Charton S, ... Hausman J-F (2016). MdMyb93 is a regulator of suberin deposition in russeted apple fruit skins. New Phytologist 212(4):977-991. <https://doi.org/10.1111/nph.14170>
- Ma C, Wang X, Yu M, Zheng X, Sun Z, Liu X, ... Wang C (2021). PpMYB36 encodes a MYB-type transcription factor that is involved in russet skin coloration in pear (*Pyrus pyrifolia*). Frontiers in Plant Science 12:776816. <https://doi.org/10.3389/fpls.2021.776816>
- Maia JDG (2012). Origem da videira Niágara [Origin of the grapevine Niágara]. In: Maia JDG, Camargo UA (Eds). O cultivo da videira Niágara no Brasil [Niágara vineyard cultivation in Brazil]. Embrapa, Brasília pp15-22.
- Malinowski E (1935). Studies on unstable characters in petunia. I. The extreme flower types of the unstable race with mosaic color patterns. Genetics 20(4):342-356 <https://doi.org/10.1093/genetics/20.4.342>
- Marcotrigiano M (1997) Chimeras and variegation: patterns of deceit. HortScience 32(5):773-784. <https://doi.org/10.21273/HORTSCI.32.5.773>
- Mortensen JA, Harris JW, Hopkins D, Andersen PC (1994). 'Southern Home': an interspecific hybrid grape with ornamental value. HortScience 29(11):1371-1372. <https://doi.org/10.21273/HORTSCI.29.11.1371>
- Olson J, Clark M (2021). Characterization of anatomical and physiological effects of variegation mutation on grapevine. HortScience 56(10):1251-1257. <https://doi.org/10.21273/HORTSCI15929-21>
- Olson J, Zou C, Karn A, Reisch B, Cadle-Davidson L, Su Q, Clark M (2022). Genetic analyses for leaf variegation in hybrid grape population (*Vitis spp.*) reveals two loci, *Lvar1* and *Lvar2*. HortScience 57(11):1416-1423. <https://doi.org/10.21273/HORTSCI16763-22>
- Pierozzi NI, Moura MF (2014). Cytological analyses in 'Niágara Branca' grape and in its somatic mutant 'Niágara Rosada'. Notulae Botanicae Horti Agrobotanici Cluj-Napoca 42(2):460-465. <https://doi.org/10.15835/nbha4229540>
- Pires EJP, Pommer CV, Passos IRS, Terra MM (1988). Mutante somático sem sementes em videira 'Niágara Rosada' [Seedless somatic mutant in the grapevine 'Niágara Rosada']. Bragantia 47(2):171-176. <https://doi.org/10.1590/S0006-87051988000200003>
- Pires EJP, Sawazaki HE, Terra MM, Botelho RV, Conagim A, Nogueira NAM (2003). Redimeire: A natural mutation of cv. Italia *Vitis* in Brazil. VITIS-GEILWEILERHOF 42(1):55-56. <https://doi.org/10.5073/vitis.2003.42.55-56>
- Raman VS, Rangasamy SRS, Manimekalai G (1971). Triploidy and seedlessness in guava (*Psidium guajava* L.). Cytologia 36(3):392-399. <https://doi.org/10.1508/cytologia.36.392>
- Reisch, BI, Watson JP (1984). Inheritance of leaf variegation in *Vitis* species. The Journal of Heredity 75(5):417-418. <https://doi.org/10.1093/oxfordjournals.jhered.a109973>
- Rick CM, Barton DW (1954). Cytological and genetical identification of the primary trisomics of the tomato. Genetics 39(5):640-666. <https://doi.org/10.1093/genetics/39.5.640>
- Sharma RL, Mukherjee SK (1972). Morphological descriptions of some induced systematic mutants of grapes (*Vitis vinifera* L.). Vitis 11(3): 177-188. <https://doi.org/10.5073/vitis.1972.11.177-188>
- Sharma RL, Mukherjee SK (1973). Some anatomical features of radiation-induced grape variegata. Vitis 12(3):189-191. <https://doi.org/10.5073/vitis.1973.12.189-191>
- Silva LAC, Pagliarini MS, Santos AS, Silva N, Souza VF (2012). Chromosome number, microsporogenesis, microgametogenesis and pollen viability in the Brazilian native grass *Mesosetum chaseae* (Poaceae). Genetics and Molecular Research 11(4):4100-4109. <https://doi.org/10.4238/2012.September.12.1>
- Sousa JSI (1959). Mutações somáticas na videira Niágara. [Somatic mutations in the Niágara grapevine] Bragantia 18(unique number):387-415. <https://doi.org/10.1590/S0006-87051959000100027>
- Sousa JSI (1996). Uvas para o Brasil [Grapes to Brazil]. FEALQ, Piracicaba, pp 791.
- Staudt G, Kassrawi M (1972). Die meiosis von di- und tetraploidem *Vitis vinifera* 'Riesling' [The meiosis in di and tetraploids *Vitis vinifera* 'Riesling']. Vitis 11(2):89-98. <https://doi.org/10.5073/vitis.1972.11.89-98>

- Tofanelli MBD, Botelho RV, Pires EJP, Vilela LAF, Ribeiro DO (2011). Phenology of “Niagara Rosada” grapevines grafted on different rootstocks grown on Cerrado (Brazilian savanna) of Goiás State, Brazil. African Journal of Biotechnology 10(17):3387-3392 <https://doi.org/10.5897/AJB09.1950>
- Vezzulli S, Leonardelli L, Malossini U, Stefanini M, Velasco R, Moser C (2012). Pinot Blanc and Pinot Gris arose as independent somatic mutations of Pinot Noir. Journal of Experiment Botany 63(18):6359-6369. <https://doi.org/10.1093/jxb/ers290>
- Walker AR, Lee E, Robinson SP (2006). Two new grape cultivars, bud sports of Cabernet Sauvignon bearing pale-coloured berries, are the result of deletion of two regulatory genes of the berry colour locus. Plant Molecular Biology 62(4-5):623-635. <https://doi.org/10.1007/s11103-006-9043-9>
- Winkler A; Athoo T; Knoche M (2022). Russeting of fruits: etiology and management. Horticulturae 8(3):231 <https://doi.org/10.3390/horticulturae8030231>
- Zhao M-H, Li X, Zhang X-X, Zhang H, Zhao X-Y (2020). Mutation mechanism of leaf color in plants: A review. Forests 11(8):851 <https://doi.org/10.3390/f11080851>



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