Fruit color and post-harvest shelf life in tomato affected by the *og^c*, *nor^A*, and *rin* alleles

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Abstract – The objective of this work was to evaluate the effects of the mutant alleles *alcobaça* (*nor^A*), *ripening inhibitor* (*rin*), and *old gold crimson* (*og^c*), in heterozygosity or homozygosity, on the expression of color and on the postharvest quality of fruit of experimental tomato hybrids. Fourteen hybrids with contrasting genotypic constitutions in the *nor^A*, *rin*, and *og^c* loci were evaluated in a randomized complete block design with four replicates. The following fruit postharvest quality traits were evaluated: firmness in the breaker stage, color, and soluble solids content. The *rin$^+$/rin* and *nor$^+$/nor^A* genotypes increased firmness of tomato fruit at harvest (breaker stage). The *rin$^+$/rin* genotypes displayed the worst internal fruit color. There was a positive effect of *og^{c+}/og^c* in improving the internal color of *rin$^+$/rin* and *nor$^+$/nor^A* fruit, making the color similar to that of the normal genotypes. The combination of the *og^c/og^c rin$^+$/rin nor$^+$/nor^A* genes is effective to improve tomato fruit firmness, besides maintaining or improving internal color.

Index terms: *Solanum lycopersicum*, *alcobaça*, *nonripening*, *old gold crimson*, plant breeding, *ripening inhibitor.*

Coloração e conservação pós-colheita de frutos de tomateiro influenciadas pelos alelos *og^c*, *nor^A* e *rin*

Resumo – O objetivo deste trabalho foi avaliar os efeitos promovidos pelos alelos mutantes *alcobaça* (*nor^A*), *ripening inhibitor* (*rin*) e *old gold crimson* (*og^c*), em heterozigose ou homozigose, na expressão da coloração e da conservação pós-colheita de frutos de híbridos experimentais de tomateiro. Quatorze híbridos com contituições genotípicas contrastantes entre si nos locos *nor^A*, *rin* e *og^c* foram avaliados em delineamento em blocos ao acaso, com quatro repetições. As seguintes características de qualidade pós-colheita dos frutos foram avaliadas: firmeza no estádio de amadurecimento incipiente, coloração e teor de sólidos solúveis. Os genótipos *rin$^+$/rin* e *nor$^+$/nor^A* condicionaram a maior firmeza dos frutos de tomate no ponto de colheita (estádio de amadurecimento incipiente). Os genótipos *rin$^+$/rin* apresentaram as piores colorações internas de frutos. Houve efeito positivo de *og^{c+}/og^c* na melhoria da coloração interna dos frutos *rin$^+$/rin* e *nor$^+$/nor^A*, que tornou a coloração semelhante à dos genótipos normais. A combinação dos genes *og^c/og^c rin$^+$/rin nor$^+$/nor^A* é eficiente para melhorar a firmeza dos frutos de tomate, além de manter ou melhorar a coloração interna.

Termos para indexação: *Solanum lycopersicum*, *alcobaça*, *nonripening*, *old gold crimson*, melhoramento genético, *ripening inhibitor.*

Introduction

Tomato (*Solanum lycopersicum* L.) is one of the most highly consumed vegetable crops in the world. Brazil ranks eighth in the worldwide ranking for tomato production (FAO, 2013). Tomato fruit is highly perishable, with losses of up to 21% after harvest (Rinaldi et al., 2011); it has high-water content, about 90 to 95%, which makes it fragile (Rocha et al., 2009). In tomato breeding programs, the main studied aspects are yield increase, pest and disease resistance, and the improvement of fruit quality. One of the factors associated with this last aspect is the greater natural conservation of the fruit postharvest, and this conservation may be achieved by the production of F1-hybrid fruit that is firmer and has better color (Andrade Júnior et al, 2001).

An alternative to extend the shelf life of tomato fruit has been to affect the natural maturation process of *Solanum lycopersicum* lines by the incorporation

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of mutant genes, such as *rin* (*ripening inhibitor*), *nor* (*nonripening*), *Nr* (*never ripe*), and *nor^{A}* (*alcobaça*) (Barry & Giovannoni, 2006). These genes block and retard the maturation process, which lends a longer life to fruit. Currently, most post-harvest long-life tomato hybrids are heterozygous for the genes *rin*, *nor*, or *nor^{A}*, which may lead to reduced quality because of the pleiotropic effects on other metabolic pathways involved in promoting fruit color, flavor, aroma, and texture (Kovács et al., 2009). However, alleles such as *high pigment (hp)* and *old gold crimson (og^{c})* increase lycopene production in the fruit, which improves color, and they can be used simultaneously with mutant maturation alleles (Andrade Júnior et al., 2005).

Araújo et al. (2002) have shown that the *alcobaça* allele in homozygosity (*nor^{A}/nor^{A}*) prolongs the post-harvest shelf life by the reduction of weight loss and the increase of firmness, and reduces the lycopene and beta-carotene concentrations, besides increasing the Brix/acidity ratio, mainly in association with the genotypes *hp/hp* and *hp^{+}/hp* or with *og^{c}/og^{c}*; however, its external color limits its commercial use. Cá et al. (2006) in the evaluation of the viability of simultaneous use of mutant genes, for maturity and for color in tomato hybrids, in the loci *nor^{A}*, *rin*, *og^{c}*, or *hp*, found that these genotypes exhibited better post-harvest shelf life than the normal genotypes, and concluded that the genotypic constitutions *nor^{+}/nor^{A} og^{c+}/og^{c}*, together with *nor^{+}/nor^{A} og^{c+}/og^{c} hp^{+}/hp*, were considered most promising, both for improving post-harvest conservation and for maintaining or improving the internal color of tomato fruit.

Faria et al. (2006) showed that the use of the mutants *og^{c}* or *hp*, in heterozygosity, improves the color and increases the concentrations of beta-carotene and lycopene of fruit in *rin^{+}/rin* and *rin^{+}/rin nor^{+}/nor^{A}* long shelf life hybrids. Andrade et al. (2015) concluded that the best combination of *high beta-carotene* (*B*) in heterozygosity for the hue angle and chroma depends on the mutant gene *og^{c}* in heterozygosity; they also showed that the *og^{c}* homozygous genotypes in the absence of *high beta-carotene* had a significant shift in the red direction in the placenta and in the columella, which confirms their effect on improving the internal color of fruit.

The objective of this work was to evaluate the effects of the mutant alleles *alcobaça* (*nor^{A}*), *ripening inhibitor* (*rin*), and *old gold crimson* (*og^{c}*), in heterozygosity or homozygosity, on the expression of color and on the postharvest quality of fruit of experimental tomato hybrids.

Materials and Methods

The study was carried out in a greenhouse and in the field, at the experimental station of HortiAgro Sementes S.A., in the Palmital farm (21°14'16"S, 45°08'00"W, at 920 m altitude), Ijaci, Minas Gerais state, Brazil, as well as in the Departamento de Agricultura, of Universidade Federal de Lavras (Ufla), and in the laboratory of vegetable crop post-harvest, of Departamento de Ciências dos Alimentos of Ufla.

The experiment in the greenhouse was carried out in a randomized complete block design, with 14 treatments, and four replicates. Homozygous lines for the genes *rin*, *nor^{A}*, and *og^{c}*, from the germplasm bank of HortiAgro Sementes S.A., were used as parent lines for different combinations in the loci *rin*, *nor^{A}*, and *og^{c}*, and the following hybrids were obtained: TEX-432=F1(TOM-667 x TOM-756), TEX-433=F1 (TOM-591 x TOM-756), TEX-434=F1(TOM-714 x TOM-756), TEX-435=F1(TOM-756 x TOM-761), TEX-436=F1(TOM-713 x TOM-761), TEX-437=F1 (TOM-591 x TOM-761), TEX-438=F1(TOM-756 x TOM-762), TEX-439=F1(TOM-713 x TOM-762), TEX-440=F1(TOM-591 x TOM-762), TEX-441=F1 (TOM-763 x TOM-TOM-713), TEX-442=F1(TOM-757 x TOM-713), and TEX-443=F1(TOM-758 x TOM-713). These hybrids, all with determinate growth habit, together with two commercial hybrids as controls ('Giselle' and 'Colono', also of determinate growth habit), constituted 14 treatments that were placed in five different categories, in relation to their genotypic constitutions in the loci *rin, nor^{A}*, and *og^{c}* (Table 1).

The plots consisted of a single row of 10 plants. The 14 genotypes were sown on 04/27/2015, and seedlings were produced in 128-cell expanded polystyrene trays that contained the commercial substrate Topstrato. Plants were thinned to one seedling per cell, at 15 days after sowing. Seedlings were transplanted in the field at 30 days after sowing. The experiment was conducted in the field according to the commercial growing recommendations for tomato under field conditions (Alvarenga, 2013), with a 0.5 m spacing between plants and 1.5 m between rows. The plants were tied on a half-length stake and the experiment was irrigated by drip.

The plants were pruned up to the height of the first flowering shoot, and from that point on, they were not pruned. Fertilization at planting and topdressing, and plant health control treatments were also performed, according to the specific recommendations for the tomato cropping.

The soluble solids content (°Brix) was evaluated through direct reading in a refractometer. Post-harvest conservation of fruit was evaluated (N m^{-2}) by fruit firmness collected at the beginning of the maturity stage, which is characterized by change of the fruit green color to the appearance of slightly red spots in the region of the stem scar – "breaker stage") using the applanation technique described by Calbo & Nery (1995).

The same samples harvested at the beginning of the maturity stage for the firmness evaluation were used for color evaluation. Fruit were stored at 15°C and 60% relative humidity until reaching full maturity. Color readings were made at 4 parts of the fruit – epidermis, pericarp, placenta, and columella – with the Minolta CR-400 colorimeter in the CIE mode L*, a*, and b*, in which L* (lightness) is the coordinate of brightness (z axis), which ranges from -100 (black) to +100 (white), in which the higher values indicate brighter colors; a* is the hue coordinate (x axis), which ranges from +60 (red) to -60 (green); b* is the hue coordinate (y axis), which ranges from +60 (yellow) to -60 (blue). From the a* and b* values, the hue angle [defined as arc tg (b/a)] and chroma or saturation [the square root of (a^2+b^2)] were obtained. The hue angle is considered from the 'a' axis and is expressed in degrees: 0° is defined as +a (red); 90°, as +b (yellow); 180° as -a (green), and 270° as -b (blue). Mature tomato fruit in general range from 0° (red) to 90° (yellow); the nearer the values are to zero (0°), the redder the fruit; and the nearer to 90°, the yellower the fruit (Konica Minolta, 2014). For saturation or chroma, the values range from 0 to 60. Values equal to zero correspond to the origin center of the coordinates; values near zero indicate less saturated colors, and the value of 60 indicates the maximum saturation (Konica Minolta, 2014).

Data were subjected to the analysis of variance, and means were compared by the Tukey test's, at 5% probability, by the SAS statistical application (SAS Institute, Inc., Cary, NC, USA). Experimental precision was checked by the estimate of selection accuracy (Resende & Duarte, 2007). Contrasts of interest were also calculated for comparisons between genotypes with different genotypic constitutions in the loci *rin*, og^c, and nor^A.

Table 1. Description of the treatments.

Treatment	Characteristics
Giselle	Normal *(rin^+/rin^+ nor^+/nor^+ og^{c+}/og^{c+})*
Colono	*rin* heterozygote *(rin^+/rin)*
TEX-432 (TOM-667 x TOM-756)	*rin* heterozygote *(rin^+/rin)*
TEX-433 (TOM-591 x TOM-756)	nor^A heterozygote; og^c heterozygote *(nor^+/nor^A ogc^+/og^c)*
TEX-434 (TOM-714 x TOM-756)	Normal *(rin^+/rin^+ nor^+/nor^+ og^{c+}/og^{c+})*
TEX-435 (TOM-756 x TOM-761)	*rin* heterozygote; og^c heterozygote *(rin^+/rin ogc^+/og^c)*
TEX-436 (TOM-713 x TOM-761)	*rin* heterozygote; og^c heterozygote *(rin^+/rin ogc^+/og^c)*
TEX-437 (TOM-591 x TOM-761)	nor^A heterozygote; *rin* heterozygote; og^c homozygote *(nor^+/nor^A rin^+/rin og^c/og^c)*
TEX-438 (TOM-756 x TOM-762)	*rin* heterozygote; og^c heterozygote *(rin^+/rin ogc^+/og^c)*
TEX-439 (TOM-713 x TOM-762)	*rin* heterozygote; og^c heterozygote *(rin^+/rin og^{c+}/og^c)*
TEX-440 (TOM-591 x TOM-762)	nor^A heterozygote; *rin* heterozygote; og^c homozygote *(nor^+/nor^A rin^+/rin og^c/og^c)*
TEX-441 (TOM-763 x TOM-713)	Normal *(rin^+/rin^+ nor^+/nor^+ og^{c+}/og^{c+})*
TEX-442 (TOM-757 x TOM-713)	Normal *(rin^+/rin^+ nor^+/nor^+ og^{c+}/og^{c+})*
TEX-443 (TOM-758 x TOM-713)	Normal *(rin^+/rin^+ nor^+/nor^+ og^{c+}/og^{c+})*

Results and Discussion

In the evaluated fruit quality characteristics – firmness, soluble solids content, and fruit color –, significant differences were observed between treatments, and all the quality traits showed accuracy higher than 70% (Table 2). Experimental precision was, therefore, considered high, according to Resende & Duarte (2007). The content of soluble solids, in the evaluated genotypes, varied from 3.72 °Brix, for 'Colono', to 4.72 °Brix for TEX-435 and TEX-441, respectively (Table 2). Only the differences between these extreme values were significant. The other genotypes, with intermediate °Brix values, did not differ from either of the two extreme values; this indicates that all the hybrids used were satisfactory for soluble solids content, and similar to the controls

'Giselle' or 'Colono'. Fruit firmness was affected by both the genotypic constitution in the loci (*rin*, *nor*^{A}, and og)^{c} and the genetic base (Table 2). The normal genotypes proved to be less firm at the beginning of the maturity stage than the heterozygous genotypes *rin* (C1, Table 3), *og*^{c} and *nor*^{A} (C3), and the genotypes *og*^{c}*/og*^{c} *rin*$^{+}$*/rin nor*$^{+}$*/nor*^{A} (C4); besides, they did not differ significantly from the genotypes *og*$^{c+}$*/og*^{c} *rin*$^{+}$*/rin* (C2). These results confirmed reports on *rin*$^{+}$*/rin* and *nor*$^{+}$*/nor*^{A} as responsible for the shelf life increase of the fruit (Araújo et al., 2002; Faria et al., 2003; Santos Júnior et al., 2005; Cá et al., 2006). These reports also showed that the genetic base also plays an important role in fruit firmness, when considering, for instance, the amplitude of the firmness values found among the normal genotypes 'Giselle', TEX-434, TEX-441, TEX-442, and TEX-443, among the heterozygous genotypes *rin* ('Colono', TEX-432), and among the genotypes *og*$^{c+}$*/og*^{c} *rin*$^{+}$*/rin* (TEX-435, TEX-436, TEX-438, TEX-439). The *og*^{c} allele in heterozygosity seems not to have a direct effect on firmness, since the heterozygous hybrids *og*^{c} and *rin* were less firm than the heterozygous genotypes *rin* alone (C5, Table 3). Heterozygous genotypes *og*^{c} and *nor*^{A} were firmer than the heterozygous *rin* (C6). However, simultaneous heterozygosity in the loci *rin* and *nor*^{A} seems to be the most effective combination for increasing firmness in relation to the presence of only one of these heterozygotes, when the magnitude and significance of the contrasts C7, C8, and C9 and of the negative signal (though nonsignificant) of the C10 contrast are considered.

Among the experimental hybrids, TEX-440 had the firmest fruit, and the fact that it has the genotypic constitution *og*^{c}*/og*^{c} *rin*$^{+}$*/rin nor*$^{+}$*/nor*^{A} confirms the effectiveness of obtaining firm hybrids through the homozygous genotype *og*^{c} and heterozygous genotypes for both *rin* and *nor*^{A}.

As to fruit color, differences were observed among the treatments for all the evaluated characteristics (epidermis, pericarp, placenta, and columella) for both hue angle and chroma values (Table 4). The hue angle was affected both by the presence of the mutants *rin, nor*^{A}, and/or *og*^{c} and by the genotypic background.

Table 2. Mean values of firmness in the breaker stage and of soluble solids in tomato (*Solanum lycopersicum*) fruit[(1)].

Treatment	Firmness (10^4 N m^{-2})	Soluble solids (°Brix)
Giselle	2.40bcd	4.35ab
Colono	3.50a	3.72b
TEX-432 (TOM-667 x TOM-756)	2.02d	4.01ab
TEX-433 (TOM-591 x TOM-756)	3.06abcd	3.98ab
TEX-434 (TOM-714 x TOM-756)	2.03d	3.97ab
TEX-435 (TOM-756 x TOM-761)	2.70abcd	4.72a
TEX-436 (TOM-713 x TOM-761)	2.12d	4.44ab
TEX-437 (TOM-591 x TOM-761)	3.31abc	4.15ab
TEX-438 (TOM-756 x TOM-762)	2.33bcd	4.29ab
TEX-439 (TOM-713 x TOM-762)	2.35bcd	4.42ab
TEX-440 (TOM-591 x TOM-762)	3.44ab	4.13ab
TEX-441 (TOM-763 x TOM-713)	2.21cd	4.72a
TEX-442 (TOM-757 x TOM-713)	2.52abcd	4.62ab
TEX-443 (TOM-758 x TOM-713)	2.30bcd	4.25ab
Accuracy (%)	92.97	76.60

[(1)]Mean values followed by equal letters, in the columns, do not differ by the Tukey's test, at 5% probability.

Table 3. Estimates of contrasts of interest for firmness ($2x10^4$ N m^{-2}) in tomato (*Solanum lycopersicum*) fruit.

Contrast	Description	Firmness estimate (N m^{-2})
C1	Normal genotypes vs heterozygous genotypes *rin*	-0.47*
C2	Normal genotypes vs heterozygous genotypes *og*^{c} and *rin*	-0.08^{ns}
C3	Normal genotypes vs heterozygous genotype *og*^{c} and *nor*^{A}	-0.77**
C4	Normal genotypes vs homozygous genotypes *og*^{c} and heterozygous *rin* and *nor*^{A}	-1.08**
C5	Heterozygous genotypes *rin* vs heterozygous genotypes *og*^{c} and *rin*	0.39*
C6	Heterozygous genotypes *rin* vs heterozygous genotype *og*^{c} and *nor*^{A}	-0.30^{ns}
C7	Heterozygous genotypes *rin* vs homozygous genotypes *og*^{c} and heterozygous *rin* and *nor*^{A}	-0.61**
C8	Heterozygous genotypes *og*^{c} and *rin* vs heterozygous genotype *og*^{c} and *nor*^{A}	-0.68**
C9	Heterozygous genotypes *og*^{c} and *rin* vs homozygous genotypes *og*^{c} and heterozygous genotypes *rin* and *nor*^{A}	-1.00**
C10	Heterozygous genotype *og*^{c} and *nor*^{A} vs homozygous genotypes *og*^{c} and heterozygous genotypes *rin* and *nor*^{A}	-0.31^{ns}

og^{c}, presence of the *old gold crimson* allele; *rin*, *ripening inhibitor* allele; *nor*^{A}, *alcobaça* allele. ** and *Significant by the Tukey's test, at 1 and 5%, respectively. ^{ns}Nonsignificant. Normal genotypes: *rin*$^{+}$*/rin*$^{+}$ *nor*$^{+}$*/nor*$^{+}$ *og*$^{c+}$*/og*$^{c+}$.

Evidence of the effect of the genotypic background can be observed in the difference between the normal genotypes – 'Giselle' had a significantly greater hue angle (therefore, lower tendency to red) – in the placenta than the other normal hybrids: TEX-434, TEX-441, TEX-442, and TEX-443.

Differences were detected in the external color (epidermis) and internal color (pericarp, placenta, columella) of the fruit due to the presence of the loci *rin, nor*, and og^c (Table 5). The heterozygosity in the locus *rin* increased the hue angle in relation to the normal genotypes (C1), in both the external and internal parts of the fruit, which shows the detrimental effect of rin^+/rin on color. The detrimental effects of rin^+/rin on internal color of the fruit were however reversed by the presence of og^c in heterozygosity (C2 and C5).

In the present study, it was not possible to measure the effect of the genotype nor^+/nor^4, separately, on the internal and external colors, due to the absence of a heterozygous genotype nor^4 in the absence of og^c. However, it is clear that this detrimental effect, if it exists, is counterbalanced by the presence of og^c

Table 4. Mean values of hue angle (in degrees) for epidermis, pericarp, placenta, and columella of tomato (*Solanum lycopersicum*) fruit[(1)].

Treatment	Epidermis	Pericarp	Placenta	Columella
Giselle	38.77ab	38.90abcd	58.63a	39.11ab
Colono	38.34abcd	39.76ab	59.08a	34.81bc
TEX-432 (TOM-667 x TOM-756)	38.58abc	41.02a	55.21ab	34.43bc
TEX-433 (TOM-591 x TOM-756)	33.84e	35.10d	52.99bc	39.09ab
TEX-434 (TOM-714 x TOM-756)	35.32de	36.79bcd	51.31bc	34.73bc
TEX-435 (TOM-756 x TOM-761)	37.89abcd	36.78bcd	50.05c	35.03bc
TEX-436 (TOM-713 x TOM-761)	38.92a	38.65abcd	55.36ab	40.66a
TEX-437 (TOM-591 x TOM-761)	35.81bcde	36.06bcd	49.27c	39.13ab
TEX-438 (TOM-756 x TOM-762)	38.47abc	38.17abcd	52.38bc	32.66c
TEX-439 (TOM-713 x TOM-762)	38.33abc	39.73abc	55.57ab	37.87abc
TEX-440 (TOM-591 x TOM-762)	37.20abcd	37.13abcd	48.89c	37.99abc
TEX-441 (TOM-763 x TOM-713)	35.53cde	35.93cd	51.06bc	36.64abc
TEX-442 (TOM-757 x TOM-713)	36.85abcde	37.55abcd	51.91bc	40.62a
TEX-443 (TOM-758 x TOM-713)	36.68abcde	37.82abcd	52.71bc	38.52ab
Accuracy (%)	89.53	85.03	95.41	85.47

[(1)]Mean values followed by equal letters, in the columns, do not differ by the Tukey's test, at 5% probability.

Table 5. Estimates of contrasts of interest for hue angle of the epidermis (EP), pericarp (PE), placenta (PL), and columella (CO) of tomato (*Solanum lycopersicum*) fruit.

Contrast	Description	Estimates			
		EP	PE	PL	CO
C1	Normal genotypes vs heterozygous genotypes *rin*	-1.83**	-2.99**	-4.02**	3.30**
C2	Normal genotypes vs heterozygous genotypes og^c and *rin*	-1.77**	-0.93^{ns}	-0.22^{ns}	1.37^{ns}
C3	Normal genotypes vs heterozygous genotypes og^c and nor^4	2.79**	2.30*	0.13^{ns}	-1.16^{ns}
C4	Normal genotypes vs homozygous genotypes og^c and heterozygous genotypes *rin* and nor^4	0.12^{ns}	0.80^{ns}	4.04**	-0.63^{ns}
C5	Heterozygous genotypes *rin* vs heterozygous genotypes og^c and *rin*	0.06^{ns}	2.05**	3.80**	-1.93*
C6	Heterozygous genotypes *rin* vs heterozygous genotypes og^c and nor^4	4.62**	5.29**	4.15**	-4.47**
C7	Heterozygous genotypes (HG) *rin* vs homozygous genotypes og^c and HG *rin* and nor^4	1.95**	3.79**	8.07**	-3.94**
C8	Heterozygous genotypes og^c and *rin* vs heterozygous genotypes og^c and nor^4	4.56**	3.23**	0.35^{ns}	-2.54^{ns}
C9	Heterozygous genotypes (HG) og^c and *rin* vs homozygous genotypes og^c and HG *rin* and nor^4	1.90**	1.74*	4.26**	-2.00*
C10	Heterozygous genotypes (HG) og^c and nor^4 vs homozygous genotypes og^c and HG *rin* and nor^4	-2.67**	-1.50^{ns}	3.92**	0.53^{ns}

og^c, presence of the *old gold crimson* allele; *rin*, *ripening inhibitor* allele; nor^4, *alcobaça* allele. ** and *Significant by the Tukey's test at 1 and 5% probability, respectively. ^{ns}Nonsignificant. Normal genotypes: rin^+/rin^+ nor^+/nor^+ og^{c+}/og^{c+}.

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in heterozygosity because the combination og^{c+}/og^{c} nor^{+}/nor^{A} improved the epicarp and placenta colors in comparison to the normal genotypes (C3, Table 5). The possible detrimental effect of nor^{+}/nor^{A}, if it exists, appears to be less than that brought about by rin^{+}/rin because the beneficial effects of ogc^{+}/og^{c} are more accentuated in the first case (C6) than in the second one (C5).

The simultaneous presence of heterozygosity in *rin* and nor^{A} besides homozygosity in og^{c}/og^{c} provided not only a red color comparable to that of normal genotypes in the epidermis, pericarp, and columella, but also improved it in the placenta (C4, Table 5). Andrade et al. (2015) found that homozygous genotypes og^{c} did not differ from the normal genotypes for the hue angles in the epidermis and pericarp, but had a significant shift in the red direction in the placenta and columella.

As to the genotype rin^{+}/rin alone, the combination og^{c}/og^{c} rin^{+}/rin nor^{+}/nor^{A} exhibited color closer to red in all the measured points (C7, Table 5), and the same happened in the combination og^{c+}/og^{c} rin^{+}/rin (C9). In relation to the genotype og^{c+}/og^{c} nor^{+}/nor^{A}, the genotypic combination og^{c}/og^{c} rin^{+}/rin nor^{+}/nor^{A} brought about the best color in the placenta and the worst color in the epidermis (C10). Araújo et al. (2002) reported that the mutants og^{c} and *hp*, with a genotypic background 'Floradade', isolated or in combination, in both homozygosity and in heterozygosity, brought about significant increases in internal and external colors.

From these results it can be inferred that the allele og^{c}, both in homozygosity and in heterozygosity, is able to counterbalance the negative effects of the heterozygotes rin^{+}/rin or nor^{+}/nor^{A} in fruit color, mainly in internal color. Similar effects on fruit color were found by Faria et al. (2003) and Cá et al. (2006). In fact, the three smallest chromaticity angles (nearest to red) among the studied genotypes were TEX-435 of the genotype og^{c+}/og^{c} rin^{+}/rin, and TEX-437 and TEX-440 of the genotype og^{c}/og^{c} rin^{+}/rin nor^{+}/nor^{A}.

The chroma saturation seems to have been less affected than the hue angle among the studied genotypes (Table 6), particularly in the epidermis, pericarp, and placenta, whereas the normal genotype control 'Giselle' had the lowest numerical chromas among the treatments, and the hybrid TEX-441, which is also of normal genotype, had some of the highest chromas at these points.

The most significant differences in the chromas occurred in the columella (Table 7). The genotypes rin^{+}/rin had significantly lower chromas than the normal genotypes (C1), but due to the magnitude of variation among the normal genotypes themselves, it is possible that this difference arises mainly from the effect of different genotypic backgrounds, and not from the allele *rin* only. Nevertheless, the effect of the allele og^{c} in heterozygosity is clear as for the increasing of the chroma of the hybrids rin^{+}/rin, both in the external and internal part of the fruit (C5). Genotypes og^{c+}/og^{c} nor^{+}/nor^{A} also showed greater internal chroma than the $rin^{+}/$

Table 6. Mean values of chroma for epidermis, pericarp, placenta, and columella of tomato (*Solanum lycopersicum*) fruit.

Treatment	Epidermis	Pericarp	Placenta	Columella
Giselle	28.23ab	27.39a	24.22ab	31.68abcd
Colono	26.72b	26.15a	21.87b	25.93d
TEX-432 (TOM-667 x TOM-756)	28.91ab	28.74a	23.51ab	32.77abc
TEX-433 (TOM-591 x TOM-756)	29.81ab	30.13a	26.08ab	32.19abc
TEX-434 (TOM-714 x TOM-756)	27.90ab	31.38a	27.40a	35.55a
TEX-435 (TOM-756 x TOM-761)	29.66ab	30.89a	25.46ab	32.99abc
TEX-436 (TOM-713 x TOM-761)	31.98a	29.76a	24.96ab	30.95abcd
TEX-437 (TOM-591 x TOM-761)	29.89ab	31.03a	23.86ab	29.54bcd
TEX-438 (TOM-756 x TOM-762)	28.26ab	28.98a	24.46ab	31.60abcd
TEX-439 (TOM-713 x TOM-762)	30.14ab	29.76a	23.79ab	32.08abc
TEX-440 (TOM-591 x TOM-762)	30.26ab	29.58a	25.15ab	29.10cd
TEX-441 (TOM-763 x TOM-713)	30.32ab	30.41a	27.61a	35.31ab
TEX-442 (TOM-757 x TOM-713)	28.72ab	29.12a	27.09a	30.48abcd
TEX-443 (TOM-758 x TOM-713)	28.13ab	27.20a	25.24ab	29.59bcd
Accuracy (%)	75.43	70.53	75.79	90.32

(1)Mean values followed by equal letters, in the columns, do not differ by theTukey's test, at 5% probability.

Table 7. Estimates of contrasts of interest for chroma of the epidermis (EP), pericarp (PE), placenta (PL), and columella (CO) of tomato (*Solanum lycopersicum*) fruit.

Contrast	Description	Estimates			
		EP	PE	PL	CO
C1	Normal genotypes vs heterozygous genotypes *rin*	0.84^{ns}	1.66^{ns}	3.62**	3.17**
C2	Normal genotypes vs heterozygous genotypes *og^{c}* and *rin*	-1.35^{ns}	-0.75^{ns}	1.65*	0.62^{ns}
C3	Normal genotypes vs genotypes *og^{c}/og^{c} nor^{+}/nor^{A}*	-1.15^{ns}	-1.03^{ns}	0.23^{ns}	0.33^{ns}
C4	Normal genotypes vs genotypes *og^{c}/og^{c} rin^{+}/rin nor^{+}/nor^{A}*	-1.42^{ns}	-1.20^{ns}	1.81*	3.20**
C5	Heterozygous genotypes *rin* vs heterozygous genotypes *og^{c}* and *rin*	-2.19*	-2.41*	-1.98*	-2.55*
C6	Heterozygous genotypes *rin* vs heterozygous genotype *og^{c}* and *nor^{A}*	-2.00^{ns}	-2.69*	-3.39**	-2.84*
C7	Heterozygous genotypes (HG) *rin* vs homozygous genotypes *og^{c}* and HG *rin* and *nor^{A}*	-2.26*	-2.86*	-1.82^{ns}	0.03^{ns}
C8	Heterozygous genotypes *og^{c}* and *rin* vs heterozygous genotype *og^{c}* and *nor^{A}*	0.20^{ns}	-0.28^{ns}	-1.42^{ns}	-0.29^{ns}
C9	Heterozygous genotypes (HG) *og^{c}* and *rin* vs homozygous genotypes *og^{c}* and HG *rin* and *nor^{A}*	-0.07^{ns}	-0.46^{ns}	0.16^{ns}	2.58*
C10	Heterozygous genotype (HG) *og^{c}* and *nor^{A}* vs homozygous genotypes *og^{c}* and HG *rin* and *nor^{A}*	-0.26^{ns}	-0.17^{ns}	1.58^{ns}	2.87^{ns}

og^{c}, presence of the *old gold crimson* allele; *rin*, *ripening inhibitor* allele; *nor^{A}*, *alcobaça* allele. ** and *Significant by the Tukey's test, at 1 and 5% probability, respectively. ^{ns}Nonsignificant. Normal genotypes: *rin^{+}/rin^{+} nor^{+}/nor^{+} og^{c+}/og^{c+}*.

rin (C6). Under the simultaneous presence of *rin^{+}/rin* and *nor^{+}/nor^{A}*, the allele *og^{c}* in homozygosity increased the chroma in the epidermis and in the pericarp of tomato fruit (C7).

Conclusions

1. The genotypic constitutions *rin^{+}/rin nor^{+}/nor^{A}* are responsible for providing greater firmness to tomato (*Solanum lycopersicum*) fruit at the time of harvest, and the allele *og^{c}* does not have a direct effect on firmness.

2. Heterozygosity in the locus *rin* has a detrimental effect on color of the external and internal parts of the fruit. The *og^{c}* allele is able to counterbalance the negative effects of the heterozygotes *rin^{+}/rin* or *nor^{+}/nor^{A}* on internal color.

3. The genotypic constitutions *og^{c}/og^{c} rin^{+}/rin nor^{+}/nor^{A}* or *og^{c+}/og^{c} nor^{+}/nor^{A}* are considered the most promising ones, improving the firmness and the internal or external color of tomato fruit.

4. The simultaneous presence of heterozygous alleles *rin* and *nor^{A}*, associated with *og^{c}*/og^{c}, is effective in developing tomato hybrids with better post-harvest conservation.

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