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POSTHARVEST SHELF LIFE OF TOMATO (*Lycopersicon esculentum Mill.*) MUTANATS (*nor* and *rin*) AND THEIR HYBRIDS

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Two tomato lines with normal maturation (NR-1 and NR-2) have been crossed with two mutant lines (NR-10 nor and NR-12 rin) with delayed maturation (shelf life). Determination of mutant genes has been done by χ^2 test on 100 fruits from F_2 generation. Fruits have been picked 65 days from antesis and kept for 60 days, when six evaluations have been done. Data have been collected every 10 days on parental lines and progeny F_1 and F_2 generation. Variance testing has been done on the basis of one- and two-factorial analysis and groups compared by contrasts. Fruits have been preserved in controlled conditions (in dark at 5^{o}C). Tomato genotypes with nor or rin gene had desirable traits (delayed ripening, long shelf life and firm fruits) for modern selection, so they should be included in programmes aiming to create commercial F_1 hybrids.

Key words: hybrid, mutant, nor, rin, tomato

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INTRODUCTION

Modern ways of tomato selection emphasize ticker pericarp as an imperative. This trait together with firmness of ectocarp and firmness of placenta enables longer shelf life and postharvest preservation. Mutant *nor* (non-ripening) and *rin* (ripening inhibitor) tomato lines have ticker pericarp besides the delayed ripening, which therefore means better firmness of fruits (HO, 2003).

Fruits with greater firmness can be selected by accumulating firmness traits (ZDRAVKOVIĆ *et al.,* 2010). Genotypes with "fruit firmness" gene cause long shelf life of mature tomato (ZDRAVKOVIĆ *et al*., 2003).

Heterozygous combination of *rin* and the mutants nor^A, increases fruit firmness and the effect of the rin^+/rin, nor^+/nor^A genotypes on fruit firmness was roughly the sum of the individual effects of each locus. The *rin* allele was more efficient than nor^A to keep fruit firmness (FARIA *et al.* 2006).

During shelf life, especially for fruits harvested in light green phase, synthesis of β-carotene is faster than lycopene synthesis (BRASHLYANOVA and PEVICHAROVA, 2009). Fruits harvested in light red phase had increased beta carotene (BRASHLYANOVA and GANEVA, 2009). Packaging also affects traits and quality of fruits since fruits turn red from pink due to the change of carotene (NASRIN *et al.,* 2008)

During postharvest period tomato fruits can change quality. Fruits are stored, packed and transported to the end consumers. Loss and changes of quality are important for consumers who use tomato fresh. Complex selection programmes enable commercial F_1 hybrids and quality during postharvest shelf life.

MATERIALS AND METHODS

Four parental lines (NR-1, NR-2, NR-10 (*nor*) and NR-12 (*rin*) were analysed genetically. Lines NR-1 and NR-2 with normal period of ripening and lines NR-10 (*nor*) and NR-12 (*rin*) with delayed ripening.

Hybridization of parental lines obtained F_1 generation, and self-pollination of F_1 hybrids F_2 generation. Parents, F_1 and F_2 generation have been compared in random block system with three replications. Fruits were picked 65 days after pollination and kept in controlled conditions for 60 days. Data regarding the number of decayed fruits were taken every 10 days.

Ten fruits per parent and F_1 generation and 20 per F_2 generation were taken. For testing the number of decayed fruits the proportion according to base line of units – fruits was calculated. The transformed data have been processed by one-way and two-way ANOVA. The transformation included:

$$X_{ijk} = 2\arcsin\sqrt{Xijk}$$

$$X_{ijk} = 2\arcsin\sqrt{Xijk \pm 1/2n}$$

Sign + is used in case of $X_{ijk} \approx 0$, sign – in case of $X_{ijk} \approx 1$ and n is the number on the basis of which is calculated proportion (10 or 20). The transformed data have been processed by two-way ANOVA.

For the data of trait – fruits storage analysis included 10 genotypes measured three times (usual ripening dynamics) and 6 genotypes measured six times (prolonged ripening). One-way ANOVA aimed to estimate the significance of factors and variance homogeneity was calculated:

$$\chi^2 = \frac{2.306 f \left(K \log Sp^2 - \sum_{i=1}^{k} \log Si^2\right)}{1 + \left[(K+1)/3Kf\right]}$$

$$Sp = \frac{\sum_{i=1}^{k} Si^2}{K},$$

with f representing the number of degrees of freedom for each variance and K representing the number of variances. If the calculated value exceeds the table value, the data are transformed according to log(x+1) formula, otherwise raw data are processed.

The data were processed by complex contrast comparison. Complex contrast with multiple degrees of freedom is a group of simple contrasts. It is usually defined as comparison among groups (s-number of groups):

$M=g_1$ with g_2 with g_3 with ... g_s

with g_i representing i-th group composed of m_i treatment.

The hypothesis of equality of group means was tested:

$$SS(M) = \frac{1}{rgs} \sum_{i=1}^{r} \frac{Gi^2}{m_i} - \frac{\left(\sum_{i=1}^{s} Gi^2\right)^2}{rgs \sum_{i=1}^{s} m_i}$$

with r representing the number of replications, g terms of measurements, $M=g_1$ with g_2 has (s-1)=2-1=1 degrees of freedom.

$$F = \frac{\frac{SS(M)}{(s-1)}}{MSa}$$

F-table value for $v_1=1$ and $v_2=60$ degrees of freedom is 4.00 and 7.08 for 0.05 and 0.01 levels of probability, respectively.

Two groups (s=2) of treatments, namely genotypes without prolonged shelf life (4) and mutant genotypes (6) have been considered:

- group g_1: Korona, R-83, Korona x R-83 F_1 and Korona x R-83 F_2, ($m_1=4$);
- group g_2: L-10, Korona x L-10 F_1, Korona x L-10 F_2, L-12, Korona x L-12 F_1 and Korona x L-12 F_2, ($m_2=6$).

If the calculated F value exceeds the table value, the means of the observed groups are significantly different.

RESULTS AND DISCUSSION

Determination of *nor* and *rin* genotypes were determined according to colour of fruits of researched genotypes (per 100 fruits from F_2 generation), where:

- *nor* (light green),
- *rin* (light yellow),
- Normal (red).

All F_1 hybrid combinations of *nor* and normal ripening genotypes, as well as *rin* and normal ripening, gave light red and red fruits, which proves earlier studies regarding recessive character of this trait.

In F_2 generation there was an expected segregation (3:1) *nor* and *rin* genotypes comparing to genotypes with normal ripening, which proves the recessive character of single gene (*nor* and *rin*), Table 1.

Table 1. χ^2 test of splitting of F_2 generation to normal (nor and rin) genotypes

Combination	Number of fruits: normal exp.	normal teor.	(nor – rin) exp.	(nor – rin) teor.	total	χ^2
NR-1 x NR-10	70	75	30	25	100	0.53
NR-1 x NR-12	77	75	23	25	100	0.21
NR-2 x NR-10	71	75	29	25	100	0.85
NR-2 x NR-12	81	75	29	25	100	1.92
total	299	300	118	100	400	0.43

r=1	0.05=3.84	r=4	0.05=9.49
r=1	0.01=6.63	r=4	0.01=13.28

Results of average number of decayed fruits during shelf life for researched genotypes (uniform and delayed ripening) are presented in Table 2.

Table 2. Average number of decayed fruits tested during shelf life

genotypes \ date	04.08.	14.08.	24.08.	03.09.	13.09.	23.09.
NR-1	10	3	7	-	-	-
NR-2	10	5	5	-	-	-
NR-1 x NR-2 F_1	10	5	5	-	-	-
NR-1 x NR-2 F_2	20	12	8	-	-	-
NR-10	10	-	-	-	-	1
NR-1 x NR-10 F_1	10	-	-	-	2	1
NR-1 x NR-10 F_2	20	-	-	3	1	5
NR-12	10	-	-	-	1	2
NR-1 x NR-12 F_1	10	-	-	1	2	4
NR-1 x NR-12 F_2	20	-	-	5	5	2

Fruits of genotypes with uniform ripening (parents, F_1 and F_2 hybrids) decayed after the third measurement term, 20 days after harvest. Fruits of parents NR-2 lasted the longest. For *nor* and *rin* genotypes the first fruits started to senescence in the forth term of measurement, 30 days after harvest. Obviously, genotypes with *nor* gene last longer than genotypes with *rin* gene (they senescence slower, to be more precise) while both mutants can be preserved longer comparing to genotypes with uniform ripening.

Fruits with *nor* gene construction preserved longer (about three months after harvest) comparing to *rin* (about two months after harvest). These results have been incorporated by TANKSLY *et al.* (1998).

Nor and *rin* genotypes with delayed ripening have late or absent synthesis of lycopine, in favour of synthesis of β-carotene (KUZEMENSKI, 2007).

Analysis of variance for all 10 genotypes (with uniform and delayed ripening) was significantly different in six terms of measurements. Differences among the researched genotypes (311.256**), among the terms of measurements (167.384**) and for interaction genotype x measurement term (33.836**) were significant (Table 3).

Table 3. Two-factor analysis for number of decayed fruits for 10 genotypes in three measurement terms

Source of variation	df	SS	MS	F-calculated	F-table	
					0.05	0.01
Among measurement terms	1	8.236	8.236	311.256**	4.08	7.31
Among genotypes	9	39.861	4.429	167.384**	2.12	2.80
Interaction genot x meas. term	9	8.058	0.895	33.836**	2.12	2.80
Error	40	1.058	0.026			
Total	59	57.212				

Group comparisons, mean value for the observed treatments of genotypes with uniform ripening (four) with nor and rin genotypes (six) clearly shows that average values of the observed groups of treatments were statistically different (F=821.45**). Obtained results (for genotypes with uniform ripening G_1=51.32 and for nor and rin genotypes G_2=22.81) proved that nor and rin genotypes senescence less since not a single fruit decayed during three terms of measurement comparing to genotypes with normal ripening. Comparisons refer to first three measurement treatments.

G_1=51.32
G_2=22.81
SS(M)=21.73

F=821.45** v_1=1 v_2=40 $F_{00.5}$=4.08 $F_{00.1}$=7.31

Results of two-factorial analysis of variance for the number of decayed fruits in postharvest period in *nor* and *rin* genotypes showed significant values (Table 4). Values among researched *nor* and *rin* genotypes (26.862**), measurement terms (12 357**) and interaction genotype x terms of measurements (2.068*) were significant at level of significance 0.05 (Table 4).

Table 4. Two-factor analysis of variance for number of decayed fruits in six generations in six measurements

Among measurement terms	df	SS	MS	F-calculated	F-table	
					0.05	0.01
Among measurement terms	4	9.504	2.376	26.862**	2.52	3.83
Among genotypes	5	5.465	1.093	12.357**	2.37	3.51
Interaction genot x meas. term	20	3.658	0.183	2.068*	1.81	2.20
Error	60	5.307	0.088			

CONCLUSION

χ^2 test proved that hypothesis of mono-gene and recessive character of *nor* and *rin* gene. In F_2 generation there was expected segregation 3:1 of genotypes with normal ripening comparing to *nor* and *rin* genotypes.

Shelf life of fruits and dynamics of change of parameters of quality of tomato fruits (B-carotene, total acids and dry matter) after harvest was different depending on variety and presence of inhibitor gene of ripening (*nor* and *rin*).

During shelf life of harvested fruits, significant longer shelf life has been noted for *nor* and *rin* genotypes comparing to genotypes with uniform ripening. *Nor* fruits had longer shelf life than *rin*, and some fruits of *nor*-genotype were shelved three months after harvest and *rin*-genotype about two months.

Obviously, tomato genotypes with *nor* or *rin* gene have desirable traits (delayed ripening, long shelf life, good firmness) which are prerogative of modern selection, so they should be included in selection programs aiming to create commercial F_1 hybrids. Creation of such hybrids would enable longer shelf life, successful transport to further destination with minimal losses of quantity and quality.

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DUŽINA ČUVANJA PLODOVA PARADAJZA (*Lycopersicon esculentum Mill.*) MUTANATA (*nor* i *rin*) I NJIHOVIH HIBRIDA U POSLE ŽETVENOM PERIODU

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Izvršena su ukrštanja dve linije paradajza sa normalnim periodom sazrevanja plodova (NR-1 i NR-2) i dve mutantne linije (NR-10 nor i NR-12 rin) kod kojih je period sazrevanja odložen (shelf life). Izvršena je determinacija mutantnih gena pomoću χ^2 testa na uzorku od 100 plodova F_2 generacije.
Plodovi su ubrani 65 dana od antezisa i čuvani u periodu od 60 dana, kada je izvršeno šest evaluacija. Podaci su uzimani u intervalima na svakih 10 dana na roditeljskim linijama i potomstvu F_1 i F_2 generacije. Testiranje varijanse izvršeno je na osnovu jednofaktorijalne i dvofaktorijalne analize na osnovu kojih su izvršena grupna poređenja korišćenjem složenih kontrasta. Plodovi su čuvani u kontrolisanim uslovima (u mraku, na temperaturi od 5^{o}C).

Možemo reći da genotipovi paradajza sa nor ili rin genom poseduju poželjne osobine (odloženo sazrevanje, mogućnost dugog čuvanja, kao i veliku čvrstinu ploda) koje zahtevaju savremeni pravci selekcije, te ih s toga treba uključiti u selekcione programe koje za cilj imaju stvaranje komercijalnih F_1 hibrida.

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