

# GENETIC DIVERGENCE OF STRAWBERRY CULTIVARS UNDER DIFFERENT MANagements

## DIVERGÊNCIA GENÉTICA DE CULTIVARES DE MORANGO EM DIFERENTES MANEJOS

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**ABSTRACT:** Developing strawberry cultivars that can be grown on a large scale, it is necessary to gather desirable characteristics such as: tolerance to *Tetranychus urticae*, high fruit yield and wide adaptability to several cropping managements. Therefore, our objective was to study the genetic diversity among 13 strawberry cultivars under different managements and to recommend promising crosses to obtain segregating populations with high fruit yield and *T. urticae* tolerance. Trial was performed under field conditions at the Centro Regional de Desenvolvimento Rural Centro Serrano of the Instituto Capixaba for Technical Assistance and Rural Extension (Incaper), Domingos Martins-ES. We evaluated strawberry cultivars Albion, Aleluia, Aromas, Camarosa, Camino Real, Campinas, Diamante, Dover, Festival, Seascap, Toyonoka, Tudla, and Ventana, cultivated in three cropping managements: open field, low tunnel and high tunnel. Experimental design was randomized complete blocks with three replications. Variables evaluated were: number of two-spotted spider mite/cm<sup>2</sup> on the leaf (NTSSM), total number of fruits (TNF), number of commercial fruits (NCF) and fruit yield (YIE, t/ha). We applied the generalized Mahalanobis distance and Tocher's optimization method to study the genetic diversity among cultivars in each management, and the relative contribution of traits to genetic diversity was evaluated according to the criterion described by Singh (1981). For the low tunnel and high tunnel environments, the crosses Aleluia x Camarosa, Aleluia x Aromas and Aleluia x Festival are the most promising to generate segregating populations with a higher possibility to appearance transgressive individuals, while for the open field cultivation system, we recommend the cross among Aleluia x Toyonoka. The variables that most contributed for genetic dissimilarity were total number of fruits, fruit yield and number of commercial fruits for the environments open field, low tunnel and high tunnel, respectively.

**KEYWORDS:** *Fragaria x ananassa*. Genotype x environment interaction. Plant breeding. *Tetranychus urticae*

### INTRODUCTION

Strawberry (*Fragaria x ananassa* Duch.) is a herbaceous and creeping plant, which although has characteristics of perennial crop, it is cultivated as annual (CALVETE et al., 2008). Its fruit has great economic importance, being the most popular, cultivated and consumed in the small fruit group (TAZZO et al., 2015). In Brazil, the crop is widespread in temperate and subtropical regions, where strawberries are produced for *in natura* consumption and also for industrialization (RADMANN et al., 2006). Its cultivation has important socio-economic role in Brazil's agricultural sector due to the increase of income in small properties and maintaining workers in rural areas (COSTA et al., 2015).

The main Brazilian strawberry producing regions are located in tropical high altitude areas. Commercial production of strawberries in Brazil is based on a few cultivars (CONTI et al., 2002a), the

most of them coming from breeding programs of other countries, such as The United States (Aromas, Camarosa, Dover, Oso Grande and Sweet Charlie) and Spain (Milsei-Tudla). Therefore, it is necessary to develop new cultivars adapted to Brazilian soil and climate conditions by encouraging national breeding programs (OLIVEIRA; SCIVITTARO, 2011).

In subtropical regions as Brazil, it is mainly cultivated during the winter and spring, predominantly under open field conditions. However, the area with protected cultivation in tunnels has increased, mainly to minimize the effects of environmental climatic factors, such as rainfall and high humidity, and to reduce the incidence of diseases (XIAO et al., 2001; OZDEMIR, 2003; OZDEMIR; GUNDUZ, 2004). Two types of tunnels have been most used in this crop: low tunnel, where the beds are protected individually and high tunnel with cultivation of several beds (OLIVEIRA et al., 2008; ANTUNES;

PEREZ, 2013). However, if the tunnels are not managed properly they may lead to increases in the population of two-spotted spider mite (*Tetranychus urticae*) Koch (Acari: Tetranychidae) (SVENSSON, 2006).

Thus, developing strawberry cultivars that can be grown under adverse conditions, it is necessary to gather desirable characteristics such as tolerance to two-spotted spider mite (TSSM), high fruit yield and wide adaptability to several cropping managements. In this sense, studying the genetic diversity among commercial cultivars based on agronomic traits and under different managements can contribute to identify segregating populations with high genetic variability and frequency of favorable alleles. Therefore, our objective was to evaluate the genetic diversity among 13 strawberry cultivars under different managements and to recommend promising crosses to obtain segregating populations with high fruit yield and TSSM tolerance.

## MATERIAL AND METHODS

Trial was conducted under field conditions at the Centro Regional de Desenvolvimento Rural Centro Serrano of the Instituto Capixaba para Assistência Técnica e Extensão Rural (Incaper), Domingos Martins, highlands of Espírito Santo State, (20° 22' 20 S and 41° 03' 76 W, and 950 m of altitude). We evaluated the following strawberry cultivars: Albion, Aleluia, Aromas, Camarosa, Camino Real, Campinas, Diamante, Dover, Festival, Seascape, Toyonoka, Tudla and Ventana. Three managements were evaluated: open field (without plastic cover), low tunnel (milky-colored plastic cover, 75 microns, suspended on one meter high) and high tunnel (milky-colored plastic cover, 100 microns, suspended on 2.5 m height), deployed in October (spring). The trial was conducted in a randomized complete block design, with three replications and 15 plants per plot (plot area 2.4 m<sup>2</sup>), spacing 40 x 40 cm, in three rows, on beds of 30 cm high and black mulching.

Drip irrigation was used and fertigation was carried out according to the soil analysis. Natural infestation of the *T. urticae* (TSSM) occurred from the trial implantation and it was evaluated using a non-destructive method, counting the number of two-spotted spider mite per cm<sup>2</sup> (NTSSM) on leaf (trifoliolate) per plant and five plants per plot, using manual lens with 20-fold increase. After 120 days, infestation was homogeneously controlled in all treatments to standardize the mite effect on production traits. Other agronomic traits were

evaluated: total number of fruits (TNF) per plot, number of commercial fruits per plot (NCF) and fruit yield (YIE, t/ha), which was measured in each plot and extrapolated to 1 ha considering a useful area of 7,500 m<sup>2</sup>/ha.

Initially, the count data (NTSSM, TNF and NCF) were transformed into ( $\sqrt{x + 1/2}$ ) and subjected to the Lilliefors and Bartlett tests. After checking that the relationship between the largest and smallest mean squared error of each management was less than seven, we performed the joint analysis of variance according to the following statistical model (Equation 2):

$$Y_{ijk} = \mu + B/M_{jk} + G_i + M_j + GM_{ij} + \epsilon_{ijk}$$

Wherein  $Y_{ijk}$  is the value observed in the  $i$ -th genotype evaluated in the  $j$ -th management in the  $k$ -th repetition;  $\mu$  is the overall mean;  $B/M_{jk}$  is the effect of the  $k$ -th block within  $j$ -th management;  $G_i$  is the effect of the  $i$ -th genotype considered as fixed;  $M_j$  is the effect of the  $j$ -th management considered as fixed;  $GM_{ij}$  is the effect of genotypes and managements interaction considered as fixed;  $\epsilon_{ijk}$  is the random error associated with  $Y_{ijk}$  the observation. Means were clustered by Skott and Knott test (1974).

For genetic diversity analysis, the genetic distance between different pair of genotypes was calculated, for each management, applying the generalized Mahalanobis distance ( $D_{ij}^2$ ). Thereafter, Tocher optimization method was used to cluster the cultivars based on the criterion that the intragroup are smaller than the intergroup distances (CRUZ et al., 2012). The relative contribution of traits to genetic diversity was evaluated according to the criterion described by Singh (1981). All analyzes were performed using Genes software (CRUZ, 2013).

## RESULTS AND DISCUSSION

By the joint analysis of variance, we verified that the effects of genotypes (G), managements (M) and GxM interaction were significant ( $p < 0.01$ ) for all evaluated traits (Table 1), corroborating the results observed by Costa et al. (2015). The existence of significant GxM interaction indicates that there were differentiated agronomic responses as a function of the management used, hindering a broad recommendation. Therefore, it is necessary to perform analyzes that can detect the phenotypic stability of these genotypes throughout the management used, which will allow to make a more reliable recommendation.

**Table 1.** Summary of joint analysis of variance for the traits number of two-spotted spider mite per cm<sup>2</sup> (NTSSM) total number of fruits (TNF), number of commercial fruits (NCF) and fruit yield (YIE, t/ha) evaluated in 13 strawberry cultivars grown in different managements.

Sources of variation	Degrees of freedom	NTSSM <sup>1</sup>	TNF <sup>1</sup>	NCF <sup>1</sup>	YIE
Blocks/Management	6	1.31	22.07	7.91	110.06
Genotypes (G)	12	9.40*	89.82*	58.70*	418.13*
Managements (M)	2	7.96*	707.17*	384.87*	1542.04*
G x M	24	4.83*	27.89*	17.93*	141.49*
Residue	72	0.99	4.59	3.29	27.55
Coefficient of variation (%)		12.31	6.41	7.45	14.18

\*: significant at 1% probability by F test; <sup>1</sup>: values for transformed data. G x M: genotypes x managements interaction.

It is important to mention that the estimates of coefficient of variation obtained were lower than 15% for all traits, indicating high experimental precision and credibility of the joint analysis. Estimates of coefficient of variation observed in the joint analysis are similar to those reported in other strawberry studies (RADMANN et al., 2006; MONCADA et al., 2008; RESENDE et al., 2010; RANDIN et al., 2011; COSTA et al., 2015; COSTA et al., 2016). According to Cruz et al. (2012), coefficient of variation lower than 20% indicates high experimental precision for quantitative traits, such as those evaluated in this study.

Number of two-spotted spider mite per cm<sup>2</sup> (NTSSM) observed in the strawberry genotypes as a function of the different managements is contained

in Table 2. The evaluated management systems influenced differently the TSSM incidence in each genotype. Camarosa cultivar was the one that showed lower values of NTSSM in all managements. At open field, only three cultivars had lower NTSSM (Toyonoka, Camarosa and Aromas). Toyonoka and Camarosa also had low NTSSM at low tunnel, together with Aleluia, Campinas, Diamante and Ventana. In general, high tunnel provided a lower TSSM incidence, except for Aleluia, Dover and Toyonoka. (Table 2). These results go in the opposite direction to that reported by Svensson (2006) and Demchak (2009), who states that production systems in plastic tunnel creates favorable conditions to incidence of two-spotted mite.

**Table 2.** Mean values for the two-spotted spider mite per cm<sup>2</sup> (NTSSM) evaluated in 13 strawberry cultivars grown in different managements.

Cultivar	Open field	Low tunnel	High tunnel
Albion	8.37 <sup>1</sup> aA 7.90 aA	9.93 aA	7.30 bB
Aleluia		6.80 bB	8.17 aA
Aromas	6.67 bB	9.00 aA	7.07 bB
Camarosa	5.80 bB	6.67 bB	6.73 bA
Camino Real	9.03 aA	8.77 aB	7.90 bB
Campinas	8.87 aA	7.97 aB	7.97 bB
Diamante	8.03 aA	7.27 bB	6.57 bB
Dover	8.37 aA	12.27 aA	10.87 aA
Festival	7.10 aB	8.90 aA	7.43 bA
Seascape	7.53 aB	9.67 aA	8.00 bA
Toyonoka	5.40 bC	7.13 bB	10.60 aA
Tudla	9.83 aA	10.10 aA	7.33 bB
Ventana	8.20 aA	7.17 bB	5.93 bC

<sup>1</sup>: values transformed via  $\sqrt{(x+1/2)}$ . Group of means followed by equal lower case letters in the same column and equal capital letters in the same line not differ by the Skott and Knott test (1974) at 5% probability.

Table 3 show the total number of fruits evaluated in strawberry cultivars according to each management. Again, high tunnel management provided satisfactory results, with the largest number of high performance cultivars (Aleluia, Aromas, Camarosa, Dover, Festival, Seascape, Toyonoka and Tudla). The lower means were observed for open field, where only two cultivars

(Aromas and Campinas) presented higher TNF. Camarosa cultivar showed high means both for open field and low tunnel, confirming the agronomic superiority of this cultivar under protect environments already reported in the literature (DUARTE FILHO et al., 2007; RESENDE et al., 2010).

**Table 3.** Mean values for total number of fruits (TNF) evaluated in 13 strawberry cultivars grown in different managements.

Cultivar	Open field	Low tunnel	High tunnel
Albion	24.10 <sup>1</sup> bB	29.83 bB	33.83 bA
Aleluia	25.77 cB	32.90 bB	42.47 aA
Aromas	33.23 bA	33.87 bA	39.27 aA
Camarosa	26.00 bB	38.10 aA	41.80 aA
Camino Real	24.00 bB	30.07 bA	32.80 bA
Campinas	39.50 aA	39.20 aA	33.10 bB
Diamante	24.37 bB	30.43 bA	30.80 bA
Dover	28.90 bB	35.67 bA	35.90 aA
Festival	32.97 bA	40.23 aA	42.67 aA
Seascape	30.23 bA	35.83 bA	38.00 aA
Toyonoka	28.87 bB	36.43 bA	39.03 aA
Tudla	30.37 bA	33.97 bA	36.60 aA
Ventana	24.60 bB	35.30 bA	33.33 bA

<sup>1</sup>: values transformed via  $\sqrt{(x + 1/2)}$ . Group of means followed by equal lower case letters in the same column and equal capital letters in the same line not differ by the Skott and Knott test (1974) at 5% probability.

Differently from what occurred for the previously mentioned traits, the three managements had similar behavior regarding the NFC (Table 4). Again, Camarosa presented great performance under protected environments (low and high tunnel), while the Festival was the only one that presented high means in all environments. These results indicate

the presence of G x M interaction, since the ranking of the best genotypes changes with the variation of the environment. Therefore, evaluating the performance of genotypes under different types of management is important, since it contributes to the recommendation of the most suitable genotypes for each cropping system.

**Table 4.** Mean values for number of commercial fruits (NCF) evaluated in 13 strawberry genotypes grown in different managements.

Cultivar	Open field	Low tunnel	High tunnel
Albion	18.70 <sup>1</sup> Bb	23.83 cB	27.57 bA
Aleluia	19.57 Bb	27.10 bA	33.23 aA
Aromas	26.60 Ab	26.73 bB	30.53 aA
Camarosa	20.20 Bb	29.23 aA	28.77 bA
Camino Real	16.20 Bb	24.33 cA	24.97 cA
Campinas	24.63 Aa	23.37 cA	22.33 cA
Diamante	18.77 Bb	25.10 cA	23.87 cA
Dover	18.80 Bb	22.77 cA	20.20 cA
Festival	24.70 Ab	30.97 aA	33.40 aA
Seascape	21.97 Ba	24.80 cA	25.07 cA

Toyonoka	20.80 Ba	25.40 cA	22.60 cA
Tudla	20.87 Ba	24.60 cA	25.93 cA
Ventana	17.63 Bb	27.30 bA	25.57 cA

<sup>1</sup>: values transformed via  $\sqrt{(x + 1/2)}$ . Group of means followed by equal lower case letters in the same column and equal capital letters in the same line not differ by the Skott and Knott test (1974) at 5% probability.

Fruit yield of strawberry cultivars according to the different managements is shown in Table 5. As in the other yield components (TNF and NCF), protected environments provided to all genotypes increased yield compared to the management open field. Aleluia cultivar obtained the high mean, much higher than the average crop yield in Brazil, which is 32.7 ton ha<sup>-1</sup> (ANTUNES; DUARTE FILHO,

2005). Festival presented high yield in all evaluated environments, confirming that this cultivar presents good performance regardless of the type of management. Resende et al. (2010) also found higher yield provided by low and high tunnel in comparison to open field, allowing to conclude that applying correctly the crop technology in protected environment allows to achieve high yield.

**Table 5.** Mean values for fruit yield (YIE, t/ha) evaluated in 13 strawberry cultivars grown in different managements.

Cultivar	Open field	Low tunnel	High tunnel
Albion	26.77 <sup>1</sup> bB	35.23 bA	41.73 bA
Aleluia	32.27 bB	45.07 aB	71.87 aA
Aromas	40.40 aA	36.63 bA	45.53 bA
Camarosa	28.47 bB	49.83 aA	49.63 bA
Camino Real	24.97 bB	42.20 aA	42.73 bA
Campinas	35.07 aA	32.30 bA	27.00 cB
Diamante	28.97 bB	40.27 bA	35.93 cA
Dover	20.83 bB	27.57 bA	24.37 cA
Festival	37.33 aA	49.90 aA	53.97 bA
Seascape	31.33 bA	37.93 bA	36.60 cA
Toyonoka	25.80 bB	36.20 bA	31.03 cA
Tudla	31.40 bA	34.07 bA	38.93 cA
Ventana	24.30 bB	48.50 aA	40.77 bA

<sup>1</sup>: values transformed via  $\sqrt{(x + 1/2)}$ . Group of means followed by equal lower case letters in the same column and equal capital letters in the same line not differ by the Skott and Knott test (1974) at 5% probability.

The lower yield, total number of fruits and number of commercial fruits observed for open field may be related mainly to the wetting of the aerial part, which besides contributing to the development of fungal diseases and bacterial growth, facilitates pollen washing of the inflorescences, reducing the fertility (DUARTE FILHO et al., 2007). In general, protected environment provides better conditions for the development and sanity of the plants, since the climatic conditions inside it allow greater expression of the physiological activities through a greater net photosynthesis (TAIZ; ZEIGER, 2004), which provides, consequently, improvement in traits related to yield.

Using protected cropping in strawberry cultivation has been an increasingly frequent practice, but its effect on the TSSM incidence in

strawberry has not been evaluated yet. High tunnel protected system has provided satisfactory agronomic performance for growing conditions of the State of Espírito Santo, Brazil, possibly due to the lower interference in the microclimate (BALBINO et al., 2006; COSTA et al., 2015). However, TSSM infestation levels under protected cropping system (high and low tunnel) were higher than the open field system for most genotypes. On the other hand, the use of protected environment in the strawberry crop has the advantage of discouraging diseases incidence. This is due to the lower accumulation of water on the leaves, which reduces the use of fungicides and enables strawberry production with greater food safety, greater plant longevity and yield (BALBINO et al., 2006). An alternative to attenuate the effects of protected

environments on the TSSM incidence is to find genotypes with greater tolerance to this pest.

The presence of GxM interaction indicates the need to study the genetic diversity among the cultivars in each management, since the most promising combinations for one type of management may not be the best for another. Thus, we estimated the Mahalanobis distances between the pairs of cultivars in each management and used the Tocher's optimization method, which clustered the cultivars into three groups in the open field, four groups in the low tunnel and five groups in the high

tunnel (Table 6). Although the number of groups is different for each management system, the cultivars Diamante, Ventana, Camino and Albion were maintained into Group I in the three environments. Dover and Toyonoka cultivars are allocated in the same groups in the environments open tunnel (Group II) and high tunnel (Group III). The identification of similar genotypes regardless of the management system is important because it indicates that these cultivars has many genes or alleles in common, and their expression is not altered by the management system.

**Table 6.** Cluster by Tocher's optimization method of 13 strawberry cultivars grown in different managements.

Group	Cultivars		
	Open field	Low tunnel	High tunnel
1	Diamante, Aleluia, Albion, Ventana, Camino, Camarosa Seascap, Tudla, Festival and Aromas	Diamante, Aleluia, Ventana, Camino and Albion Seascap, Tudla, Toyonoka	Ventana, Diamante, Camino and Albion
2	Dover and Toyonoka	Aromas and Festival	Aromas and Festival
3	Campinas	Dover and Campinas	Dover and Toyonoka
4		Camarosa	Seascap, Campinas, Tudla, and Camarosa
5			Aleluia

As the number of groups and allocation of genotypes into each group varied over the management, it was not possible to choose crosses that are promising for all environments. Thus, for the low tunnel and high tunnel environments, the crosses Aleluia x Camarosa, Aleluia x Aromas and Aleluia x Festival are the most promising to generate segregating populations with a higher possibility to appearance transgressive individuals, since they have a high yield potential (Aleluia cultivar) and a lower TSSM incidence (Aromas, Camarosa and Festival). As for the open field cultivation system, we recommend the cross among Aleluia x Toyonoka, which besides being divergent, they have a high yield (Aleluia) and a lower susceptibility to the TSSM, since the Toyonoka cultivar showed the lowest TSSM incidence at open field and low tunnel (Table 2). These results reinforce the presence of genetic variability among strawberry cultivars currently cultivated (GRAHAN

et al., 1996; CONTI et al., 2002a; CONTI et al., 2002b; RADMANN et al., 2006).

Using the Singh methodology (1981) was possible to evaluate the relative importance of the traits for the study of genetic diversity based on variance (Table 7). Variables with a low magnitude, redundancy and who have no stability (differential behavior in different environments) can be considered as unattractive in the diversity study (CRUZ et al., 2012). Relative contribution of the traits was distinct over the environments, and there was a trait with greater contribution for each environment.

**Table 7.** Relative contribution of the traits number of mites per cm<sup>2</sup> (NTSSM), total number of fruits (TNF), number of commercial fruits (NCF) and fruit yield (YIE) evaluated by Singh criterion (1981) for genetic divergence among 13 strawberry cultivars grown in different managements.

Trait	Open field		Low tunnel		High tunnel	
	S.j	Value (%)	S.j	Value (%)	S.j	Value (%)
NTSSM	504.80	10.99	572.72	8.18	346.72	4.89
TNF	3072.64	66.88	2809.50	40.13	1718.72	24.25
NCF	704.64	15.34	511.22	7.30	3037.53	42.86
YIE	312.15	6.79	3106.91	44.38	1983.80	27.99

For the open field, the most important variable for genetic dissimilarity was TNF, contributing with 66.88% of the diversity. The variable YIE was the one that presented the greatest contribution to diversity in the low tunnel environment, representing 44.38% of the variation. At last, the NFC, with a contribution of 42.86%, it

was the most important trait for genetic dissimilarity in the high tunnel. The NTSSM contributed little to the diversity among cultivars, regardless of management. However, its disposal in breeding programs is not advisable because it allows to select genotypes that are more tolerant to TSSM and require less use of insecticides.

**RESUMO:** Para desenvolver cultivares de morango que podem ser cultivados em larga escala é necessário reunir características desejáveis como: tolerância ao *Tetranychus urticae*, alta produtividade de frutos e ampla adaptabilidade a diversos sistemas de cultivo. Portanto, o objetivo do trabalho foi estudar a diversidade genética entre 13 cultivares de morango sob diferentes manejos e recomendar cruzamentos promissores para obtenção de populações segregantes com alta produtividade de frutos e tolerantes ao *T. urticae*. O experimento foi conduzido sob condições de campo no Centro Regional de Desenvolvimento Rural Centro Serrano do Instituto Capixaba de Assistência Técnica e Extensão Rural (Incaper), Domingos Martins-ES, no mês de outubro (primavera). Foram avaliadas as cultivares Albion, Aleluia, Aromas, Camarosa, Camino Real, Campinas, Diamante, Dover, Festival, Seascape, Toyonoka, Tudla e Ventana, cultivadas em três sistemas de cultivo: campo aberto, túnel baixo e túnel alto. O delineamento experimental utilizado foi blocos casualizados com três repetições. As variáveis avaliadas foram: número de ácaro/cm<sup>2</sup> na folha (NTSSM), número total de frutos (TNF), número de frutos comerciais (NCF) e produtividade de frutos (YIE, t/ha). Foram empregadas a distância generalizada de Mahalanobis e o método de otimização de Tocher para o estudo da diversidade genética entre os cultivares em cada manejo, e a contribuição relativa dos caracteres para a diversidade genética foi avaliada segundo o critério de Singh (1981). Para os manejos túnel baixo e túnel alto, os cruzamentos entre os cultivares Aleluia x Camarosa, Aleluia x Aromas e Aleluia x Festival são os mais promissores para gerar populações segregantes com alta possibilidade de aparecimento de indivíduos transgressivos, enquanto que para o campo aberto recomenda-se o cruzamento entre os cultivares Aleluia x Toyonoka. As variáveis que mais contribuíram para a dissimilaridade genética foram o número total de frutos, produtividade e número de frutos comerciais para os ambientes campo aberto, túnel baixo e túnel alto, respectivamente.

**PALAVRAS-CHAVE:** *Fragaria* x *ananassa*. Interação genótipo x ambiente. Melhoramento de plantas. *Tetranychus urticae*

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