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Characterization of potato genotypes for resistance to *Diabrotica speciosa*

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ABSTRACT

The potato crop is attacked by many pests, among which the *Diabrotica speciosa* (Coleoptera: Chrysomelidae) is considered the main one. Considering chemical control failures associated with the requirements of integrated potato production (IPP) requiring sustainable measures, this study aimed to characterize advanced clones breeding program of Embrapa for resistance to insect infestation in artificial conditions. Tests were carried out in a greenhouse without choice of plants and tubers of four clones of Embrapa (C2337-06-02, C2337-18-02, C2342-1-02, C2362-02-02), a standard-resistant clone (NYL 235-4) and a susceptible standard cultivar (Asterix). Results of susceptibility of 'Asterix' and resistance of 'NYL 235-4' and 'C2342-1-02', both in shoots and in tubers, predominated. Thus indicating a typical behavior of tolerance of 'NYL 235-4' and 'C2337-06-02' to insect attack, both in the shoots and in the tubers. Clones C2337-06-02 and C2362-02-02 expressed resistance to the attack of the young stage of the insect in the tubers. Three potato genotypes derived from the wild species *Solanum berthaultii* (NYL 235-4, C2337-06-02, C2362-02-02) even under infestation of *D. speciosa*, showed agronomic traits determinant of greater commercial acceptance of tubers such as steady productivity, elongated shape, skin smoothness and shallower depth of the eye (vegetative bud).

Keywords: *Solanum tuberosum*, corn rootworm, leaves, tubers, plant resistance.

RESUMO

Resistência de genótipos de batata a *Diabrotica speciosa*

A cultura da batata é hospedeira de diversos insetos, sendo a vaquinha, *Diabrotica speciosa* (Coleoptera: Chrysomelidae), um dos mais nocivos. Devido às falhas de controle químico, associadas às exigências da produção integrada da batata (PIB) exigindo medidas sustentáveis, objetivou-se caracterizar clones avançados do programa de melhoramento genético de batata da Embrapa quanto à resistência ao inseto, em condições de infestação artificial. Realizaram-se ensaios sem chance de escolha em casa-de-vegetação, utilizando-se plantas e tubérculos de quatro clones da Embrapa (C2337-06-02, C2337-18-02, C2342-1-02 e C2362-02-02), um clone padrão-resistente (NYL 235-4) e uma cultivar padrão-suscetível (Asterix). Predominaram resultados de suscetibilidade da 'Asterix' e de resistência do 'NYL 235-4' e 'C2342-1-02', tanto na parte aérea como em tubérculos. Evidenciou-se comportamento típico de tolerância de 'NYL 235-4' e 'C2337-06-02' ao ataque do inseto, tanto na parte aérea das plantas como nos tubérculos. Os clones C2337-06-02 e C2362-02-02 manifestaram resistência ao ataque de larvas nos tubérculos. Três clones de batata derivados da espécie silvestre *Solanum berthaultii* (NYL 235-4, C2337-06-02, C2362-02-02), mesmo sob infestação de *D. speciosa*, apresentaram caracteres agrônômicos determinantes de maior aceitação comercial de tubérculos tais como manutenção de produtividade, formato alongado, película lisa e menor profundidade do olho (gema vegetativa).

Palavras-chave: *Solanum tuberosum*, vaquinha, folhas, tubérculos, resistência de plantas.

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The potato (*Solanum tuberosum*) is one of the most important crops in the world, surpassed in production, in terms of human consumption, only by rice and wheat (CIP, 2010). Due to high demands on the quality of tubers, it is essential to know the main phytosanitary problems in crop in the growing environment, besides the influence of management, fertilization and the cultivars chosen (Nunes, 2002; Quadros *et al.*, 2009), as these will set the quality of the final product.

Among the various species of insects that attack the potato crop, *Diabrotica speciosa* is considered one of the major (Souza *et al.*, 2006). In adult form (beetle), feeding on leaflets causes the roughness of the leaf and therefore reduces production of tubers (Lara *et al.*, 2004). In the larval stage, known as rootworm, acting as soil pest, the insect damages stolons and forms galleries in tubers (Furiatti, 2009). Control is done systematically with chemical insecticides, applied in a generalized

way in planting furrows or via foliar sprays (Souza *et al.*, 2008).

The integrated potato production system, besides being economic, aims to meet the final product, prioritizing the use of proper management and rational use of agrochemicals, in order to minimize environmental impact and to improve the quality of human health. Thus, the overall goal of the system is to raise the quality standards and competitiveness of the potato, through the sustainable management of

agricultural production (Boulomytis & Júnior, 2013).

Due to some disadvantages of using chemical insecticides on potatoes, other control methods against *D. speciosa* have been studied, including the host plant resistance (Furiatti, 2009). This method, among other advantages, can keep the insect population in lower levels in relation to the economic thresholds, reducing the expenses on control and environmental impact (Lara *et al.*, 2004). In order to obtain cultivars resistant to arthropod pests like *D. speciosa*, studies need to be done to clarify the causes of different types of resistance (antibiosis, antixenosis and/or tolerance), which in *Solanum* spp. can be associated with morphological and/or chemical mechanisms of plants such as trichomes and hydrocarbon respectively (Fernandes *et al.*, 2012).

The leave trichomes of wild species of *Solanum berthaultii*, by releasing adhesive viscous substances, provide protection against numerous insect pests of potato (Castell *et al.*, 2006), including Coleoptera Chrysomelidae family, as the Colorado potato beetle *Leptinotarsa decemlineata* (Coleoptera: Chrysomelidae) and *D. speciosa* (Lara *et al.*, 2000).

Combining the resistance of wild potato species and agronomic traits of cultivated species is one of the efforts of the crop breeding programs. One of the main results is the cultivar “Prince Hairy” NYL 235-4, resistant to *L. decemlineata*, derived from crosses of wild potato (*S. berthaultii*), which, besides containing good agronomic traits, loses three to seven times less productivity than genotypes susceptible to the insect. The high level of resistance of NYL 235-4, non-preference for feeding (antixenosis) type, has been associated with the presence of glandular trichomes on the foliage (Fisher *et al.*, 2002).

In Brazil, NYL 235-4, a clone with resistance to the Colorado potato beetle, has been used as standard in studies of the resistance of potato to *D. speciosa* (Lara *et al.*, 2000, 2004; Souza *et al.*, 2005, 2006; Furiatti, 2009), including as a gene provider. In this context, clonal families C2337 and C2362, developed

by the potato breeding program of Embrapa and containing NYL 235-4 genes, demonstrated foliar resistance to *Diabrotica* spp. (Souza *et al.*, 2008). The cultivar Asterix has been used as a pattern of susceptibility to the insect (Salles, 2000).

Experiments on the resistance of potato to *D. speciosa* have been carried out in greenhouse, in tests with and without choice of plant by the insect. Initially, plants, in pots hidden by cages lined with screen, are infested with adult insects, for visual estimation of leaf area consumed (Lara *et al.*, 2004), being subsequently evaluated the index of damage caused by larvae to tubers (Salles, 2000).

Based on available knowledge about the effects that plants of *Solanum* spp. can exert on different species of Chrysomelids, this research aimed to characterize the behavior of six advanced clones of the potato breeding program of Embrapa in relation to the resistance to *D. speciosa*, in conditions of artificial infestation at greenhouse.

MATERIAL AND METHODS

The experiment was conducted on April 4, 2012, in a greenhouse at Temperate Climate Center of Brazilian Agricultural Research Corporation, Rio Grande do Sul state, Brazil. Six genotypes were evaluated, four being advanced potato clones developed by Embrapa Potato Breeding Program, with *S. berthaultii* in their genealogies (C2337-06-02, C2337-18-02, C2342-01-02, C2362-02-02) and unknown reaction to the insect; one clone resistant to *D. speciosa*, cv. “Prince Hairy” NYL 235-4 (Furiatti, 2009) and one susceptible cultivar, Asterix (Salles, 2000).

Tubers were planted in polyethylene bags, with a capacity of 5 L, containing Plantmax® peat substrate. The plants (one per pot), under temperature of 18 to 30°C were alternately irrigated every two days and treated, on June 8, 2012, with the fungicide mefenoxan (Metalaxyl-M) + mancozeb at doses of 250 mL/100 L, aiming to avoid the incidence of diseases such as late blight, caused by the fungus

Phytophthora infestans. In the previous plant infestation period, a luminous trap was turned on, daily, at sunset, in the greenhouse, to catch insect pests of natural occurrence.

During the stolon emission phase at 30 days after planting (DAP), the pots with the plants were arranged in factorial design (two plants/genotype) in six replications, and covered individually by a cylindrical cage [1.2 m (height) x 0.6 m (diameter)] of wire and screened mesh fabric (tulle). The experimental plot consisted of two plants of each genotype. Right after the isolation in cages, the plants from one of the rows were infested by five *D. speciosa* couples; the couples were sexed according to White (1977), establishing a no-choice condition of the host plant. The plants from the other row (controls) remained uninfested in order to establish the rate of production decrease.

Adults of *D. speciosa* used in this research were obtained by the method of creation described by Ávila *et al.* (2000), however, with modifications. In the larval stage, the insect was raised on a natural diet consisting of maize seedlings, containing the expanded vermiculite as substrate (sterile) in plastic pots (20 cm diameter x 10 cm height). For this purpose, a first layer of vermiculite (40 g) was placed in the bottom of a plastic container and on this layer distilled water was used (80 mL). Then, 50 maize seeds were placed on the first layer of vermiculite and covered by one second layer of this material (50 g), and more 100 mL of distilled water was added, being the containers packed in heated room (25±2°C; UR 60±10%). After four days, when the seminal radicles have emerged, the seedlings were infested in the roots with 150 neonate larvae (3 larvae/seedling) using a thin bristle brush. After 10 days of growth, about 100 larvae were transferred, by sieving, from the infested container to plastic container containing vermiculite and 50 new maize seedlings (obtained by the process described above). The larvae were kept in this container up to reach their pupal stage.

Adults of *D. speciosa* that emerged, still in the larval rearing containers were packed in acrylic cage (39 cm wide x

51 cm long x 51 cm high) containing a diet consisting of one plant of bean, cultivar Carioca (maintained in an Erlenmeyer flask with water), carrot slices (1 cm wide x 2 cm diameter) and honey solution (10%) in cotton wool. The petioles of beans were fixed (with cotton) to the neck of the Erlenmeyer flask, whereas the sliced carrot and honey solution (cotton) were kept in Petri dishes.

A month after infestation, at the phenological stage of tuberization, injuries on the leaflets of potato caused by *D. speciosa* were visually estimated, attributing grades (% of consumed area) in the lower, middle and top thirds of the plants. For this purpose, the method of Furiatti (2009) was adopted, based on the degree of defoliation using the following grading scale: zero = no defoliation; 1 = up to 5%; 2 = 5 to 10%; 3 = 10 to 15%; 4 = 15 to 20%; 5 = 20 to 30%. In the experiment, however, two more defoliation grades were used: 6 = 30 to 45% and 7 = greater than 50%.

At 70 DAP, at tuber growth stage, in order to keep the infestation of the potato genotype by *D. speciosa*, of the leaves of each plant, more three couples (adult) were released and in the roots, using a fine moistened brush, eight larvae until 3rd instar. The experiment was completed on July 16, 2012 at 103 DAP (stage of tuber maturation and plant senescence), when the number of perforations, caused by the larvae, in all tubers harvested from each plant was recorded.

The evaluation of the agronomic traits was based on the methods describe by Souza *et al.* (2005), attributing grades to the appearance of tubers according to the following scale: 1 = excellent appearance (attractive light or intense red-colored tubers, smooth skin, elongated and uniform shape, shallow eyes, medium to large size) and 5 = very bad appearance (rough tubers, deep eyes, low productivity, irregular shape and other defects); In relation to tuber uniformity, the grades were 1 = uniform (tubers with uniform sizes and shapes) up to 5 = uneven (tubers with different sizes and shapes). The tubers were classified according to the transverse diameter, in

order to evaluate its production. Those with more than 45 mm diameter were considered commercial, the number and the weight were recorded. The roughness of the tubers was evaluated in comparison to the witnesses, using the grades 1 (smooth) up to 5 (rough). For the eye-depth (vegetative bud), the grades were also from 1 (shallow eye) to 5 (deep eye).

The rate of the reduction of production (% RP) of potato genotypes was calculated using the formula: $RP\% = [(S - C) / S] \times 100$, where S and C are the weight of tubers in plots without and with infestation of adults and larvae of *D. speciosa* respectively.

The original data obtained, processed in $\sqrt{x+0.5}$ (grades attributed to the defoliation in the lower, middle and top thirds of the plants; number of holes in the tubers; average weight; total production; total number of tubers; appearance; uniformity; roughness of the skin and eye depth) or arc sen $\sqrt{x/100}$ (reduction of production; % of marketable tubers) to standardize and reduce the heterogeneity of variations, were submitted to the analysis of variance using Genes, v. 7.0 software (Programa Genes, 2009), and the averages were compared by the Scott & Knott test at 5% significance.

RESULTS AND DISCUSSION

Under without choice condition of potato genotypes by adults of *D. speciosa*, at 30 days after the infestation, some significant differences were observed in relation to defoliation of the lower and middle thirds of the plants (Table 1). The genotypes differed, however, in relation to the attack on the top third of the plant. The resistant clone NYL 235-4 and the advanced clones C2342-01-02 and C2337-18-02 formed a less defoliated group, whereas the clones C2337-06-02 and C2362-02-02, together with cultivar Asterix, were the most defoliated group. In the general context of defoliation, in the three parts of the plants, a higher level of resistance behavior was observed in clones C2342-01-02 and C2337-18-02, similarly to the resistant clone NYL 235-4.

The significant differences between

potato genotypes in relation to defoliation by *D. speciosa* observed one month after the infestation only on the top third of the plants, where the attack was greater (Table 1), highlighting the clone NYL 235-4 (resistance pattern) among the least attacked ones, showed this part of the plant to be the most favorable for evaluation of insect resistance. This distinction of greater attack and, therefore, better differentiation about leaf consumption on the top third of the plants becomes important, considering that variation in the rate of damage due to the age and plant part attacked may occur (Cranshaw & Radcliffe, 1980). Furthermore, as in preference for feeding test, without choice of the host plant, the insect can cause considerable damage even to genotypes with resistance characteristics (Lara, 1991), the low rate of defoliation of the clones C2342-01-02 and C2337-18-02 is even more evident.

Three groups of potato genotypes differed significantly in relation to drilling of tubers by larvae of *D. speciosa* (Table 1). The cultivar Asterix, the most attacked one, confirmed to be susceptible to the insect. The clone C2337-18-02 showed intermediate damage whereas the clones C2337-06-02, C2342-01-02, C2362-02-02 and NYL 235-4 showed resistant behavior to the attack by larvae. The authors observed that the clone C2337-18-02, with the lowest level of defoliation, showed intermediate rate of drilling on tubers. This behavior can be related to the lack of relationship between the damage in leaves and tubers, caused by adults and larvae of *D. speciosa*, respectively (Furiatti, 2009). On the other hand, potato plants submitted to defoliation stresses, resulting from innumerable factors, increase in tubers, the content of glycoalkaloid solanine, which can be considered a natural insecticide for many insect species as *L. decemlineata* and *Empoasca fabae* (Hemiptera: Cicadellidae) (Hlywka *et al.*, 1994) which may have interfered, at different levels, in the behavior of genotypes evaluated. Therefore, the behavior of the clone C2337-06-02, with higher and lower rates of defoliation and drilling of tubers by *D. speciosa*, respectively (Table 1), can be the result of an increase in alkaloid content.

Table 1. Grades attributed to defoliation transformed into $\sqrt{x+0.5}$, in lower (TI), medium (TM) and superior third of the plants (TS), at 59 days after emergence, caused by artificial infestation of adults and number of holes in tubers (NFT), at 103 days post planting caused by the larvae *Diabrotica speciosa* in six potato genotypes in greenhouse [notas atribuídas à desfolha transformadas em $\sqrt{x+0.5}$, nos terços inferior (TI), médio (TM) e superior (TS), aos 59 dias após a emergência, provocada por infestação artificial de adultos e número de orifícios nos tubérculos (NFT), aos 103 dias pós plantio, provocado pela larva-alfinete de *Diabrotica speciosa*, em seis genótipos de batata em casa-de-vegetação]. Capão do Leão, Embrapa Clima Temperado, 2012.

Genotype	TI	TM	TS	Average value	NFT
NYL 235-4	1.8 a A ¹	1.8 a A	2.2 b C	2.3 b	12.0 (n:6.5) ² c
C2342-1-02	1.6 a A	2.6 a A	3.2 b B	2.9 b	31.0 (n:3.2) c
C2337-18-02	2.5 a A	2.7 a A	3.4 b B	3.3 b	50.8 (n:6.7) b
C2337-06-02	2.4 a A	2.7 a A	4.1 a A	3.7 a	18.0 (n:3.5) c
Asterix	2.1 a A	3.1 a A	4.3 a A	4.2 a	81.8 (n:5.0) a
C2362-02-02	2.3 a A	2.6 a A	4.7 a A	4.2 a	25.8 (n:4.7) c
Means	2.1	2.6	3.7	3.4	36.5
CV (%)	16.97	12.59	8.32	8.25	20.14

¹Means followed by the same letter, lowercase in columns and uppercase in rows, do not differ, Scott & Knott test, 5%; ²averages of assessed tubers. Scale of zero = no defoliation; 1 = up to 5%; 2 = 5 to 10%; 3 = 10-15%; 4 = 15-20%; 5 = 20-30%; 6 = 30-45% and 7 = greater than 50% (valores médios seguidos por letras minúsculas, iguais na coluna e maiúsculas iguais na linha, não diferem, teste Scott & Knott, 5%; ²média de tubérculos avaliados. Escala de notas zero= sem desfolha; 1= até 5%; 2= 5 a 10%; 3= 10 a 15%; 4= 15 a 20%; 5= 20 a 30%; 6= 30 a 45% e 7= superior a 50%).

Despite the possibility of the defense mechanisms against *D. speciosa* in leaves and tubers of the potato plants being independent (Souza *et al.*, 2006), identifying genotypes as NYL 235-4 and C2342-1-02, which expresses resistant behavior in both parts of the plants, is advantageous (Table 1).

For the evaluation of agronomic traits of potato genotypes, with and without infestation of larvae and adults of *D. speciosa* (Table 2), significant differences between the average weights of tubers of the plants submitted to the insect attack were not observed. On the contrary, in the condition of uninfected plants, the clones C2362-02-02, C2342-01-02 and NYL 235-4 stood out, forming a group with heavier tubers. The cultivar "Asterix" and the clones C2337-06-02 and C2337-18-02 formed another group with tubers with lower average weight.

Under infestation of larvae and adults of *D. speciosa*, the clones NYL 235-4 and C2337-18-02 reached highest total weight of tubers, producing an average of 6.5 more tubers/plant than the other genotypes, whose production did not differ (Table 2). This result is in accordance with the finding that the

clones NYL 235-4 and 116-20 NDG, in field conditions, showed higher total production compared to cultivars Baronesa and Elvira (Souza *et al.*, 2006), showing their good adaptation to growing conditions in the Southern region.

Under the condition of plants without infestation by larvae and adults of *D. speciosa*, the clone C2337-18-02 stood out in relation to the production (total weight of tubers) not differing from the clone NYL 235-4. The clones C2337-06-02, C2342-01-02, C2362-02-02 and the cultivar Asterix (susceptible witness) formed a group of genotypes with the lowest total weight of tubers. These results corroborate the observations of a population composed of combinations performed between materials which are resistant and also adapted to the South part of Brazil, under natural infestation of larvae and adults of *D. speciosa*, highlighting C2337 (Whyte Lady/ND140) and C2362 (Cristal/NYL 235-4), that means, the same parental genotypes C2337-06-2, C2337-18-02 and C2362-02-02 for the proper performance of the total tuber production (Souza *et al.*, 2008).

The clones C2337-06-02 and

NYL-235-4 showed tolerance to *D. speciosa* attack, showing the lowest rate of reduction in tubers production, differing from the other genotypes which formed another group with higher losses (Table 2). Thus, the clones C2341-01-02 and C2337-18-02 that expressed non-preference for feeding as a resistance, when evaluated under aspect of tolerance to the insect attack, showed susceptibility behavior. This may have resulted from a likely preference for oviposition in certain genotypes associated with many factors of plants, as described by Lara (1991). Moreover, the potato plants have high sprouting ability, allowing to tolerate drastic defoliation levels, avoiding significant reductions in the production of tubers (Furiatti, 2009).

For the number of tubers produced per plant (NTT) (Table 2), no genotype difference in relation to the insect attack was observed. However, under the condition of plants without infestation by larvae and adults of *D. speciosa*, the genotype C2337-18-02 stood out with an average of 8.0 tubers, differing from the rest, which formed another group.

In the evaluation of commercial tuber production, no difference between the advanced clones and the commercial witnesses Asterix and NYL- 235-4, under artificial infestation of larvae and adults of *D. speciosa*, was noticed. However, for the condition of plants without insect infestation, the clone C2342-1-02 did not show any difference comparing to the resistant witness NYL 235- 4, with production of 100% of commercial tubers. The other clones formed one group together with the cultivar (susceptible witness). These observations reinforce the predominance of resistant parents of the wild species *S. berthaultii* for these yield component traits, total number of tubers produced per plant and commercial classification, observed by Souza *et al.* (2008), since in potato breeding it is necessary to direct those combinations wherein the resistance in significant level is transmitted to most progenies providing maintenance and rising agronomic quality standards of the material on which is being working (Darmo & Peloquin, 1991).

Table 2. Potato genotypes evaluated with and without infestation (CI and SI) of *Diabrotica speciosa* under greenhouse and means of treatments for the variables weight (PM) (g), total production (PT) (g), reduction of production (RP) (%), total number of tubers (NTT) and % of marketable tubers (%TC) {genótipos de batata avaliados com e sem infestação (CI e SI) de *Diabrotica speciosa* em casa-de-vegetação e média dos tratamentos para as variáveis: peso médio (PM) (g), produção total (PT) (g), redução de produção (RP) (%), n° tubérculos totais (NTT) e % tubérculos comerciais (%TC)}. Capão-do-Leão, Embrapa Clima Temperado, 2012.

Genotype	PM (g)		PT (g)		RP (%)	NTT		TC (%)	
	CI	SI	CI	SI		CI	SI	CI	SI
Asterix	13.6 a A ¹	14.5 b A	62.1 (n:30) ² b A	78.2 (n:33) b A	20.6 a	5.0 a A	5.7 b A	82.8 a A	87.8 b A
C2337-06-02	29.9 a A	18.3 b A	83.7 (n:21) b A	86.5 (n:30) b A	3.2 b	3.5 a A	4.8 b A	92.5 a A	88.5 b A
C2362-02-02	18.2 a A	21.8 a A	76.8 (n:28) b A	101.8(n:31) b A	24.6 a	4.7 a A	4.6 b A	94.4 a A	93.3 b A
C2342-1-02	31.8 a A	29.1 a A	95.2 (n:19) b A	108.2(n:24) b A	12.0 a	3.2 a A	4.7 b A	95.8 a A	100.0 a A
NYL 235-4	29.3 a A	22.4 a A	124.2 (n:39) a A	132.1(n:36) a A	6.0 b	6.5 a A	6.0 b A	84.2 a A	100.0 a A
C2337-18-02	19.5 a A	18.8 b A	127.5 (n:40) a A	150.1 (n:48) a A	15.1 a	6.7 a A	8.0 a A	91.5 a A	87.5 b A
Means	23.7	20.8	94.9	109.5	13.6	4.9	5.6	90.2	92.8
CV (%)	30.41	29.29	14.41	24.52	18.05	19.91	24.09	20.02	11.34

¹Means followed by same letters, lowercase in columns, and uppercase on lines, do not differ, Scott & Knott, 5%; ²average values of evaluated tubers (¹médias seguidas da mesma letra, minúscula na coluna, e maiúscula na linha, não diferem, Scott & Knott, 5%; ²média de tubérculos avaliados).

Regarding the characteristics of tubers, the cultivar Asterix and the clone C2337-06-02 showed intermediate appearance of the tubers (grade= 3), differing from the other genotypes, whose appearance was very bad (grade= 5), including clone NYL-235-4, highly resistant to the attack of rootworm (Table 3). In the condition of plants without infestation by larvae and adults of *D. speciosa*, the genotypes Asterix, NYL 235-4, C2337-06-02 and C2337-18-02 showed excellent appearance of tubers, comparing to clones C2342-1-02 and C2362-02-

02, with lower appearance. Although the overall appearance is a subjective character, whose evaluation depends on the experience of the breeder, in the fall, the degree of appearance decreases as climate conditions becomes adverse to the normal development of potato plants (Andreu, 2005).

No differences between the genotypes, in relation to the uniformity of the tubers produced by plants submitted to infestation of larvae and adults of *D. speciosa* were observed. However, under the condition of plants without infestation, the clones C2337-

06-02, C2337-18-02 and C2362-02-02 formed tubers with more elongated size and shape, differing from the other genotypes, which showed rounded shape, including witness cultivars Asterix and NYL 235-4.

Under the condition of plants infested by *D. speciosa*, six genotypes were graded ($4.3 \leq AP \leq 5.0$) for greater roughness of the tuber skin (Table 3). The same result was noticed in relation to most of the genotypes, in the condition of plants without infestation, except clone C2337-06-02, which showed smoother tubers (grade= 1.7).

Table 3. Appearance (A), uniformity (U), skin roughness (AP) and depth of eyes (PO) of potato genotypes with and without infestation (CI and SI) of *Diabrotica speciosa* under greenhouse [aparência (A), uniformidade (U), aspereza da película (AP) e profundidade de olho (PO) de genótipos de batata com e sem infestação (CI e SI) de *Diabrotica speciosa* em casa-de-vegetação]. Capão-do-Leão, Embrapa Clima Temperado, 2012.

Genotype	A		U		AP		PO	
	CI	SI	CI	SI	CI	SI	CI	SI
Asterix	3.0 b A ¹	2.3 b A	3.0 a A	3.0 a A	5.0 a A	5.0 a A	4.3 a A	3.0 a A
NYL235-4	5.0 a A	1.0 b A	5.0 a A	3.7 a A	5.0 a A	5.0 a A	5.0 a A	1.0 b A
C2337-06-02	2.3 b A	1.7 b A	2.3 a A	1.7 b A	4.3 a A	1.7 b A	2.3 a A	2.3 a A
C2337-18-02	5.0 a A	1.0 b A	4.3 a A	1.0 b A	5.0 a A	5.0 a A	3.7 a A	1.0 b A
C2342-1-02	5.0 a A	4.3 a A	4.3 a A	4.3 a A	5.0 a A	4.3 a A	4.3 a A	2.3 a A
C2362-02-02	5.0 a A	5.0 a A	3.0 a A	2.3 b A	5.0 a A	5.0 a A	1.7 b A	1.0 b A
Means	4.2	2.6	3.7	2.7	4.9	4.3	3.5	1.8
CV (%)	16.19	22.49	23.66	27.31	8.07	11.99	25.02	29.85

¹Means followed by same letters, lowercase on the column, and uppercase on the line, do not differ, Scott & Knott, 5%; A= tuber appearance (1= very good to 5= poor); U= uniformity of tubers (1= uniform to 5= disuniform); AP= skin roughness (1= smooth to 5= rough); PO= depth of the eyes (1= shallow to 5= deep) {¹médias seguidas da mesma letra, minúscula na coluna e maiúscula na linha, não diferem, Scott & Knott, 5%; A= aparência de tubérculo (1= ótima a 5= péssima); U= uniformidade de tubérculo (1= uniforme a 5= desuniforme); AP= aspereza de película (1= lisa a 5= áspera); PO= profundidade do olho (1= raso a 5= profundo)}.

The roughness of the tubers has great influence on commercial acceptance of *in natura* product by consumers, who prefer the smooth and shiny plants (Souza *et al.*, 2008). The texture of the skin, despite being a high heritability trait, is highly affected by environmental changes such as humidity, soil type and disease (Andreu, 2005), which could be related to the results obtained in this work.

In relation to eye-depth (vegetative bud), in the tubers, the clone C2362-02-02 stood out from the others, showing shallower structures, in the condition of plants infested by larvae and adults of *D. speciosa* (Table 3). Under the condition of plants without infestation, the clones NYL 235-4, C2337-18-02 and C2362-02-02 formed one group with lower value for eye depth, differing from the other genotypes. Some studies infer the possibility of selecting, in the South-Brazilian conditions, clones of potato introduced with high levels of resistance to attack by insect pests and acceptable agronomic traits (Souza *et al.*, 2006) on the contrary, that resistant clones from crosses between *S. tuberosum* and wild species can add numerous genetic traits that are undesirable from the agronomic point of view, such as tubers with very deep eyes (Kalazich & Plaisted, 1991). Thus, in the potato breeding program process, some strategies should be adopted to give greater efficiency in the selection of genotypes, in any growing season, searching for desirable agronomic traits, such as the shallowest depth of vegetative buds ('eyes').

A further analysis of the results of this work shows that the methodology used in artificial infestation of plants and tubers of potato, in greenhouse conditions, with larvae and adults of *D. speciosa*, without choice of host genotype (advanced clones and cultivars), serves to characterize the degree of resistance to insect attack. More specifically, the authors highlight the following results: 1) confirmation of the susceptibility of the cultivar Asterix and the resistance of clones C2342-01-02 and NYL 235-4, both in the shoots and tubers; 2) response for the resistance of the clones C2337-06-02, C2342-

01-02, C2362-02-02 in relation to the drilling of tubers by larvae; 3) evidence that the clones NYL 235-4 and C2337-06-02 tolerate attack of *D. speciosa* larvae, based on a smaller loss of tuber production. Additionally, among the evaluated potato genotypes, derived from the wild species *S. berthaultii* under infestation of *D. speciosa*, some showed to add one or more determinant agronomic traits of greater commercial tubers acceptance, as elongated shape, smooth skin and shallower eyes.

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