

Characterization of cowpea genotype resistance to *Callosobruchus maculatus*

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Abstract – The objective of this work was to characterize the resistance of 50 cowpea (*Vigna unguiculata*) genotypes to *Callosobruchus maculatus*. A completely randomized design with five replicates per treatment (genotype) was used. No-choice tests were performed using the 50 cowpea genotypes to evaluate the preference for oviposition and the development of the weevil. The genotypes IT85 F-2687, MN05-841 B-49, MNC99-508-1, MNC99-510-8, TVu 1593, Canapuzinho-1-2, and Sanzi Sambili show non-preference-type resistance (oviposition and feeding). IT81 D-1045 Ereto and IT81 D-1045 Enramador exhibit antibiosis against *C. maculatus* and descend from resistant genitors, which grants them potential to be used in future crossings to obtain cowpea varieties with higher levels of resistance.

Index terms: *Vigna unguiculata*, Bruchinae, Chrysomelidae, plant resistance, storage.

Caracterização da resistência de genótipos de feijão-caupi a *Callosobruchus maculatus*

Resumo – O objetivo deste trabalho foi caracterizar a resistência de 50 genótipos de feijão-caupi (*Vigna unguiculata*) a *Callosobruchus maculatus*. Utilizou-se o delineamento inteiramente casualizado com cinco repetições por tratamento (genótipo). Testes de confinamento foram realizados com os 50 genótipos de feijão-caupi, para avaliar a preferência para oviposição e o desenvolvimento do caruncho. Os genótipos IT85 F-2687, MN05-841 B-49, MNC99-508-1, MNC99-510-8, TVu 1593, Canapuzinho-1-2 e Sanzi Sambili apresentam resistência do tipo não preferência (oviposição e alimentação). IT81 D-1045 Ereto e IT81 D-1045 Enramador apresentam antibiose contra *C. maculatus* e descendem de genitores resistentes, o que lhes confere potencial para serem utilizados em futuros cruzamentos, para obtenção de variedades de feijão-caupi com maior nível de resistência.

Termos para indexação: *Vigna unguiculata*, Bruchinae, Chrysomelidae, resistência de plantas, armazenamento.

Introduction

Cowpea [*Vigna unguiculata* (L.) Walp.] is a major crop in the North and Northeastern regions of Brazil. This legume has great economic and nutritional importance. Besides its low production costs, due to its short cycle, low water requirement, and ability to survive in inhospitable conditions, such as low fertility soils, it is a key source of proteins and carbohydrates, with a high content in fibers, vitamins, and minerals, as well as a low lipid content (Singh et al., 2002; Freire Filho et al., 2005).

Insect pests that harm the various stages of crop development in the field and that damage stored grains are prominent problems related to cowpea cultivation.

The cowpea weevil *Callosobruchus maculatus* (Fabr.) (Coleoptera: Chrysomelida, Bruchinae) is considered the most important pest that occurs during the storage period. The attack, which starts before harvest and intensifies during storage, may cause total losses (Faroni & Sousa, 2006).

The damage by *C. maculatus* is caused by oviposition on the surface of grains and subsequent larval penetration in the grains. The attack results in weight loss, decreased retail and nutritional value, reduced level of product hygiene (presence of droppings, eggs, and insects), and reduced seed germination rate (Almeida et al., 2005; Faroni & Sousa, 2006).

Chemical control by fumigation has been a common practice in grain disinfestations. However,

several insect pests have developed resistance to many active ingredients of synthetic products due to failure to comply with usage recommendations, underdosing, and disrespect for the residual period of the insecticides. Furthermore, storage conditions available to most farmers enable re-infestation, increasing the frequency of insecticide use (Almeida et al., 2006).

Conversely, the growing concern about the possible harmful effects of pesticides, such as toxicity to applicators, environmental pollution, and the presence of residues in food, has encouraged investigations on alternative pest-control strategies, including the use of genetically resistant cultivars (Lara, 1991; Panda & Khush, 1995). This is vital considering the growing expansion of cowpea in the Central-West region of Brazil for exportation to other countries, such as the United States (Freire Filho, 2011), and given the fact that there are no registered products at the Ministry of Agriculture, Livestock and Food Supply for the control of *C. maculatus* in cowpea (Brasil, 2013).

The use of resistant genotypes is a promising strategy for the management of *C. maculatus* for the following reasons: it can maintain the population of *C. maculatus* below the economic damage threshold without causing disturbance or pollution to the environment; it does not require specific knowledge by the farmer; it has a low cost; and it is compatible with other means of control, being in accordance with the integrated pest management philosophy (Smith, 2005; Vendramim & Guzzo, 2009). Some Brazilian studies based on oviposition preference tests (Costa & Boiça Júnior, 2004) and antibiosis tests (Costa & Boiça Júnior, 2004; Carvalho et al., 2011) have shown promising results in obtaining cowpea genotypes resistant to *C. maculatus*. However, these studies have only tested a small number of genotypes, and further research is needed to assess a larger variety.

The objective of this work was to characterize the resistance of 50 cowpea (*Vigna unguiculata*) genotypes to *C. maculatus*.

Materials and Methods

The tests were conducted at Laboratório de Resistência de Plantas a Insetos e Plantas Inseticidas

from Universidade Estadual Paulista, state of São Paulo, Brazil, from 2010 to 2011. A completely randomized design with five replicates per treatment (genotype) was used. The lines and cultivars used were obtained from the Embrapa Meio-Norte program for the genetic breeding of cowpea. Several of the selected genotypes had been genetically modified for commercial characteristics and resistance to multiple viruses (Table 1).

An initial population of *C. maculatus* was provided through insect breeding in the entomology laboratory of Embrapa Meio-Norte. Clear glass jars with a 500 mL capacity were used for population multiplication. The jars were closed at the top with a screw cap, where a round hole was made and a 30 mesh nylon screen was adapted to enable internal airing. Each jar was filled with 300 g of cowpea grains (cultivar BRS Guariba) and maintained under $25\pm 2^{\circ}\text{C}$, relative humidity of $60\pm 10\%$, and photoperiod of 12 hours, in a biochemical oxygen demand (BOD) incubator. The grains contained in these jars were sieved every 28 days, and the newly emerged adults were used in the infestation of the new jars.

After harvesting and processing, the grains were placed in plastic bags and kept in cold storage ($2-3^{\circ}\text{C}$) to control potential infestations brought from the field. Prior to performing the tests, the grains were removed from the cold chamber, placed in plastic containers, and stored inside the BOD incubator for ten days to achieve hygroscopic equilibrium.

Oviposition and other biological aspects of the insect were evaluated through no-choice tests in the different genotypes. Based on the method of Costa & Boiça Júnior (2004), five weevils (unsexed adults) with a maximum age of 48 hours were kept in round plastic containers (6 cm diameter by 1 cm height), with 10 g of each genotype, for five days, in a BOD incubator, to evaluate oviposition. After that, the insects were removed, and the containers with grains were returned to the same incubator. To avoid crushing the eggs during manipulation and to insure hatching of the larvae, oviposition was quantified 15 days following the initial infestation. This was done by counting the number of viable (white, opaque color) and unviable (hyaline color) eggs per genotype using a stereoscopic microscope

Table 1. List of the 50 cowpea (*Vigna unguiculata*) genotypes and their respective genealogies used in the assays for resistance against *Callosobruchus maculatus*.

Genotype	Genealogy/Origin
BR 14-Mulato ⁽¹⁾	CNC0434 x CNCx27-2E
BR 17-Gurguéia ⁽¹⁾	BR 10-Piauí x CE-315
BR 3-Tracueteua ⁽¹⁾	Selected from the local cultivar Quebra cadeira or Cheque ouro originated from Northeastern Brazil
BRS Cauamé ⁽¹⁾	TE93-210-13F x TE96-282-22G
BRS Milênio ⁽¹⁾	Selected from cultivar BR 3-Tracueteua
BRS Novaera ⁽¹⁾	TE97-404-1F x TE9-404-3F
BRS Pajeú ⁽¹⁾	CNCx405-17F x TE94-268-3D
BRS Paraguacu ⁽¹⁾	BR 10-Piauí x Aparecido Moita
BRS Potengi ⁽¹⁾	TE96-282-22E x TE93-210-13F
BRS Rouxino ⁽¹⁾	TE86-75-57E x TEx1-69E
BRS Tumucumaque ⁽¹⁾	TE96-282-22G x IT87 D-611-3
BRS Urubuquara ⁽¹⁾	Selected from cultivar BR 3 (Tracueteua)
BRS Marataoã ⁽¹⁾	Seridó x TVx 1836-013J
Canapuzinho ⁽¹⁾	Local cultivar São Raimundo Nonato, Piauí, Brazil
Canapuzinho-1-2 ⁽¹⁾	Selected from the cultivar Canapuzinho – São Raimundo Nonato, Piauí, Brazil
Capela ⁽¹⁾	Local cultivar Capela do Alto, São Paulo, Brazil
Corujinha ⁽¹⁾	Local cultivar Barbalha, Ceará, Brazil
Epace 10 ⁽¹⁾	Seridó x TVu 1888
Inhuma ⁽¹⁾	Selected from the local cultivar Inhuma – Inhuma, Piauí, Brazil
Monteiro ⁽¹⁾	Selected from the local cultivar Monteiro – Piripiri, Piauí, Brazil
Patativa ⁽¹⁾	CNC1735 x (CNCx926-4F x Paulista)
Paulistinha ⁽¹⁾	Local cultivar Barbalha, Ceará, Brazil
Pingo-de-ouro-1-1 ⁽¹⁾	Selected from the local cultivar Pingo-de-ouro – Iguatu, Ceará, Brazil
Poços de Caldas, MG ⁽¹⁾	Selected from the cultivar introduced through undefined screening from Peru, most likely the cultivar Vainablanca
Sanzi Sambili ⁽¹⁾	International Institute of Tropical Agriculture (IITA), Ibadan, Nigeria
Vainablanca ⁽¹⁾	Improved cultivar from Peru
IT81 D-1045 Enramador ⁽²⁾	International Institute of Tropical Agriculture (IITA), Ibadan, Nigeria
IT81 D-1045 Ereto ⁽²⁾	International Institute of Tropical Agriculture (IITA), Ibadan, Nigeria
IT82 D-889 ⁽²⁾	International Institute of Tropical Agriculture (IITA), Ibadan, Nigeria
IT85 F-2687 ⁽²⁾	International Institute of Tropical Agriculture (IITA), Ibadan, Nigeria
IT86 D-716-1 ⁽²⁾	International Institute of Tropical Agriculture (IITA), Ibadan, Nigeria
MN05-841 B-49 ⁽²⁾	MNC00-599 F-9 x MNC99-537 F-14-2
MNC04-786 B-87-2 ⁽²⁾	MNC01-625E-10-1-2 5 x MNC99-554 D-10-1-2-2
MNC99-505 G-11 ⁽²⁾	Canapuzinho x BR 17-Gurguéia
MNC99-507 G-4 ⁽²⁾	BR 14-Mulato x Canapuzinho
MNC99-507 G-8 ⁽²⁾	BR 14-Mulato x Canapuzinho
MNC99-508-1 ⁽²⁾	TE90 180 88F x Canapuzinho
MNC99-510 G-16 ⁽²⁾	Paulista x TE90-180-88F
MNC99-541 F-15 ⁽²⁾	TE93-210-13F x TE96-282-22G
MNC99-541 F-21 ⁽²⁾	TE93-210-13F x TE96-282-22G
TE93-244-23 F-1 ⁽²⁾	IPA 206 x TE86-73-3G
TE94-309 G-9 ⁽²⁾	CNCx405-24F x CNCx698-128G
TE97-299 G-24 ⁽²⁾	CNCx405-17F x CNCx698-128G
TE97-304 G-4 ⁽²⁾	CNCx405-17F x TE94-268-3D
TE97-309 G-18 ⁽²⁾	CNCx405-24F x CNCx698-128G
TE97-309 G-24 ⁽²⁾	CNCx405-24F x CNCx698-128G
TVu 1593 ⁽²⁾	International Institute of Tropical Agriculture (IITA), Ibadan, Nigeria
TVu 36 ⁽²⁾	International Institute of Tropical Agriculture (IITA), Ibadan, Nigeria
TVu 382 ⁽²⁾	International Institute of Tropical Agriculture (IITA), Ibadan, Nigeria

⁽¹⁾Cultivar. ⁽²⁾Line. CNC, Centro Nacional Caupi; MNC, Meio Norte Caupi.

(20 x magnification). The plots were returned to the BOD incubator until the emergence of the adult insects.

The plots were evaluated daily by sieving the grains of each container through an appropriate mesh and counting the number of insects emerging per day and per genotype, in order to assess the total number of insects emerged and the development period (egg to adult) at 25 days after the initial infestation. After counting, the emerged adults were placed in glass vials (2 cm diameter by 5 cm height) with rubber stoppers, immediately placed in a freezer for rapid death (avoiding weight loss), and maintained under conservation conditions.

Once the emergences ended (i.e., five consecutive days without emergence), the vials containing the emerged insects were opened and placed in an oven (40°C) for two days; after this period, the dry weight (mg) of the emerged adults was assessed using a precision scale (0.0001 g). The weighted mean – calculated by the formula $\sum xf/\sum x$, in which: x is the number of adults emerging that day and f is the number of emergence days – was used to determine the egg to adult development period (Costa & Boiça Júnior, 2004). The dry mass (g) of grains consumed by the larvae of all evaluated plots was calculated as the difference between the dry mass of uninfested and infested aliquots, divided by the number of adults emerging from each replicate of the corresponding treatment.

The obtained data were subjected to analysis of variance using the F test. The normality and homogeneity of the data were assessed with the Kolmogorov-Smirnov and Levene tests, respectively. Whenever the F test was significant, means were compared by the Scott & Knott test, at 5% probability, using the statistical software Sisvar, version 5.0 (Lavras, MG, Brazil).

Results and Discussion

No significant difference was observed between the genotypes as to the total number of eggs (Table 2). Regarding the percentage of viable eggs, the genotypes Sanzi Sambili, MN05-841 B-49, MNC99-508-1, and Canapuzinho-1-2 had the lowest means, whereas the genotypes MNC99-510 G-16, BRS Novaera, IT82 D-889, Monteiro, IT81

D-1045 Ereto, BRS-Marataoã, BR 17-Gurguéia, BR 3-Tracuateua, BR 14-Mulato, BRS Milênio, and BRS Urubuquara showed the highest percentages.

Seed physical characteristics, such as color, texture, size and hardness, are associated with the resistance of some varieties of cowpea to *C. maculatus*, as indicated by decreased oviposition (Gbaye & Holloway, 2011). However, most studies have only recorded the type of resistance and not investigated its causes (Costa & Boiça Júnior, 2004; Carvalho et al., 2011; Marsaro Junior & Vilarinho, 2011).

In the present study, a great diversity was observed in grain color (red, green, brown, white, and black tegument) and size (small, medium, and large) among the 50 tested genotypes, but there was little diversity in texture (only one genotype had rough seed coats). Grain color, size, and texture did not interfere in oviposition preference since no significant differences were found among genotypes or in egg viability: the genotypes with the lowest and highest egg viability shared common physical characteristics. The low egg viability may have been due to biochemical factors of the seed coat and the inside of the grain, independently of grain size or texture.

The genotypes Sanzi Sambili, Canapuzinho-1-2, MN05-841 B-49, and MNC99-508-1 presented reductions in the number of adult emergence, which was expected due to the low egg viability. However, it should be highlighted that IT81 D-1045 Ereto, IT81 D-1045 Enramador, and IT85 F-2687, even with high percentages of viable eggs, had decreased number of emergences (less than 35 individuals). The remaining genotypes displayed a high emergence number, which did not differ significantly between the genotypes (Table 2). The IT81 D-1045 genotype descends from TVu 2027, which was selected from the germplasm bank of the International Institute of Tropical Agriculture, used by improvement programs to obtain resistant varieties. The resistance of these genotypes has been associated with variant forms of the reserve protein, vicilin, which cannot be metabolized by the insect's midgut proteinases, limiting the larvae's food supply and interfering with the development of *C. maculatus* (Domingues et al., 2006). This may have been the case in the present study in

Table 2. Mean±standard error values of total number of eggs, egg viability, number of emerged adults, and viability of the immature phase of *Callosobruchus maculatus* in different cowpea (*Vigna unguiculata*) genotypes⁽¹⁾.

Genotype	Total number of eggs ^{ns}	Egg viability (%)	Number of emerged adults	Viability of immature phase (%)
IT85 F-2687	41.33±10.89	84.77±9.08d	34.00±8.08a	96.77±3.23e
Monteiro	76.60±13.99	91.75±1.31e	64.60±11.99a	92.13±2.16e
MNC04-786 B-87-2	82.00±15.09	79.75±1.88d	53.00±10.16a	82.58±3.37d
MNC99-510 G-16	82.00±16.24	89.39±3.99e	66.80±13.41a	91.69±3.00e
BRS Milênio	89.80±16.56	94.39±1.74e	81.00±14.45b	96.47±4.95e
TVu 36	90.40±17.29	70.00±2.89c	46.20±9.77a	72.75±2.00c
MNC99-541 F-15	93.00±21.16	81.02±1.65d	71.20±15.49a	94.94±3.40e
BRS Xiquexique	93.60±09.10	70.09±4.09c	58.60±8.35a	87.65±4.46d
Paulistinha	97.00±11.82	73.79±2.60c	60.00±7.82a	84.00±2.26d
BRS Rouxinol	97.40±19.37	87.95±2.35d	78.20±17.47b	89.04±2.10e
MNC99-508-1	97.80±24.15	57.96±4.54a	44.40±13.76a	72.12±1.28c
BRS Pajeu	100.80±20.18	85.82±3.61d	74.60±16.71b	84.68±2.25d
BRS Cauamé	101.40±24.59	84.41±3.99d	84.00±24.02b	96.15±2.72e
TE97-304 G-4	102.20±16.37	73.16±1.75c	67.20±11.12a	90.11±3.25e
MNC99-507 G-4	102.40±22.16	75.20±4.38c	53.40±11.08a	71.19±1.09c
BR 3-Tracueteua	102.60±18.69	95.04±1.30e	79.00±13.72b	82.73±1.63d
TE97-309 G-18	102.80±36.30	82.09±5.57d	75.40±27.55b	94.29±3.20e
TVu 1593	103.20±11.11	62.04±1.39b	43.80±5.88a	67.99±2.41c
BRS Tumucumaque	103.40±15.80	65.26±5.11b	60.20±10.09a	88.65±3.04d
Canapuzinho	105.60±14.40	87.73±3.36d	79.20±11.78b	85.14±3.12d
IT86 D-716-1	106.20±26.56	69.65±1.36c	64.40±15.30a	88.50±2.54d
MNC99-541 F-21	106.80±12.08	68.59±1.68c	65.60±6.06a	90.33±1.50e
BRS Marataoã	107.80±20.31	91.33±1.00e	84.00±16.05b	84.68±2.34d
MNC99-510-8	111.40±43.29	64.78±3.43b	48.60±17.44a	69.73±1.10c
TE97-299 G-24	114.00±32.66	88.19±1.28d	83.00±23.39b	83.24±0.91d
Epace 10	114.20±28.50	84.86±1.86d	88.80±22.12b	91.62±2.97e
TE97-309 G-24	115.20±26.31	87.76±1.22d	90.40±19.90b	89.75±4.75e
BRS Potengi	116.40±28.18	66.33±7.68b	72.00±19.44a	86.40±1.74d
Pingo-de-ouro-1-1	116.60±23.30	69.74±1.68c	59.60±13.48a	72.45±0.79c
BRS Novaera	116.60±22.66	89.04±2.60e	95.00±20.65b	89.17±1.11e
IT81 D-1045 Enramador	117.20±17.87	74.60±3.36c	17.60±4.73a	18.63±11.21a
TVu 382	120.00±21.14	81.37±3.24d	85.00±16.50b	84.49±4.39d
Vainablanca	120.40±31.65	76.80±2.32c	55.80±25.93a	49.71±1.78b
BR 17-Gurguéia	120.40±33.88	95.63±2.13e	102.40±27.83b	92.30±1.64e
Patativa	122.80±26.13	84.85±1.58d	92.00±18.70b	89.87±2.19e
MNC99-507 G-8	123.00±32.37	86.76±3.76d	94.80±26.40b	87.39±2.26d
Sanzi Sambili	124.20±26.81	56.38±3.34a	54.00±12.42a	77.30±1.87c
Poços de Caldas, MG	126.80±19.84	74.42±3.10c	74.60±10.81b	79.73±1.74c
Canapuzinho-1-2	128.20±21.11	58.53±2.51a	52.20±9.91a	68.58±1.26c
IT81 D-1045 Ereto	128.20±33.20	90.34±0.81e	15.60±3.80a	13.68±1.27a
Corujinha	128.40±06.00	64.77±3.10b	61.00±4.95a	73.16±2.94c
TE94-309 G 9	130.60±14.67	76.95±3.96c	85.60±8.13b	86.26±1.85d
MNC99-505 G-11	131.80±10.34	78.93±3.81d	85.40±8.11b	82.40±3.67d
BR 14-Mulato	132.80±36.69	95.91±1.95e	114.60±30.05b	92.34±3.34e
MN05-841 B-49	137.60±11.31	56.88±2.54a	58.20±6.77a	74.13±1.21c
Inhuma	141.20±32.75	73.34±3.57c	75.40±17.11b	74.74±2.63c
BRS Urubuquara	150.60±20.48	96.85±0.86e	135.20±16.77b	93.47±1.63e
IT82 D-889	153.00±17.81	89.97±1.48e	118.40±12.97b	86.35±1.06d
Capela	175.20±19.28	86.69±0.92d	128.60±16.33b	84.02±1.61d
BRS Paraguaçu	177.80±33.28	68.91±3.12c	106.20±24.50b	84.43±2.67d
P	0.6102	<0.001	<0.001	<0.001
CV (%)	45.77	8.82	49.73	8.24

⁽¹⁾Means followed by equal letters do not differ by the Scott & Knott test, at 5% probability. ^{ns}Nonsignificant. Temperature, 25±2°C; relative humidity, 60±10%; photoperiod, 12:12 hours.

which both IT81 D-1045 Ereto and IT81 D-1045 Enramador were detrimental to the development of *C. maculatus*, indicating antibiosis-type resistance.

A possible explanation for the low emergence registered for IT85 F-2687 (Table 2), despite the high percentage of viable eggs, is the fact that this genotype has lower oviposition than the other ones evaluated (although the differences observed were not significant). It should also be noted that IT85 F-2687 was the only one among the tested genotypes with a rough seed coat.

Since the viability of the immature phase directly reflects on the emergence of adults, the lowest percentages recorded in IT81 D-1045 Ereto and IT81 D-1045 Enramador (Table 2) suggest detrimental effects of both of these genotypes for the development of *C. maculatus*. In contrast, BRS Rouxinol, TE97-309 G-24, BRS Novaera, Patativa, IT85 F-2687, Monteiro, MNC99-510 G-16, BRS Milênio, MNC99-541 F-15, BRS Cauamé, TE97-304 G-4, TE97-309 G-18, MNC99-541 F-21, Epace 10, BR 17-Gurguéia, BR 14-Mulato, and BRS Urubuquara exhibited percentages above 90%. The fact that the egg viability was high in IT85 F-2687 also indicates that the low emergence observed in this genotype did not result from larvae mortality inside the grain.

The genotypes with the highest means for the development period (from egg to adult) were IT81 D-1045 Enramador and IT81 D-1045 Ereto (Table 3), which differed from the other genotypes. In comparison, TE97-309 G-18 and TVu 382 showed the lowest means for the development period, which favored the insect biology. These results are indicative of the expression of antibiosis-type resistance in IT81 D-1045 Enramador and IT81 D-1045 Ereto. The viability and duration of the immature phase are considered important parameters for the characterization of antibiosis to other grain weevils such as *Acanthoscelides obtectus* Say (Baldin & Lara, 2008) and *Zabrotes subfasciatus* Boh. (Baldin & Pereira, 2010). Moreover, antibiosis-type resistance is characterized by an increased span of time between the egg and adult phases, as well as by the reduction in adult emergence (Costa & Boiça Júnior, 2004; Smith & Clement, 2012). Both of these characteristics were observed in the present study. According to Smith

& Clement (2012), the ability of a resistant host to delay the development of pests results in decreased reproduction rates or number of insects in natural populations due to the increased average time of each generation.

The genotypes with the lowest intake per insect were IT85 F-2687, MNC-99-510-8, BRS Potengi, and BRS Xiquexique, whereas IT81 D-1045 Ereto and IT81 D-1045 Enramador showed the highest mean values of intake per insect (Table 3). The genotypes with lower intake per insect had higher numbers of emerged adults than the ones with higher intake, i.e., lower intake per insect was observed when the viability of the larvae was higher, which may also be related with larvae competition inside the susceptible grains. Another possible explanation for the low intake might be a non-preference-type feeding expression, usually associated with the presence of anti-nutritional or unpalatable compounds, requiring a longer insect feeding period to complete the immature stage (Smith & Clement, 2012). The higher intakes in IT81 D-1045 Ereto and IT81 D-1045 Enramador confirm antibiosis-type resistance, since the insects feed normally with this resistance mechanism.

No significant differences were observed in insect weight among the genotypes (Table 3). The insects that fed normally did not gain more weight than the ones with low intake. Domingues et al. (2006) studied the survival of *C. maculatus* larvae when grown on IT81 D-1045 resistant seeds and found that the larvae feeding on resistant cowpea embryos developed better than the ones feeding on the cotyledons. Using computed axial scanning (tomography), Tarver et al. (2006) concluded that *C. maculatus* feeds differently when grown on resistant or susceptible cowpea seeds, and that there is a region in the core of the resistant seeds that has a negative effect on the insect, which avoids this area. This may have been the case in the present study, in which larvae feeding on IT81 D-1045 Ereto and IT81 D-1045 Enramador were not affected in terms of intake, but took more time to develop, being affected in terms of weight gain, possibly due to the ingestion of a detrimental chemical substance, even if in small quantities. The lack of differences in insect weight may also be related to insect metabolism.

Table 3. Mean±standard error values for development period (egg to adult), intake, and dry weight per *Callosobruchus maculatus* insect in different cowpea (*Vigna unguiculata*) genotypes⁽¹⁾.

Genotype	Development period (days)	Intake per insect (g)	Dry weight per insect (mg)
IT81 D-1045 Enramador	37.04±0.45g	0.024±0.005d	1.85±0.17
IT81 D-1045 Ereto	35.01±1.00g	0.035±0.005e	1.77±0.03
BRS-Marataoã	31.08±0.37f	0.011±0.001a	1.89±0.08
BR 3-Tracuateua	30.31±0.05e	0.014±0.001a	1.96±0.04
MNC99-507 G-4	30.26±0.30e	0.014±0.001a	1.95±0.03
IT82 D-889	30.10±0.20e	0.012±0.001a	1.93±0.06
MNC99-507 G-8	29.96±0.19e	0.013±0.001a	1.99±0.04
BRS Cauamé	29.90±0.16e	0.015±0.001b	1.98±0.03
BRS Novaera	29.74±0.22e	0.014±0.001b	1.86±0.04
Inhuma	29.57±0.27d	0.018±0.001c	2.33±0.43
MNC99-541 F-15	28.58±0.20d	0.011±0.001a	2.00±0.05
BR 14-Mulato	29.54±0.21d	0.013±0.001a	1.75±0.03
Poços de Caldas, MG	29.51±0.27d	0.012±0.001a	1.85±0.02
Canapuzinho-1-2	29.45±0.26d	0.015±0.001b	2.04±0.08
Monteiro	29.44±0.29d	0.014±0.001a	1.91±0.02
Canapuzinho	29.24±0.30d	0.015±0.001b	1.95±0.04
MNC99-508-1	29.20±0.28d	0.011±0.001a	1.98±0.06
BR 17-Gurguéia	29.14±0.37d	0.011±0.001a	2.06±0.21
MN05-841 B-49	29.04±0.44d	0.013±0.001a	1.82±0.02
IT85 F-2687	28.96±0.41d	0.010±0.001a	2.03±0.66
MNC99-505 G-11	28.84±0.27d	0.015±0.001b	2.00±0.05
BRS Paraguaçu	28.80±0.18d	0.012±0.001a	1.94±0.08
BRS Pajeu	28.80±0.27d	0.011±0.001a	2.00±0.08
IT86 D-716 1	28.75±0.11d	0.013±0.001a	1.93±0.07
Corujinha	28.67±0.12d	0.012±0.001a	1.94±0.05
Epace 10	28.50±0.32c	0.015±0.001b	1.93±0.05
TE97-299 G-24	28.43±0.19c	0.015±0.001b	2.01±0.06
MNC-99-510-8	28.42±0.27c	0.010±0.002a	2.04±0.04
TE94-309 G-9	28.37±0.15c	0.020±0.001c	2.03±0.02
BRS Milênio	28.26±0.19c	0.013±0.001a	1.82±0.02
TVu 1593	28.22±0.43c	0.020±0.002c	2.09±0.05
MNC99-541 F-21	28.16±0.21c	0.015±0.001 b	1.94±0.04
Pingo-de-ouro-1-1	28.07±0.20c	0.016±0.001b	1.80±0.06
MNC04-786 B-87-2	28.06±0.21c	0.018±0.001c	1.93±0.06
BRS Rouxinol	28.04±0.12c	0.014±0.001a	1.86±0.01
BRS Potengi	28.04±0.42c	0.010±0.002a	1.84±0.07
BRS Urubuquara	27.98±0.28c	0.013±0.001a	1.79±0.02
Capela	27.94±0.20c	0.013±0.001a	1.82±0.03
Patativa	27.76±0.16c	0.015±0.001b	1.93±0.04
TE97-304 G-4	27.75±0.12c	0.017±0.001c	2.02±0.06
Paulistinha	27.60±0.19c	0.017±0.001c	1.95±0.01
Sanzi Sambili	27.57±0.29c	0.018±0.003c	1.77±0.09
MNC99-510 G-16	27.54±0.38c	0.016±0.001b	1.95±0.05
BRS Xiquexique	27.41±0.20c	0.010±0.001a	1.95±0.05
Vainablanca	27.09±0.38b	0.008±0.001a	1.60±0.11
BRS Tumucumaque	27.02±0.08b	0.011±0.001a	1.99±0.04
TVu 36	26.97±0.34b	0.012±0.001a	1.83±0.05
TE97-309 G-24	26.73±0.19b	0.015±0.001b	1.85±0.04
TVu 382	26.16±0.28a	0.013±0.001a	1.79±0.04
TE97-309 G-18	25.57±0.52a	0.015±0.001b	1.96±0.12
P	<0.001	<0.001	0.2128
CV (%)	2.37	21.82	11.99

⁽¹⁾Means followed by equal letters do not differ by the Scott & Knott test, at 5% probability. ^{ns}Nonsignificant. Temperature, 25±2°C; relative humidity, 60±10%; photoperiod, 12:12 hours.

Conclusions

1. The genotypes IT85 F-2687, MN05-841 B-49, MNC99-508-1, MNC99-510-8, TVu 1593, Canapuzinho-1-2, and Sanzi Sambili exhibit non-preference-type resistance to the oviposition and feeding of *Callosobruchus maculatus*.

2. The genotypes IT81 D-1045 Ereto and IT81 D-1045 Enramador exhibit antibiosis-type resistance to *C. maculatus*.

3. The IT81 D-1045 Ereto and IT81 D-1045 Enramador lines descend from resistant progeny and have potential to be used in future crossings to obtain varieties with higher levels of resistance.

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