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Screening Cowpea Genotypes for Resistance to Cowpea Aphid Borne Mosaic Virus (CABMV) and Cowpea Severe Mosaic Virus (CPSMV) in Paraguay

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ABSTRACT

Background: Cowpea [*Vigna unguiculata* (L) Walp.] is one of the Paraguayan rural families' main crops, serving as an essential protein and carbohydrate source. Cowpea aphid borne mosaic virus (CABMV) and cowpea severe mosaic virus (CPSMV) were identified infecting cowpea plants. Disease control caused by both viruses is difficult because there is no information about local cowpea resistant cultivars and vector control is not practical.

Methods: In the present work, sixteen cowpea genotypes/cultivars were mechanically inoculated with local isolates of CABMV and CPSMV to identify resistant genotypes/cultivars that can be used in breeding programs. Virus infections were determined by symptoms expression and confirmed by PTA-ELISA.

Result: Genotypes Arroz rojo (*V. angularis*), TVu 379, TVu 382, TE94-256-2E and TE97-309G-9 were resistant to CABMV. Genotypes Arroz rojo (*V. angularis*), CNCX-698-128F, TVu 379, TVu 382, TVu-3961, TE97-309G-9 and TE97-309G-3 were resistant to CPSMV. Overall, this study showed that local cowpea cultivars do not offer any resistance to virus infection and the need for resistant germplasms for cowpea breeding programs in the country.

Key words: Comovirus, Cowpea, Potyvirus, Resistance.

INTRODUCTION

Cowpea [*Vigna unguiculata* (L.) Walp.] seeds pose tremendous nutritional value for its high protein and carbohydrate content (Lambot, 2002). It is of extreme importance for Paraguayan rural families and is cultivated throughout the country on approximately 72,000 ha annually, involving about 230,000 small growers (MAG/DCEA 2017).

Diseases and pests affect cowpea production resulting in severe yield losses. In Latin America, the main viruses that infect cowpea crops are cucumber mosaic virus (CMV) (genus *Cucumovirus*, family *Bromoviridae*); cowpea aphid-borne mosaic virus (CABMV) (genus *Potyviridae*, family *Potyvirus*); cowpea severe mosaic virus (CPSMV) (genus *Comovirus*, family *Comoviridae*) and cowpea golden mosaic virus (CPGMV) (genus *Begomovirus*, family *Geminiviridae*) (Lima *et al*., 2005; Pio-Ribeiro and Assis-Filho, 1997; Barreto, 1999). In Paraguay, only CABMV and CPSMV are present in cowpea crops (González-Segnana *et al*., 2013; Delgado *et al*., 2014).

CABMV was first described in Paraguay infecting sesame plants (*Sesamum indicum*) (González-Segnana *et al*., 2011). In addition to cowpea and sesame, weeds and cover crops have been found infected with CABMV and its presence has been confirmed in at least three departments of the country (González-Segnana *et al*., 2013). Natural transmission occurs in a nonpersistent manner by various aphid species (Bashir *et al*., 2002). *Aphid crassivora* proved to be vectors of CABMV in Paraguay (Natalia de Jesús *et al*., 2013).

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CPSMV was first detected in Paraguay in seed-borne infected cowpea seedling from field symptomatic cowpea plants (Delgado *et al*., 2014). Natural transmission occurs in a semi-persistent manner by some Chrysomelidae beetles. *Cerotoma arcuata* was reported as the most important vector in Brazil (Costa *et al*., 1978; Lima *et al*., 2005).

Planting resistant cultivars is the most economical, effective and practicable method to minimize yield losses

caused by CABMV and CPSMV (Taiwo, 2003). Plant resistance against viral diseases begun with the identification of an appropriate resistance source. Since biological properties of CABMV and CPSMV may differ among isolates worldwide (Bashir *et al*., 2002; Hampton *et al*., 1997) it becomes imperative to identify resistance sources against the local prevalent viral isolates.

This work aimed to evaluate the response of some local and introduced cowpea cultivars/genotypes to infection with local isolates of CABMV and CPSMV to identify resistant material to be used in further breeding programs.

MATERIALS AND METHODS

Virus isolates

CABMV and CPSMV isolates collected from field-infected cowpea plants in the region of Choré, San Pedro Department, Paraguay, were used for the experiments. Virus isolates were separately maintained on cowpea plants through the periodical mechanical transmission. Plants were kept in a greenhouse at the Department of Biology FCA/UNA, San Lorenzo, Central department, Paraguay. Virus presence in source plants was confirmed by partial nucleotide sequencing of each virus target gene amplified by RT-PCR. Total RNA was extracted from cowpea sources plants using Trizol and analyzed by RT-PCR using potyvirus universal primers (forward: 5′ -GATTTAGGTGA CACTATAGT17-3′, reverse: 5′ -ATGGTTTGGTGYATY GARAAT-3′) (Mackenzie *et al*., 1998; Maciel *et al*., 2011) and comovirus universal primers (forward: 5′-GCATGGTCCACWCAGGT-3′, reverse: 5′-YTCRAAWCCVYTRTTKGGMCCACA-3′) (Beserra *et al*., 2011). Obtained amplicons were purified with Wizard® SV Gel and PCR Clean-Up System (Promega) and sent for direct nucleotide sequencing at Macrogen (Seoul, South Korea). Nucleotide sequence of local CABMV isolate shared 95.56%-95.70% identity with CABMV isolates available in the GenBank (accession no. MK472693- KM597165). CPSMV isolate from Paraguay was partially sequenced for the first time and two nucleotide sequences were deposited in the GenBank (accessions no. KF793279- KF793280). They shared 93.77%-99.64% identity with corresponding nucleotide sequences of CPSMV isolates from Brazil (accessions no. GQ229416- AF263549). Molecular detection was carried out at ESALQ/USP.

Samples of CPSMV source plants were analyzed by transmission electron microscopy (TEM) to complement the identification of CPSMV, since the only available evidence of its presence in the country was done by serologic detection (Delgado *et al*., 2014). Examinations were made in a Zeiss EM 900 or JEOL 1011 transmission electron microscopes at ESALQ/USP and images were recorded digitally. TEM analysis of thin sections of symptomatic cowpea leaf tissue confirmed the presence of a high concentration of CPSMV isometric virus particles ca. 25 nm in diameter, often forming a crystalline array (Fig 1).

Evaluation of cowpea resistance to CABMV and CPSMV infection

For evaluation of the response of cowpea cultivars/genotypes to infection with each virus, independent experiments were conducted in 2018 and 2019, respectively. The seed of the following cowpea cultivars and/or genotypes were separately sown in 1.5 L plastic pots containing substrate; local cultivars: Arroz Rojo (*Vigna angularis*), Arroz Negro, Blanco Ojo Negro, Choré, Crema IAN, Ñu, Pyta, San Francisco; introduced genotypes: CNCX-698-128F TVu-379, TVu-382, TVu-3961, TE94-256-2E, TE94-200-49F, TE97-309G-9 and TE97-309G-3. After germination, seedlings were transplanted to 1.5 L plastic pots containing a mixture of soil, sand and sterile organic matter.

Mechanical transmissions were performed using extracts from each viral source plants prepared by grinding symptomatic leaves in a mortar in the presence of 0.01 M phosphate buffer (pH 7.0) with 0.1% sodium sulfite, diluted 1:5 (w/v). The extracts were then applied to 10 plants of each cultivar, at 4-6 leaf stage. The inoculum was manually rubbed on the surface of two leaves previously dusted with carborundum. Ten plants per cultivar were mock inoculated as controls. Plants were kept in the greenhouse of the Dept. of Biology FCA/UNA, for further development. Eight weeks later, all plants were analyzed by symptoms expression and PTA-ELISA for virus detection. The experiment was repeated twice.

Virus detection

PTA-ELISA was performed to detect each virus in newly emerged leaves of mechanically inoculated test plants as described by Mowat and Dawson (1987). CABMV and CPSMV antisera were diluted 1:1000. CABMV antiserum was produced by one of the authors (JAMR, unpublished data), whereas the other was produced by Dr. M.T. Lin, Universidade de Brasília, DF, Brazil. Alkaline phosphatase conjugated anti-rabbit (Sigma) diluted at the ratio of 1:7000 was used. The reaction was considered positive when the OD405nm reading was at least three times that of the healthy control. A Perlong model DNM 9602 ELISA reader was used. The immunoassays were carried out at FCA/UNA.

RESULTS AND DISCUSSION

The reaction of plants of eight local and eight introduced cowpea cultivar/genotypes to infection with CABMV and CPSMV throughout mechanical inoculation is shown in Table 1.

Plants of the introduced cowpea genotypes TVu 379, TVu 382, TE94-256-2E and TE97-309G-9 were resistant to infection with CABMV, whereas CNCX-698-128F, TVu 379, TVu 382, TVu-3961, TE97-309G-9 and TE97-309G-3 were resistant to CPSMV.

CABMV and CPSMV have been accounted as the most widespread viruses on cowpea crops worldwide (Emechebe and Lagoke, 2002; Bashir *et al*., 2002). Several studies for identifying CABMV and CPSMV resistant cowpea genotypes

Table 1: Symptoms presented by sixteen cowpea cultivars subjected to mechanical inoculation with CABMV and CPSMV and their response in PTA-ELISA test.

Genotypes/Cultivars	Origin	CABMV			CPSMV		
		Number of infected plants /inoculated plants	Symptoms	PTA-ELISA	Number of infected plants /inoculated plants	Symptoms	PTA-ELISA
Local							
Arroz Rojo	IPTA (1)	0/10	Ns	-	0/10	Ns	-
Arroz Negro	IPTA	10/10	M	+	10/10	B, M, Fr	+
Blanco Ojo Negro	IPTA	10/10	M	+	10/10	B, SM, Fr, N, Ad	+
Choré	IPTA	10/10	B, SM	+	10/10	B, SM, Fr	+
Crema IAN	IAN (2)	10/10	M	+	10/10	B, SM, Fr	+
Ñu	IPTA	10/10	B, SM, Fr	+	10/10	B, SM, Fr	+
Pyta	IPTA	10/10	B, SM, M, Cl, Fr	+	10/10	B, SM, Fr	+
San Francisco	IPTA	10/10	SM, Fr	+	10/10	B, SM, Cl, Fr	+
Introduced							
CNCX-698-128F	EMEPA (3)	10/10	SM, Fr	+	0/10	Ns	-
TVu-379	IITA (4)	0/10	Ns	-	0/10	Ns	-
TVu-382	IITA	0/10	Ns	-	0/10	Ns	-
TVu-3961	IITA	0/10	SM, Cl	+	0/10	Ns	-
TE94-256-2E	Embrapa (5)	0/10	Ns	-	10/10	B, M, Fr	+
TE94-200-49F	Embrapa	10/10	Ns	+	10/10	B, M, Fr	+
TE97-309G-9	Embrapa	0/10	Ns	-	0/10	Ns	-
TE97-309G-3	Embrapa	10/10	M, Fr	+	0/10	Ns	-

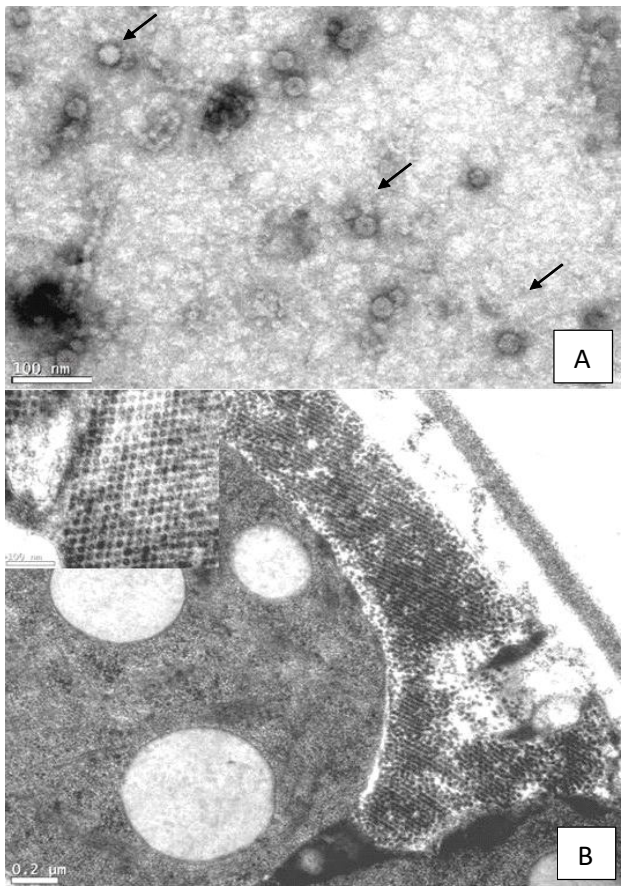
B: Blister; M: Mosaic; SM: Severe mosaic; CL: Chlorotic lesion; Fr: Foliar reduction; N: Necrosis; Ad: Apical death, Ns: No symptoms. (1) Instituto Paraguayo de Tecnología Agrícola (2) Instituto Agronómico Nacional (3) Empresa Estadual de Pesquisa Agropecuária da Paraíba (4) International Institute of Tropical Agriculture. (5) Empresa Brasileña de Investigación Agropecuaria Meio-Norte.

were made and used in cowpea breeding programs (Lima *et al*., 1986; Dhanasekar and Reddy, 2015; Leão *et al*., 2016; Assunção *et al*., 2005). In Brazil, Lima *et al*. (1986) identified four genotypes resistant to both viruses (TVu 379, TVu 382, TVu 966 and TVu 3961), two of those genotypes (TVu 379, TVu 382) evaluated in the present study were resistant to local isolates of CABMV and CPSMV. However, TVu 379, TVu 382 are considered outside the commercial standard, needing a pre-breeding before being included in a breeding program (Oliveira, 2012; Nogueira *et al*., 2011).

Cultivar TVu 382 was used as a donor parent of CPSMV resistance gene in a few works. Crosses segregate in the proportion of 3 susceptible plants: 1 resistant plant and was considered promising for obtaining resistant cowpea cultivars (Lima, 2019; Lima *et al*. 1986; Assunção *et al*. 2005). Similarly, cultivar TE97-309G-9 was also found resistant to each local virus isolates and used as donor parent to identify cowpea-resistant plants by retro-crossing. Segregation of the F2 plants in all crosses was homogeneous, in the proportion of 15 susceptible: 1 resistant (Barros *et al*., 2013).

Plants of CNCx698-128F were susceptible to CABMV in the present study; however, they were resistant to this potyvirus in the study made by Nogueira *et al*. (2011) in Brazil. This difference can be attributed to virus isolates differences since distinct CABMV serotypes/isolates have been reported worldwide and biological properties of CABMV may differ among isolates (Bashir *et al*., 2002; Huguenot *et al*. 1993; Ndiaye *et al*. 1993; Barros *et al*., 2013). The same has been reported for CPSMV, as nine serotypes have already been detected worldwide (Hampton *et al*., 1997). Four serological groups were identified (I, II, III and IV) in Brazil (Lin *et al*., 1981; Lin *et al*., 1984). In this context, genotype TE94-256-2E was susceptible to the local isolate of CPSMV and the serotype I of a Brazilian isolate, as reported by Oliveira *et al*. (2012). However, plants of this genotype were resistant to the Brazilian serotype I of CPSMV (Oliveira *et al*., 2012). The same authors showed that cultivar TE97-309G-9 was susceptible to infection with serotype I. Nevertheless, in the present study, plants of TE97-309G-9 were resistant to infection with the local isolate of CPSMV. No serotype characterization is available for the Paraguayan isolates of CABMV and CPSMV. These results can serve as biological evidence for future serotype characterization.

Local cultivars (Arroz Negro, Blanco Ojo Negro, Choré, Crema IAN, Ñu, Pyta and San Francisco) were highly susceptible to infection with both virus and developed variable symptoms. Plants of cultivar Arroz rojo (*V. angularis*) were resistant to infection with both viruses, as none of the inoculated plants developed symptoms; neither PTA-ELISA detected the viruses in the inoculated plants.



A. Isometric particles present in negatively stained leaf extract;
B. Crystalline array of isometric particles, a detail of which is shown in the insert, in leaf parenchymal cells of symptomatic cowpea.

Fig 1: Transmission electron micrographs of cowpea severe mosaic virus-infected cowpea (*Vigna unguiculata*) leaf sample.

CONCLUSION

The control of viral diseases on cowpea crops worldwide has been achieved using resistant cultivars and hybrids. Since simultaneous infection with both viruses is expected, selecting resistant cowpea cultivars for controlling this double infection is important (Lima *et al*., 1986). In the present work, cowpea genotypes TVu 379, TVu 382 and TE97-309G-9 were resistant to infection with local isolates of CABMV and CPSMV. These genotypes presented asymptomatic plants under mechanical experimental inoculation with both viruses and tested negative for virus detection in PTA-ELISA. Other studies had shown their potential for obtaining new cultivars with multiple resistance to CPSMV and CABMV (Barros *et al*., 2013; Lima *et al*., 2019).

REFERENCES

Assunção, I.P., Resende, L.V., Barros, M., Lima, G.S., Coelho, R.S.B. and Lima, J. (2005). Genes diferentes podem conferir resistência ao cowpea severe mosaic virus em caupi. Fitopatologia Brasileira. 30(3): 274-278.

Barreto, P.D. (1999). Recursos genéticos e programa de melhoramento de feijão-de-corda no Ceará: Avanços e perspectivas. In: Queirós, M.A., Goedert, C.O. and Ramos, S.R.R. (eds) Recursos genéticos e melhoramento de plantas para o Nordeste brasileiro. EMBRAPA, CPATSA, 27 pp.

Barros, G.B., Nogueira, M.S.R., Oliveira, C.R.R., Freire Filho, F.R., Ribeiro, V.Q., Veiga, C.F.M., Brioso, P.S.T. and Eiras, M. (2013). Obtenção de plantas de feijão-caupi resistentes ao cowpea severe mosaic virus e ao Cowpea aphid-borne mosaic virus. Summa Phytopathologica. 39: 130-136.

Bashir, M., Ahmad, Z. and Ghafoor, A. (2002). Cowpea aphid-borne mosaic potyvirus: A review. International Journal of Pest Management. 48(2): 155-168.

Beserra, J.E.A. andrade, E.C. Camarço, R.F.R.A., Nascimento, A.K.Q. and Lima, J.A.A. (2011). Sequence variability in the coat protein gene of cowpea severe mosaic virus isolates from northeastern Brazil. Tropical Plant Pathology. 36: 121-124.

Costa, C.L., Lin, M.T., Kitajima, E.W., Santos, A.A., Mesquita, R.C.M. and Freire, F.R.F. (1978). *Cerotoma arcuata* (Oliv.), um crisomelídeo vector do mosaico da Vigna no Brasil. Fitopatologia. Brasileira. 3: 81-82.

Delgado, G.M.L., Orué, S., Jorge, H. and González Segnana, L. (2014). Identification of viral diseases transmitted by bean [*Vigna unguiculata* (L.) Walp.] seeds. Investigación Agraria. 16(2): 93-98.

Dhanasekar, P. and Reddy, K. (2015). Serological screening of cowpea genotypes for resistance against Cowpea Aphid Borne Mosaic virus using DAS-ELISA. Asian Journal of Plant Pathology. 9: 83-90.

Emechebe, A.M. and Lagoke, S.T.O. (2002). Recent Advances in Research on Cowpea Diseases. In: Challenges and Opportunities for Enhancing Sustainable Cowpea Production, [Fatokun, C.A., Tarawali, S.A., Singh, B.B., Kormawa, P.M. and Tamo, M. (Eds.)]. IITA, Ibadan. 94-123.

González-Segnana, L.G., de Lopez, M. R., Mello, A.P.O.A., Rezende, J.A.M. and Kitajima, E.W. (2011). First Report of Cowpea aphid-borne mosaic virus on Sesame in Paraguay. Plant Disease. 95(5): 613-613.

González-Segnana, L.R., Esquivel Fariña, A., González, D.D., Mello, A.P.O., Rezende, J.A.M. and Kitajima, E.W. (2013). Alternative hosts of cowpea aphid-borne mosaic virus (CABMV) in sesame (*Sesamum indicum*) crops grown in Paraguay. Tropical Plant Pathology. 38(6): 539-542.

Hampton, R.O., Thottappilly, G. and Rossel, H.W. (1997). Viral Diseases of Cowpea and Their Control by Resistance-Conferring Genes. In: Advances in Cowpea Research. International Institute of Tropical Agriculture (UTA) and Japan International Research Center for Agricultural Sciences (JIRCAS). 159-175.

Huguenot, C., Furneaux, M., Thottappilly, G., Rossel, H.W. and Hamilton, R.I. (1993). Evidence that cowpea aphid-borne mosaic and blackeye cowpea mosaic viruses are two different potyviruses. Journal of General Virology. 74: 335-340.

Lambot, C. (2002). Industrial Potential of Cowpea. In: Challenges and Opportunities for Enhancing Sustainable Cowpea Production. [(eds) Fatokun, C.A., Tarawali, S.A., Singh, B.B., Kormawa, P.M. and Tam, M.] IITA, Ibadan. 367-375.

Leão, U.S., Eiras, M., da Rocha Nogueira, M.D.S., Rodrigues, L.K., Ribeiro, V.Q. and Lopes, A.C.D.A. (2016). Selection of phenotypic traits and resistance to cowpea severe mosaic virus and 'Cowpea aphid-borne mosaic virus' in cowpea [*Vigna unguiculata* (L.) Walp.] seeds with rugose white coat. Australian Journal of Crop Science. 10(4): 470.

Lima, J.A.A., Sittolin, I.M. and Lima, R.C.A. (2005). Diagnose e Estratégias de Controle de Doenças Ocasionadas por Vírus. In: Feijão-Caupi Avanços Tecnológicos [Freire Filho, F.R., Lima, J.A.A. and Ribeiro, V.Q., (eds.)]. Embrapa Informação Tecnológica, Brasília. 403-459.

Lima, J.D.A., Santos, C.D.G. and Silveira, L.F.S. (1986). Comportamento de genótipos de caupi em relação aos dois principais vírus que ocorrem no Ceará. Fitopatologia Brasileira. 11: 151-161.

Lima, L.F. (2019). Herança e incorporação de gene de resistência ao cowpea severe mosaic virus em genótipos de feijão-caupi da subclasse manteiga. Thesis submitted to Universidade Federal Rural da Amazônia, Brazil.

Lin, M.T., Hill, J.H., Kitajima, E.W. and Costa, C.L. (1984). Two new serotypes of cowpea severe mosaic virus. Phytopathology. 74: 581-585.

Lin, M., Anjos, J.R.N. and Rios, G.P. (1981). Serological grouping of cowpea severe mosaic virus isolates from Cental Brazil. Phytopathology. 71: 435-438.

Mackenzie, A.M., Nolan, M., Wei, K.J., Clements, M.A., Gowanlock, D. and Wallace, B.J. (1998). Ceratobium mosaic potyvirus: Another virus from orchids. Archives of Virology. 143: 903-914.

Maciel, S.C., Silva, R.F.D., Reis, M.S., Jadão, A.S., Rosa, D.D., Giampan, J.S. and Camargo, L.E. (2011). Characterization of a new potyvirus causing mosaic and flower variegation in Catharanthus roseus in Brazil. Scientia Agricola. 68: 687-690.

MAG/DCEA (Ministerio de Agricultura y Ganadería, PY); (Dirección de Censos y Estadísticas Agropecuarias). (2017). Síntesis estadísticas: Producción agropecuaria año agrícola 2016/ 2017. San Lorenzo, Paraguay. 54 p

Mowat, W.P. and Dawson, S. (1987). Detection of plant viruses by ELISA using crude sap extracts and unfractionated antisera. Journal of Virological Methods. 15: 233-247.

Natalia de Jesús, J. and de López, M.B.R. (2013). Evaluación de Aphis craccivora como vector del virus potyvirus (CABMV) en plantas de sésamo (*Sesamum indicum* L.) variedad escoba blanca. Investigación Agraria. 12(2): 85-89.

Ndiaye, M., Bashir, M., Keller, K.E. and Hampton, R.O. (1993). Cowpea viruses in Senegal, West Africa: identification, distribution, seed transmission and sources of genetic resistance. Plant Disease. 77(10): 999-1003.

Nogueira, M.D.S., freire Filho, F.R., de Oliveira, C.R.R., Barros, G.B., Brioso, P.S.T. and Ribeiro, V.Q. (2011). Avaliação da reação de genótipos de feijão-caupi a infecção por vírus por meio dos efeitos qualitativos. In congresso brasileiro de melhoramento de plantas, 6., Búzios. Panorama atual e perspectivas do melhoramento de plantas no Brasil.

Oliveira, C.R.R.D., Freire Filho, F.R., Nogueira, M.D.S.D.R., Barros, G.B., Eiras, M., Ribeiro, V.Q. and Lopes, A.C.D A. (2012). Reação de genótipos de feijão-caupi revela resistência às coinfecções pelo Cucumber mosaic virus, Cowpea aphid-borne mosaic virus e Cowpea severe mosaic virus. Bragantia. 71(1): 59-66.

Pio-Ribeiro, G. and Assis Filho, F.M. (1997). Doenças do caupi. In: Manual de Fitopatologia: Doenças das plantas cultivadas. [Kimati, H.; Amorim, L., Bergamin F.A., Camargo, L.E.A. and Rezende, J.A.M. (Ed.)]. 4. ed. São Paulo: Agronômica Ceres, p. 233-244.

Taiwo, M.A. (2003). Viruses Infecting Legumes in Nigeria: Case History. In: Plant Virology in Sub-Saharan Africa, [Hughes, J.A. and B.O. Odu (Eds.)]. International Institute of Tropical Agriculture, Ibadan, Nigeria, 365-380.