Australian Journal of Crop Science AJCS
AJCS 6(7):1154-1158 (2012) ISSN:1835-2707

Inheritance studies of sterility mosaic disease (SMD) resistance in vegetable type pigeonpea (*Cajanus cajan* (L.) Millsp.)

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Abstract

Sterility mosaic disease (SMD) is one of the most destructive diseases of the Indian subcontinent causing severe reduction in yield. Development of resistant varieties is an effective management strategy for which understanding the nature of inheritance of disease inheritance is a prerequisite. The objective of this work was to study the nature of inheritance of SMD in resistant (BRG 3 and ICP 7035) and susceptible (ICP 8863 and TTB 7) genotypes. SMD incidence observed in parents, F_1 and F_2 generations indicated resistance to be controlled by recessive gene and appeared to be monogenic in cross TTB 7 × BRG 3 and governed by two independent non-allelic genes exhibiting complementary epistasis in cross ICP 8863 × ICP 7035. Expression of at least one SMD gene in homozygous recessive condition was necessary for resistant phenotype in the above mentioned crosses. Resistant × resistant and susceptible × susceptible F_2 individuals showed no segregation indicating function of same loci/ linked loci to govern resistance and susceptibility in the parents studied. Visual observations recorded for leaf colour and texture revealed that the leaves of resistant genotypes were dark green and leathery textured while, that of susceptible individuals were light green and non-leathery indicating leaf traits to be associated with SMD resistance in the parents studied. The putative association of dark green leathery leaves with SMD resistance after confirmation could make significant implications for pigeonpea improvement by providing opportunity for indirect selection of leaf traits for development of SMD resistant cultivars

Keywords: *Cajanus cajan*, inheritance, sterility mosaic disease, dark green leathery leaves.
Abbreviations: SMD- sterility mosaic disease; PPSMV-pigeonpea sterility mosaic virus.

Introduction

Pigeonpea [*Cajanus cajan* (L.) Millsp] is a multipurpose grain legume crop grown extensively for food in the Asian and African countries. Globally, pigeonpea is cultivated in an area of about 4.75 million hectares with an average productivity of 774 $kgha^{-1}$ (FAO, 2010). India is considered as the primary centre of origin for pigeonpea due to the presence of ample variability in local germplasm and wild relatives (Saxena, 2008). In recent years, the crop is gaining importance due to its inherent ability to perform well under marginal input conditions and also its adaptability to withstand drought and other abiotic stresses. The major biotic stresses causing economic concerns in yield are the *Fusarium* wilt, sterility mosaic disease (SMD) and Phytopthora blight (Reddy et al., 1998). SMD is one among the most destructive disease of pigeonpea (Kannaiyan et al., 1984) causing yield losses up to 95 per cent (Reddy and Nene, 1981; Ganapathy et al., 2011). The causal agent of the disease is pigeonpea sterility mosaic virus (PPSMV) and is transmitted by vector eriophyid mite (*Aceria cajani* Channabasavanna) (Kumar et al., 2003). The symptoms of SMD include bushy and pale green leaves, excess vegetative growth, leaf size reduction, mosaic and mottling of leaves and cessation of reproductive structures. Most of the cultivars in Karnataka state, India especially are susceptible to SMD leading to decline in productivity levels. Chemical method of disease control though effective to control the mite population but not economical since the crop is grown under marginal input conditions. Development and cultivation of resistant varieties is considered as one of the most viable options for control of the disease. Development of SMD resistant cultivars requires clear understanding of its genetics. There are contradicting reports about genetics of resistance to SMD claiming both recessive and dominant genes. However in most cases, susceptibility was observed to be dominant (Srinivas et al., 1997; Nagaraj et al., 2004; Gnanesh et al., 2011). Mechanism of resistance to SMD was not clearly understood in spite of several reports available on nature of its inheritance. Murugesan et al. (1997) indicated monogenic segregation ratio for SMD and leaf characters and further reported joint segregation of SMD with leaf characters. Few other reports have indicated resistance in ICP 7035 is due to the thick leaf cuticle by which eriophyid mite cannot penetrate the leaf epidermal cells and hence not able to transmit the pathogen (Reddy et al., 1995). In spite of extensive research efforts for

improving the productivity, no *per se* improvement in yield was recorded. Development of SMD resistant parental lines and exploitation of heterosis by utilizing recently developed cytoplasmic genic male sterility systems will help in increasing the productivity of pigeonpea to a greater extent (Saxena et al., 2006). Development of hybrids with SMD resistance requires extra efforts and unless SMD resistance is present in male and female parents, it is unlikely to reflect in hybrids since resistance is governed by recessive genes in most cases. In view, of the above importance, the present investigation was undertaken to work out the genetics of resistance to SMD in vegetable type resistant lines (BRG 3 and ICP 7035) and to determine association of SMD resistance with leaf traits (leaf colour and texture).

Results and Discussion

Genetics of F_1s to SMD

The resistant lines BRG 3 and ICP 7035 showed 100 per cent resistance with no visible symptoms while, the susceptible lines TTB 7 and ICP 8863 exhibited 100 per cent infection with severe mosaic symptoms (Table 1). The F_1 hybrids of the susceptible × resistant cross combinations (TTB 7 × BRG 3 and ICP 8863 × ICP 7035) were susceptible indicating susceptibility to be dominant over resistance (Table 2). Similar studies showing susceptibility controlled by dominant genes have been reported. (Singh et al., 1983; Sharma et al., 1984; Srinivas et al., 1997; Nagaraj et al., 2004; Ganapathy et al., 2009; Gnanesh et al., 2010).

Genetics of F_2s to SMD

The F_2 individuals in all the four crosses were grouped into two classes *viz.,* resistant (no visible symptoms) and susceptible (severe mosaic symptoms) based on the disease reaction. The goodness of fit to the Mendelian segregation for resistance and susceptibility in the F_2 plants based on the chi-square test is presented in Table 3. Segregation in F_2 generation revealed digenic ratio of 9 (susceptible):7 (resistant) for cross ICP 8863 × ICP 7035. In contrast, the F_2 individuals in cross TTB 7 × BRG 3 exhibited monogenic segregation ratio of 3: 1 for susceptibility and resistance respectively. The study reveals that the resistant parent ICP 7035 differs from susceptible parent ICP 8863 for two independent genes while, the resistant parent BRG 3 differed from susceptible parent TTB 7 for single gene. Similar studies reporting variation in number of genes controlling resistance in different genotypes were reported by Singh et al., (1983); Sharma et al., (1984); Srinivas et al., (1997); Nagaraj et al., (2004). A digenic ratio (9 susceptible: 7 resistant) resulted in the cross ICP 8863 × ICP 7035 indicates the complementary nature of two dominant genes for susceptibility to SMD. For obtaining resistant genotype any one of the SMD locus or both locus in homozygous recessive condition was required in TTB 7 × BRG 3 and ICP 8863 × ICP 7035 while, dominant genes at both loci would result in susceptibility to SMD in above mentioned crosses. The probable genotypes for resistance and susceptibility in the parents, F_1 and F_2 generations of the above mentioned crosses are in Table 4. All the F_1 plants from resistant × resistant (BRG 3 × ICP 7035) cross were resistant (Fig. 1a) while, the F_1s of susceptible × susceptible (TTB 7 × ICP 8863) were susceptible (Fig. 1b). Resistant × resistant and susceptible × susceptible F_2 individuals showed no segregation indicating function of same loci/ linked loci to govern resistance and susceptibility in the parents studied. Similar results have been reported by Srinivas et al. (1997) in crosses involving resistant (ICP 7035, ICP 7349 and ICP 8850) and susceptible (BDN1 and LRG30) genotypes. The segregation ratios obtained from F_2 generation could not be confirmed from test crosses, due to insufficient seeds obtained from back crosses during off-season.

Association of leaf colour and texture with resistance

The parental genotypes used in the study were contrasting for their leaf colour and texture. The leaves of resistant parents (BRG 3 and ICP 7035) were dark green in colour and leathery textured while that of the susceptible parents (TTB 7 and ICP 8863) were light green coloured and non-leathery textured (Fig. 1c). Association of resistance with leaf characteristics was considered in F_2 generation to examine if the characters were associated. Visual observations from F_2 plants of cross TTB 7 × BRG 3 and ICP 8863 × ICP 7035 showed dark green and leathery textured to be associated with SMD resistance while, light green and non-leathery textured were found to be associated with susceptibility (Fig. 1d). The Chi-square tests also indicated that there exists genetic association between the resistance and leaf characters (colour and texture) studied (Table 5). Similar results showing genetic correlation between the SMD resistance and leaf morphological traits was reported by Murugesan et al. (1997). All these results provide tentative evidence that the resistance to SMD could be due to the leaf morphological traits which prevent the mites from feeding on resistant plants. Reddy et al. (1995) reported that in SMD resistant genotypes leaf cuticle and the epidermal cell wall were thicker as compared to susceptible genotypes. Out of four parental genotypes used in our study, two genotypes ICP 7035 (resistant) and ICP 8863 (susceptible) were common in the studies conducted by Reddy et al. (1995) and they observed 3.79 µm thick cuticle in ICP 7035, which is about 50% more than the leaf cuticle thickness of ICP 8863 (2.27µm). They further substantiated that resistance is due to the thick cuticle of the resistant genotypes through which the mite cannot peneterate the living epidermal cells to transmit the SMD pathogen. Association of dark green leathery leaves with SMD resistance may be further confirmed from grafting experiments where resistant scion will be grafted on to susceptible stock. If the resistance is due to the mite, the virus will be transmitted from the susceptible stock to the resistant scion and the grafted plants would become susceptible and show mosaic symptoms. Kumar et al. (2005) reported 15 wild accessions resistant to three SMD isolates. All the 15 wild resistant accessions used did not support vector multiplication and further reported that most accessions imparting resistance became susceptible by graft inoculation suggesting that vector resistance is conferring resistance to SMD. In general, our findings revealed that the leaf leatheryness attributing to leaf thickness in the resistant plants were lesser in the initial stages of crop growth as compared to later stages. Further, development of quantitative methods for measuring the leaf colour and thickness will help in more precise selection of the resistant plants. From the earlier reports, it is understood that PPSMV infects the crop in the initial stages of crop growth. All these results could further support the fact that resistance to SMD could be due to the inability of mites from feeding the resistant plants. There could be many other biochemical factors attributing to SMD resistance associated with dark green leathery leaves. Further, confirmation of association of leaf traits with SMD resistance may be done from the F_2 derived advanced progenies under replicated experiments.

Table 1. Resistant and susceptible reaction of parental genotypes to SMD during 2006 and 2007 screening experiments.

Parents	Rainy season (2006)			Rainy season (2007)			Disease incidence (%)	Disease Reaction	Leaf colour and texture
	Total plants	R	S	Total plants	R	S			
BRG 3	26	26	-	16	16	-	0	R	Dark green leathery leaves
ICP 7035	31	31	-	15	15	-	0	R	Dark green leathery leaves
TTB 7	19	-	19	17	-	17	100	S	Light green non-leathery leaves
ICP 8863	23	-	23	19	-	19	100	S	Light green non-leathery leaves

R = Number of resistant plants; S= Number of susceptible plants , '-' indicates no plants

Table 2. Resistant and susceptible reaction of F_1 hybrids to SMD indicating recessive nature of genes governing resistance in F_1 hybrids.

F_1 hybrids	Total plants	R	S	Disease incidence (%)	Disease reaction
ICP 8863 × ICP 7035	15	-	15	100	S
TTB 7 × BRG 3	16	-	16	100	S
BRG 3 × ICP 7035	14	14	-	0	R
TTB 7 × ICP 8863	16	-	16	100	S

R= Number of resistant plants; S= Number of susceptible plants, '-'Indicates no plants

Table 3. Chi-square test for segregation of resistance and susceptibility in F_2 populations during rainy season, 2007 revealing nature of inheritance to SMD.

F_2 generation	Total plants	Sterility mosaic disease (SMD)				Ratio S:R	χ^2	*P* value
		Observed		Expected				
		R	S	R	S			
ICP 8863 × ICP 7035	179	74	105	78.31	100.69	9:7	0.42	0.52
TTB 7× BRG 3	221	52	169	55.25	165.75	3:1	0.27	0.61
BRG 3 × ICP 7035	192	192	-	192	-	-	-	-
TTB 7 × ICP 8863	196		196	-	196	-	-	-

R=Number of resistant plants; S= Number of susceptible plants, '-'Indicates no plants

Table 4. Probable genotypes of parents and F_2s for SMD resistance.

Generation	Phenotype	Proposed genotype(s)
Parents		
BRG 3	Resistance	*aaBB*
ICP 7035	Resistance	*aabb*
TTB 7	Susceptible	*AABB*
ICP 8863	Susceptible	*AABB*
F_2 plants	Resistance	*aaB-, A-bb, aabb,*
	Susceptible	*AAB-, A-B-, aaB-,*

Table 5. Association of SMD with leaf colour and texture in the F_2 generation tested using chi-square.

F_2 generation	Total plants	Resistant plants		Susceptible plants		χ^2	*P* value
		Light green and non leathery leaves	Dark green and leathery leaves	Light green and non leathery leaves	Dark green and leathery leaves		
ICP 8863 × ICP 7035	179	4	70	105	-	0.37	0.54
TTB 7 × BRG 3	221	3	49	169	-	0.23	0.63
BRG 3 × ICP 7035	192	-	192	-	-	-	-
TTB 7 × ICP 8863	196	-	-	196	-	-	-

'-' indicates no plants

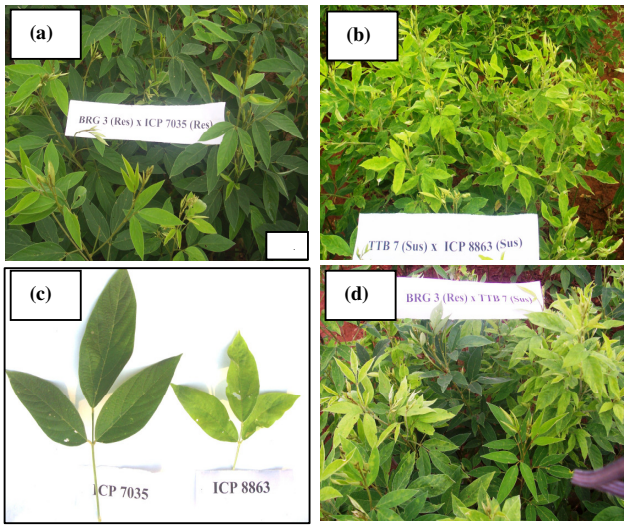

Fig 1. Segregation for SMD resistance and its association with leaf colour and texture. (a) Expression of 100% resistance in F2 of R × R cross (BRG 3 × ICP 7035). (b) Expression of 100% susceptibility in S × S cross (TTB 7 × ICP 8863). (c) Uninfected resistant (ICP 7035) and susceptible (ICP 8863) parents showing dark green leathery leaves and light green non-leathery leaves respectively. (d) Association of dark green and leathery leaves with SMD resistant plants in F2 of cross TTB 7 × BRG 3.

Association of leaf traits with SMD resistance though needs confirmation but, remains a difficult task for indirect selection of resistant plants in the early crop stages unless quantitative methods for measuring the leaf thickness are established. Hence there is need for genomic tools for assisting indirect selection of resistant plants by using marker assisted selection. Recent developments in genomics (Guo et al., 2011; Wang et al., 2011) especially pigeonpea genomics initiative (PGI) programme (Varshney et al., 2010) will aid in development and identification of markers for subsequent use in marker assisted selection. Again since the resistant parents used in the present study possessed traits of vegetable importance (bold seeds with high seed weight), desirable segregants (bold seeds, high seed weight and SMD resistance) obtained in the F_2 (data not shown) and later generations may be tested for sweetness and other cooking qualities for development of lines/varieties meeting the requirements of the end users (Byre Gowda et al., 2003; Upadhyaya et al., 2010).

Materials and methods

Plant materials

Based on the previous reports (Saifulla et al., 2003 and Rangaswamy et al., 2005) four genotypes were selected as parents for inheritance studies. The parental materials were confirmed for their resistance during 2006 and 2007 following Leaf Stapling Technique (Nene and Reddy, 1977) at All India Co-ordinated Research Project on pigeonpea, Bangalore, India. The place is located at 12°58' latitude north and 77°35' longitude east and with an altitude of 930 meters above sea level.

Hybridization programme

Two genotypes (BRG 3 and ICP 7035) selected as resistant parents (with no visible symptoms) were crossed with two susceptible genotypes TTB 7 and ICP 8863 (severe mosaic symptoms). The resistant and the susceptible genotypes were crossed among themselves to identify allelism in the resistant and susceptible genotypes. Hybridization was carried out under bee proof nylon net to prevent contamination by natural out crossing. Morphological traits such as plant type, flower colour, pod colour, seed colour and seed size were used as markers to check the trueness of F_1 plants. Part of the F_1 seeds from each of the four crosses (TTB 7 × BRG 3, ICP 8863 × ICP 7035, BRG 3 × ICP 7035 and TTB 7 × ICP 8863) were raised during January, 2007 (off-season) under bee proof nylon nets for rapid generation advancement to F_2 and the remnant seeds were retained for SMD screening.

SMD evaluation

Parents, F_1 and F_2 generations were raised in 15 × 45 cm polybags during rainy-season, 2007 and placed in SMD infected field. All the plants were artificially infected following leaf stapling technique by stapling infected leaves containing mites, carrying the disease, onto the leaves of test plants at two to three leaf stages. Stapling with infected leaves was carried out at periodic intervals to minimize disease escape. The infected plants were scored for SMD incidence at 15 days interval up to 75 days and were classified as resistant (no visible symptoms) or susceptible (severe mosaic symptoms) based on the mosaic symptoms. The same sets of F_2 plants were also recorded for their leaf colour and texture before the appearance of the mosaic symptoms and were classified as dark green leathery and light green non-leathery leaves. The resistant plants obtained after screening experiments were transferred to the field to identify desirable segregants for yield and quality traits of vegetable importance. The crop was grown according to the standard cultural recommendations for the area.

Statistical analysis

The total number of plants falling into different reaction classes (resistant and susceptible) were counted and subjected to chi-square (χ^2) analysis for goodness of fit to various classical Mendelian ratios as suggested by Panse and Sukhatme (1985). Chi-square tests were also conducted to evaluate the association between leaf traits with SMD.

Conclusions

The results of our experiment revealed resistance to be governed by two independent non-allelic genes (digenic) in cross involving ICP 7035 and ICP 8863 and monogenic in cross involving TTB 7 and BRG 3. Accordingly two gene model was proposed for the resistant lines studied. Number of genes controlling SMD resistance depends on the parental materials used for the study and it is also evident from the previous reports. The putative association of dark green leathery leaves with SMD resistance may be confirmed from grafting experiments from the advanced progenies of the above mentioned crosses. This could bring a shift in breeding for SMD resistant lines/varieties by undergoing indirect selection of leaf traits from the segregating generations for overall improvement in the pigeonpea crop.

References

Byre Gowda M, Saifulla M, Jagadeesh Babu CS, Shankaralingappa BC, Mahadevu P (2003) BRG 1: A high-yielding and bold-seeded pigeonpea variety for dhal and vegetable purpose. Int Chickpea Pigeonpea Newsl 10: 42-44

FAO (2010) http://faostat.fao.org/site/567/default.aspx#ancor

Ganapathy KN, Byre Gowda M, Venkatesha SC, Ramachandra R, Gnanesh BN Girish G (2009) Identification of AFLP markers linked to sterility mosaic disease in pigeonpea (*Cajanus cajan*) (L.) Millsp. Int J Integr Biol 7: 145-149

Ganapathy KN, Gnanesh BN, Byre Gowda M, Venkatesha SC, Gomashe SS Channamallikarjuna V (2011) AFLP analysis in pigeonpea (*Cajanus cajan* (L.) Millsp.) revealed close relationship of cultivated genotypes with some of its wild relatives. Genet Resour Crop Evol 58: 837-847

Gnanesh BN, Ganapathy KN, Ajay BC, Byregowda M (2011) Inheritance of sterility mosaic disease resistance to Bangalore and Patancheru isolates in pigeonpea (*Cajanus cajan* (L.) Millsp.) Electronic J Plant Breed 2:218-223

Guo Y, Wang XB, He W, Zhou GA, Guo BF, Zhang L, Liu ZX, Luo ZQ, Wang LH, Qiu LJ (2011) Soybean omics and biotechnology in China. Plant Omics J 4: 318-328

Kannaiyan J, Nene YL, Reddy MV, Ryan JG Raju TN (1984) Prevalence of pigeonpea disease and associated crop losses in Asia, Africa and the Americas. Trop Pest Manage 30: 62-71

Kumar PL, Jones AT Reddy DVR (2003) A novel mite transmitted virus with a divided RNA genome closely associated with pigeonpea sterility mosaic disease. Phytopath 93: 81- 91

Kumar PL, Latha TKS, Kulkarni NK, Raghavendra N, Saxena KB, Waliyar F, Rangaswamy KT, Muniyappa V, Sabitha Doraiswamy Teifion A (2005) Broad based resistance to pigeonpea sterility mosaic disease in wild relatives of pigeonpea (*Cajanus*: Phaseoleae) Ann Appl Biol 146: 371-379

Murugesan S, Murugan E, Nadarajan N (1997) Inheritance of duration, leaf colour, sterility mosaic disease resistance and growth habit in pigeonpea. Madras Agric J 84: 10-12

Nagaraj KM, Chikkadevaiah, Kulkarni RS (2004) Inheritance of resistance to sterility mosaic virus in pigeonpea (*Cajanus cajan* (L.) Millsp.). Indian J Genet 64: 118-120

Nene YL, Reddy MV (1977) Leaf stapling technique to screen pigeonpea for resistance to sterility mosaic. Indian Phytopath 30: 153

Panse VG Sukhatme PV (1985) Statistical methods for agricultural workers, Indian Council of Agricultural Research, New Delhi

Rangaswamy KT, Muniyappa V, Kumar PL, Saxena KB, Byre Gowda M, Raghavendra N, Pandurangaiah K, Kumar RV, Waliyar F, Jones AT (2005) ICP 7035 - A sterility mosaic resistant vegetable and grain purpose pigeonpea variety. SAT eJournal 1:1- 3

Reddy MV Nene YL (1981) Estimation of yield loss in pigeonpea due to sterility mosaic. In: Proc. International workshop on Pigeonpeas Vol. 2: 15–19 December 1980, ICRISAT Center, Patancheru, AP, India, p 305-312

Reddy MV, Raju TN, Lenne JM (1998) Diseases of pigeonpea. In: Allen DJ and Lenne JM (eds) The pathology of food and pasture legumes. CAB International, ICRISAT. India p 517-558

Reddy MV, Sheila VK, Murthy AK, Padma N (1995) Mechanism of resistance to *Aceria cajani* in pigeonpea. Int J Trop Plant Dis 13: 51- 57

Saifulla M, Byre Gowda M (2003) Sterility mosaic resistant genotypes of pigeonpea. In: National symposium on Pulses for crop diversification and natural resource management. December 20 – 22, Kanpur, India. p 235.

Saxena KB (2008) Genetic improvement of pigeonpea- A review. Tropical Plant Biol 1: 159-178

Saxena KB, Kumar RV, Madhavi Latha K, Dalvi VA (2006) Commercial pigeonpea hybrids are just a few steps away. Indian J Pulses Res 19:7-16

Sharma D, Gupta SC, Rai GS, Reddy MV (1984) Inheritance of resistance to sterility mosaic disease in pigeonpea. Indian J Genet 44: 84 -90

Singh BV, Pandya BP, Gautam PL, Beniwal SPS, Pandey MP (1983) Inheritance of resistance to sterility mosaic virus in pigeonpea. Indian J Genet 43: 487- 493

Srinivas T, Reddy MV, Jain KC, Reddy MSS (1997) Studies on inheritance of resistance and allelic relationships for strain 2 of pigeonpea sterility mosaic pathogen. Ann Appl Biol 130: 105-110

Upadhyaya HD, Reddy KN, Gowda CLL, Sube Singh (2010) Identification and evaluation of vegetable type pigeonpea (*Cajanus cajan* (L.) Millsp.) in the world germplasm collection at ICRISAT genebank. Plant Genet Res 8: 162-170

Varshney RK, Penmetsa RV, Dutta S, Kulwal PL, Saxena RK, Datta S, Sharma TR, Rosen B, Carrasquilla-Garcia N, Farmer AD, Dubey A, Saxena KB, Gao J, Fakrudin B, Singh MN, Singh BP, Wanjari KB, Yuan M, Srivastava RK, Kilian A, Upadhyaya HD, Mallikarjuna N, Town CD, Bruening GE, He G, May GD, McCombie R, Jackson SA, Singh NK, Cook DR (2010) Pigeonpea genomics initiative (PGI): an international effort to improve crop productivity of pigeonpea (*Cajanus cajan* L.). Mol Breed 26:393-408

Wang XJ, Liu ST, Xia H, Wan SB, Zhao CZ, Li AQ (2011) Peanut (*Arachis hypogaea* L.) omics and biotechnology in China. Plant Omics J: 4:339 -349.