



Utilization of synthetic amphidiploids in resistance breeding of groundnut

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Abstract: Eight diploid wild species of groundnut were hybridized to produce eight amphidiploids. Both the parental diploid species and synthetic amphidiploids were screened for foliar diseases viz. early leaf spot and rust, besides for sucking pests viz. thrips and leaf hopper. All the diploid species had high level of resistance for rust disease. *Arachis cardenasii* registered high level of resistance for both leaf hopper. Those desirable attributes were combined in the amphidiploids. The pollen fertility ranged from 19.0 to 67.4 per cent in the seven amphidiploid involving both 'A' genome species. Whereas it was only 1.5 per cent in *A. villosa* x *A. batizocoi* where 'A' and 'B' genomes were involved.

Key words : Synthetic amphidiploid, Resistance breeding, Genome.

Introduction

The groundnut crop (*Arachis hypogaea* L) is widely cultivated throughout tropical and subtropical regions. Increased variability is needed in cultivars for genes conditioning disease and insect resistance. One germplasm source available for introgressing genes to *A. hypogaea* is the vast array of wild species. The genus *Arachis* has been divided into seven sections (Gregory and Gregory, 1979). Although 40 to 70 species probably exist, only members of section *Arachis* are cross compatible with the cultivated groundnut. Included in this group are two tetraploids viz. *A. hypogaea* and *A. monticola* ($2n=40$) and 10 to 15 diploid species.

The *A. hypogaea* is a segmental allotetraploid made up of two A and two B genomes (Husted, 1936; Singh and Moss, 1984). The A genome is found in several diploid species, while the B genome is present only in *A. batizocoi* (Smart *et al.* 1978a).

The wild species have high level of resistance or immunity to many of the most important pests, including *Cercospora arachidicola* and *Phaeoisariopsis personata* (Abdou *et al.* 1974) rosette virus (Gibbons 1969 and Nevill, 1978); Peanut stunt virus (Hebert and Stalker, 1981); Peanut mottle virus (Denski and Sowell, 1981); rust (Bromfield and Cevrio, 1970 ; Hammons, 1977 and Subrahmanyam *et al.*

1980), Northern root - knot nematode (Castillo *et al.* 1973); mites (Johnson *et al.* 1977 ; Leuck and Hammons, 1968) and thrips, leaf hoppers and corn ear worm (Campbell and Stalker, 1982).

The genomic relationship between wild and cultivated species suggested that hybridization between synthetic amphidiploids of wild species and *A. hypogaea* should be a practical approach for the transfer of desirable genes from wild species into *A. hypogaea* (Stalker and Wynne, 1979 ; Moss 1980 ; Gardner and Stalker, 1983 ; Singh and Moss 1984 and Singh, 1985). Hence, the present study was conducted to study the usefulness of some amphidiploids in resistance breeding.

Materials and Methods

The diploid ($2n=2x=20$) wild species numbering eight viz. *A. batizocoi*, *A. duranensis*, *A. stenosperma*, *A. helodes*, *A. villosa*, *A. correntina*, *A. cardenasii* and *A. kempff-mercadori* were chosen for producing amphidiploids. Among which *A. batizocoi* and *A. duranensis* were annuals and the rest were perennials. Further, *A. batizocoi* was the only species with B genome and all others were having A genomes. The eight species were utilized in the random hybridization to produce eight amphidiploids viz. *A. villosa* x *A. stenosperma*, *A. duranensis* x *A. villosa*, *A. villosa* x *A. batizocoi*, *A. cardenasii* x *A. villosa*, *A. correntina* x *A. helodes*,

Table 1. Reaction of parental diploid species and their synthetic amphidiploids to diseases and insect pests.

S.No.	Name of the species	Foliar diseases ¹			Sucking pests ¹	
		Early leaf spot	Late leaf spot	Rust	Thrips	Leaf hopper
A. Parental diploid species						
1	<i>A. batizocoi</i>	4.0	4.0	1.0	1.0	2.0
2	<i>A. duranensis</i>	2.5	3.0	1.0	4.0	1.0
3	<i>A. stenosperma</i>	3.5	4.0	2.0	2.5	2.0
4	<i>A. helodes</i>	3.5	3.5	2.0	4.0	4.0
5	<i>A. villosa</i>	2.5	2.5	1.0	4.0	2.0
6	<i>A. correntina</i>	3.5	4.0	1.0	2.0	2.0
7	<i>A. cardenasii</i>	2.0	2.0	1.0	2.0	2.0
8	<i>A. kempff-mercadoi</i>	3.0	3.0	1.0	3.5	4.0
B. Synthetic amphidiploids						
1	<i>A. villosa</i> x <i>A. stenosperma</i>	2.5	2.5	1.0	3.0	2.0
2	<i>A. duranensis</i> x <i>A. villosa</i>	2.5	2.0	1.0	3.5	3.0
3	<i>A. villosa</i> x <i>A. batizocoi</i>	3.0	3.0	1.0	2.0	2.5
4	<i>A. cardenasii</i> x <i>A. villosa</i>	2.0	2.0	1.0	2.5	2.5
5	<i>A. correntina</i> x <i>A. helodes</i>	3.5	3.0	1.5	3.0	3.5
6	<i>A. stenosperma</i> x <i>A. cardenasii</i>	2.0	2.5	1.0	2.0	2.0
7	<i>A. stenosperma</i> x <i>A. kempff-mercadoi</i>	3.0	3.5	1.5	3.0	3.0
8	<i>A. duranensis</i> x <i>A. stenosperma</i>	3.0	3.5	2.0	3.0	1.5
C. Check varieties						
1	TMV 2	8.5	8.0	7.5	6.5	7.0
2	VRI 2	8.0	8.0	7.0	6.0	7.0

Note : The screening was done under field condition with adequate diseases pressure

¹ - 1 to 9 scale scores

A. stenosperma x *A. cardenasii*, *A. stenosperma* x *A. kempff-mercadoi*, *A. duranensis* x *A. stenosperma*. unreduced pollen was very much important as they will be useful in further back crosses with tetraploid *A. hypogaea* parents.

The eight amphidiploids and eight parents were raised during *kharif* '99 season. The plants were screened for foliar diseases *viz.* early leaf spot, late leaf spot and rust besides sucking pests *viz.* thrips and leaf hopper under field conditions with adequate disease pressure by adopting infector row technique.

The plants were also studied for pollen stainability by staining with acetocarmine. Those pollen grains which took stain were considered as fertile. Even among fertile pollen grains some were larger in size which were considered as unreduced pollen. The occurrence of such

Results and Discussion

The reaction of the parental diploid species and amphidiploids to foliar diseases and sucking pests is furnished in Table 1. All the eight species exhibited high level of resistance to rust disease. *A. cardenasii* registered high level of resistance to both the leaf spots. *A. batizocoi* was highly resistant to thrips, whereas *A. duranensis* had immunity for leaf hopper.

The morphology of the amphidiploids and their level of resistance to diseases and pests are briefly described.

Table 2. Pollen stainability in synthetic amphidiploids and their parental diploid species

S. No.	Name of the species	Percentage of fertile pollen		
		Large pollen	Uniform size pollen	Percentage of sterile pollen
A. Parental diploid species				
1	<i>A. batizocoi</i>	0	99.0	1.0
2	<i>A. duranensis</i>	0	99.0	1.0
3	<i>A. stenosperma</i>	0	100.0	0
4	<i>A. helodes</i>	0	99.0	1.0
5	<i>A. villosa</i>	0	98.0	2.0
6	<i>A. correntina</i>	0	100.0	0
7	<i>A. cardensii</i>	0	100.0	0
8	<i>A. kempff-mercadoi</i>	0	99.0	1.0
B. Synthetic amphidiploids				
1	<i>A. villosa</i> x <i>A. stenosperma</i>	0.5	40.0	59.5
2	<i>A. duranensis</i> x <i>A. villosa</i>	0.2	57.5	42.3
3	<i>A. villosa</i> x <i>A. batizocoi</i>	0	1.5	98.5
4	<i>A. cardenasii</i> x <i>A. villosa</i>	0.5	45.6	53.9
5	<i>A. correntina</i> x <i>A. helodes</i>	0.8	50.5	48.7
6	<i>A. stenosperma</i> x <i>A. cardenasii</i>	0.4	56.9	42.7
7	<i>A. stenosperma</i> x <i>A. kempff-mercadoi</i>	0.6	67.4	32.0
8	<i>A. duranensis</i> x <i>A. stenosperma</i>	0.3	19.0	80.7

A. villosa x *A. stenosperma*

The standard petal colour of *A. stenosperma* was lemon yellow. This trait dominated in the amphidiploid. However, the lanceolate leaf shape of *A. villosa* expressed in the amphidiploid. *A. villosa* registered resistance to the two leaf spot diseases, and the resistance for sucking pests were moderate. Whereas, the reverse was true for *A. stenosperma*. However, the amphidiploid combined both the desirable attributes.

A. duranensis x *A. villosa*

The traits of *A. duranensis* viz. smaller size leaflets and standard petals were dominated in the amphidiploid. *A. duranensis* recorded immune reaction to leaf hopper and *A. villosa* recorded only moderate score for this pest. However, the amphidiploid was intermediate between the two.

A. villosa x *A. batizocoi*

The characters of *A. batizocoi* viz. shy branching, longer vines, round leaflets and lemon yellow standard petal colour were

dominated in the amphidiploid. The size of the standard petal was intermediate between the two. *A. batizocoi* recorded high level of resistance to sucking pests and moderate score for both the leaf spots, whereas, the reverse was true for *A. villosa*. However, the amphidiploid combined the desirable attributes of both.

A. cardenasii x *A. villosa*

The amphidiploid combines the bigger size standard petal of *A. villosa* and larger size leaflets of *A. cardenasii*. *A. cardenasii* registered high level of resistance to both foliar diseases and sucking pests studied. The amphidiploid improved its performance over *A. villosa* in respect of the sucking pests.

A. correntina x *A. helodes*

The characters of *A. helodes* viz. deep orange standard petal colour, dark green leaves and pinkish pigmentation in the stem were dominated in the amphidiploid. *A. correntina* exhibited resistance for sucking pests as compared to moderate score for the same by *A. helodes*.

However, the amphidiploid was intermediate in reaction for all the pest and diseases studied.

A. stenosperma x *A. cardenasii*

The length of the main axis was taller than both the parents in the amphidiploid. The standard petal colour was lemon yellow, resembling *A. stenosperma*. The pod size was intermediate between the two parents. Eventhough, *A. cardenasii* registered resistant reaction to all pest/diseases evaluated, *A. stenosperma* exhibited moderate reaction for both the leaf spots. However, the amphidiploid combined the desirable attributes of both the parents.

A. stenosperma x *A. kempff-mercadoid*

The amphidiploid expressed the bigger size standard petal as that of *A. kempff-mercadoid*. The colour of the standard petal was lemon yellow resembling, *A. stenosperma*. It was vigorous and the seed setting was profuse. *A. kempff-mercadoid* recorded moderate score for leaf spots, thrips and leaf hopper. The amphidiploid was better in respect of thrips and leaf hopper.

A. duranensis x *A. stenosperma*

The amphidiploid resembled *A. duranensis* in respect of smaller size leaflets and standard petal. Whereas the colour of the standard petal (lemon yellow) resembled *A. stenosperma*. In respect of resistance, the amphidiploid was intermediate between the parents.

The pollen fertility of the parental diploid species and the amphidiploids was estimated (Table 2). Among the fertile pollen, the fraction of large pollen was also estimated. None of the diploid species studied had large pollen. However, it was present in very low frequency in some of the amphidiploids. The occurrence of large pollen was due to unequal chromosome segregation which resulted in hyperdiploid gametes and spindle break down resulted in the formation of restitution nuclei and unreduced gametes (Singh, 1985).

The pollen fertility ranged from 19.0 to 67.4 per cent in the seven amphidiploids involving both 'A' genome species, whereas the pollen fertility of the only amphidiploid involving 'A' and 'B' genomes, namely,

A. villosa x *A. batizocoi* was only 1.5 per cent (Table 2). Karyological group of section *Arachis* species revealed that *A. batizocoi* had a distinct chromosome morphology (Stalker and Dalmacio, 1981). The cytological analysis of the amphidiploid *A. duranensis* x *A. villosa* revealed the formation of 8.9 bivalent and 2.2 univalent. Ressler and Gregory (1979) observed in hybrids between 'A' genome species of section *Arachis*, the absence of quadrivalents, anaphase bridges, and heteromorphic bivalents. Further, the differences resulting in the formation of univalents were cryptic differences rather than structural rearrangements between homologues. The pollen stainability studies revealed that the hybrids between species having 'A' genome were fertile, except the hybrid involving *A. batizocoi* ('B' genome) which was sterile. Smart *et al.* (1978a, 1978b) reported that the hybrid between *A. batizocoi* and other members of section *Arachis* exhibited structural heterozygotes, as well as disturbed reduction division and irregular segregation of chromosomes. However, doubling the chromosomes of this amphidiploid by colchicine treatment will restore fertility to the desired extent.

Thus, crossability, chromosome pairing and pollen and pod fertility in hybrids between *A. hypogaea* and amphidiploids have revealed that these amphidiploids can be used as a genetic bridge for the transfer of genes from the wild species into the cultivated groundnut. At Regional Research Station, Vriddhachalam, the following amphidiploids, viz. *A. cardenasii* x *A. villosa*, *A. correntina* x *A. helodes*, *A. stenosperma* x *A. cardenasii* and *A. stenosperma* x *A. kempff-mercadoid* were hybridized with *A. hypogaea* and their hybrids were utilized for further back crosses.

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