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Conventional rice breeding has met with limited success in trying to combine gene pools of *japonica* and *indica* subspecies. Hybridization of these two subspecies often results in high levels of spikelet sterility in the F1. To avoid this problem anther culture was explored as a tool to incorporate desirable traits from upland (*japonica sensu lato*) to irrigated (*indica*) rice. Progenies of four upland x irrigated crosses were produced through both anther culture and single seed descent breeding methods. Field performances of progenies derived from these two methods were evaluated at two rice-growing locations, Bagua (Perú) and Palmira (Colombia). Comparisons were made between breeding methods, F1- and F2-derived doubled haploids, and for random and selected populations, in terms of means, variances, and proportion of favorable recombinant lines for eight traits. These traits included: spikelet sterility, days to maturity, leaf pubescence, number of grains per panicle, number of

panicles per square meter, 1000-grain weight, plant height, and endosperm dispersion.

Single seed descent progenies had higher population means than doubled haploid progenies for leaf pubescence, 1000-grain weight, and plant height. Lower spikelet sterility was also observed in the populations resulting from the single seed descent method. No consistent differences between the two breeding methods were detected in terms of variances for the traits studied. Proportion of progenies with various combinations of desirable traits was higher in single seed descent populations. Doubled haploid progenies derived from the F1 had lower spikelet sterility and higher 1000-grain weight than those derived from the F2. Selection in the F2 prior to the production of doubled haploid lines was effective in increasing the proportion of lines for leaf pubescence and plant height as compared to random selection in the F2. However, lines derived from selected F2 plants did not have a higher proportion of desirable traits than those derived from the F1. These results suggest that the F1 is the generation of choice for deriving doubled haploid lines through anther culture in upland x irrigated rice crosses.

BRIDGING UPLAND-IRRIGATED RICE (*Oryza sativa* L.) GENE POOLS VIA ANTHER CULTURE

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Typed by researcher for Carlos B. Bruzzone Córdova

DEDICATED TO:

my parents Carlos and Virginia,
my brothers Miguel, Tito and Darío,
and my sister María,

my wife Rosa,
my children Rossana,
Carla and Arturo.

and to the memory of Lucho Flores Murrieta, late member of the Peruvian Rice Program - Selva.

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INTRODUCTION

Over the past 30 years rice production has experienced a substantial growth in Latin America. A significant amount of this enhanced production is attributed to the adoption of high yielding semidwarf irrigated *indica* cultivars. However, during the last decade, rice production growth has remained stagnant. Poor management practices and the lack of adequate resistance to key biotic and abiotic stresses has impeded reaching the yield potential of the modern cultivars. The short lived resistance of these cultivars to key diseases, such as blast (*Magnaporthe grisea*), and their expansion into less favorable environments, underlines the necessity of broadening the genetic makeup of the irrigated germplasm.

Genetic diversity is urgently needed in irrigated breeding programs for traits such as resistance to blast, brown spot (*Dreschlera oryzae*), leaf scald (*Rynchosporium oryzae*) and grain spotting (several pathogens). Other traits such as tolerance to Fe and Al toxicity, deep rooting patterns, and earliness are also desirable attributes in certain marginal rice production areas. Most of these traits are found in the Centro Internacional de Agricultura Tropical (CIAT)'s upland germplasm. Unfortunately, when this material is hybridized with irrigated *indica* germplasm, high levels of sterility and undesirable progenies often result. This

phenomenon is common in *japonica-indica* hybridizations and agrees with recent findings that African and South American upland rice germplasm, the core of CIAT's upland crossing program, is genetically close to the *japonica* type. Problems related to intersubspecific hybridization and successful deployment of high yielding semidwarf *indica* germplasm resulted in a declining interest in *japonica-indica* hybridizations and led to the narrowing of the genetic base of the rice cultivars currently planted in Latin America.

The importance of upland/*japonica* traits for selected rice production environments makes it imperative to reconsider how best to transfer those traits to irrigated/*indica* cultivars.

Anther culture (AC) techniques are acquiring increasing attention in rice improvement programs mainly due to their time-saving potential and their apparent ability to alleviate some of the difficulties that conventional breeding has met when dealing with wide crosses. However, doubled haploids (DH) derived from AC, unlike those lines developed by conventional breeding, are usually the result of only one round of recombination between the parental genomes. This may not be enough for breaking undesirable gene combinations and getting a reasonable number of favorable progeny.

This investigation compares upland (*japonica* wide sense) -irrigated (*indica*) progenies derived by the AC method **versus** those derived by the single seed descent method (SSD), the

conventional method most similar to DH breeding. The effect of the AC technique on spikelet sterility of DH progenies and the possible occurrence of gametophytic selection were also investigated in this research.

The F2 rather than the F1 generation has been proposed for deriving DH plants as it permits an additional round of recombination and allows a cycle of selection before the *in vitro* AC procedure. This study compares the field performance of F1- **versus** F2-derived DH progenies and estimates the effect of selection **versus** no-selection in the F2.

LITERATURE REVIEW

Rice, *Oryza sativa* L., is generally recognized to comprise three subspecies: *indica* Kato (= continental = hsien), *japonica* Kato (= temperate-insular = keng), and *javanica* Morinaga (= tropical-insular = bulu and gundil) (Takahashi, 1984). Of these, *indica* and *japonica* subspecies are the most important in geographical distribution, economic value, and rice improvement programs (Swaminathan, 1984).

More recent studies using isozyme analysis provide evidence that two extreme forms exist within rice: group I, to which the *indica* cultivars belong; and group VI, *japonica* wide sense, or non-*indica*, which incorporates *japonica* and *javanica* types (Glaszmann *et al.*, 1984; Glaszmann, 1985). Two minor groups, II and V, corresponded to cultivars found along the Himalayan range extending from Burma to Iran; while groups III and IV, are considered as "satellites" of the major groups, and found only in Bangladesh and Manipur State, in India. This classification has shown similarities with previous descriptions made by Oka (1957), and more recently by Jacquot and Arnaud (1979), based on 46 quantitative traits, and by Cheng (1985), using five diagnostic observations: phenol reaction, interval between first and second nodes of panicle axis, leaf pubescence, glume color at heading and length-width ratio of spikelets.

Rice is ecologically differentiated into three main groups: lowland, upland, and deep-water types (Chang, 1976).

Morphological and physiological studies indicate that African and South American upland cultivars are closely related to the *javanica* type (Jacquot and Arnaud, 1979). When compared, these studies are complementary to enzymatic rice classifications making it possible to confirm a likeness between African and South American upland rice types and the *javanica* type and show that these are genetically close to the *japonica* type (Glaszmann *et al.*, 1984; Second, 1984).

In Latin America modern high yielding semidwarf rice cultivars (HYRV) are generally grown in irrigated areas, however they are also grown in upland areas in Central America. Their use is expanding as well in more-favored upland areas in South America (Dalrymple, 1986; Rice Program, Internal Annual Report, CIAT, 1990). A CIAT study based on 1986-88 FAO data suggests that 76.5 % of the HYRV area in Latin America was irrigated and 23.5 % grown in upland areas (Rice Program, Internal Annual Report, CIAT, 1990).

The genetic and cytoplasmic base of the modern cultivars in the Latin American region is narrow, with one cultivar IR8 predominating in the lineages of most modern commercial cultivars. An analysis conducted by CIAT to estimate the genetic base of rice cultivars in Latin American rice fields revealed that 56% of the genes distributed in commercial cultivars come from the three land races which gave rise to IR8. This cultivar triggered the so called Green Revolution in rice (Rice Program, Internal Annual Report, CIAT, 1990).

Breeders at CIAT have acknowledged the potential threat that such a situation represents and have started a series of crosses involving the savanna (upland) and irrigated rice types. However, wide hybridization within rice, either through *japonica-indica* or irrigated-upland crosses, has not attained the desired outcome as a breeding method for bringing together new traits from different genetic backgrounds.

Several attempts have been made to transfer into the tropical *indica* cultivars such *japonica* traits as fertilizer responsiveness, cold tolerance, reduced plant height, earliness, slow leaf and stem senescence, and tough leaves. Similarly, a reciprocal interest exists in improving *japonica* cultivars by incorporating specific traits from *indica* cultivars including grain dormancy, glabrous leaves and glumes, disease resistance, and specific cooking and processing characteristics (Lee, 1979; Chung and Heu, 1980; Seetharaman, 1981).

Conventional breeding has met with limited success in developing *japonica-indica* cultivars. In the mid-1960's a cooperative project between South Korean and International Rice Research Institute (IRRI) scientists was initiated to transfer some of the desirable characteristics of the semidwarf indicas, particularly with respect to resistance to lodging and blast, to the japonicas. These scientists also tried to retain the ecological adaptability and eating

quality of japonicas. They started making *japonica-indica* crosses in 1968 and released their first *japonica-indica* cultivar, called Tongil, in 1972. To overcome grain sterility in *japonica-indica* hybrid populations and incorporate high productivity, the F1 of all *japonica-indica* crosses were crossed back to semidwarf indicas (Chung and Heu, 1980). Although 18 Tongil-type rice cultivars were released to farmers during the period 74-79, Korean scientists still had problems trying to combine the cold-water tolerance, grain properties and eating qualities of their traditional *japonica* cultivars with the high productive plant type and disease resistance of semidwarf *indica* cultivars (Chung and Heu, 1980; Dalrymple, 1986). Similar attempts by Taiwanese breeders have met with the same problems (Lee, 1979). A crossing program sponsored by FAO in Asia was equally unsuccessful in trying to combine the fertilizer responsiveness of *japonica* cultivars with the tropical adaptation of *indica* cultivars (Wasano, 1982, cited by Clement and Poisson, 1986).

Likewise, attempts to incorporate some upland traits into an irrigated background, and vice versa, have faced similar difficulties. In an effort to incorporate drought resistance into a high-yielding genotype, hundreds of crosses between traditional upland cultivars and semidwarf lowland cultivars have been made and evaluated at IRRI. Difficulties were encountered in obtaining a plant type that combines

intermediate plant stature, moderate tillering, drought resistance, moderately long panicles and long-and-slender grains. Numerous F2 and F3 progenies were either the semidwarf or the upland parental type (IRRI, 1974; 1975).

The limited success reached with *japonica-indica* hybridizations has been attributed to the presence of a high degree of sterility in the F1, extended segregating generations, and the differentiation of characters into two extreme opposites in *japonica-indica* progenies (Toriyama, 1979; Ikehashi, 1982; Shen Jin-Hua *et al.*, 1983). Oka (1957) considers that this partial sterility limits the probability of lines accumulating the characteristics of each parent and that such an obstacle to recombination leads to the evolution of the progenies towards their original respective parental forms. Nakagahara (1976), Toriyama (1979), and Seetharaman (1981), also related this lack of recombinants within the progenies of *indica* by *japonica* crosses to the F1 sterility.

Hung and Chang (1976), using three marker genes, have provided evidence of aberrant segregation in upland/lowland crosses. Atypical segregation for plant height and genotypic association among tall stature, long panicles, and large and broad grains was also found in upland/lowland cultivar crosses, by Lin and Chang (1981). Some aspects of the hybrid sterility and restricted segregations found in these crosses were similar to those observed in wide crosses between rice subspecies (Oka, 1964).

The term "hybrid sterility" is used to describe an array of spikelet sterilities caused for a lack of affinity between the parents, and the percentage of sterility has been considered as a function of the genetic distance between those genotypes (Chandraratna, 1963). In general, *indica-japonica* hybrids show F1 sterility more frequently than crosses between cultivars of the same group. Within the F1 of *indica* by *japonica* crosses, the compatibility may be expressed in various ways, and is specific to each particular parental combination. Most F1 hybrids show a sterility rate ranging from 40 to 90 % (Shankara Gowda *et al.*, 1972) or nule (Jennings, 1966), depending on the crosses considered. Therefore, the sterility relationship cannot be a discriminant character between the two types, and it is a mistaken belief that the *indica* and *japonica* types are isolated by F1 sterility (Oka, 1988). The F1 sterility occurs in both pollen and embryo sac, either in parallel or most often in pollen. Usually, pollen sterility and reduced seed set of F1 plants are observed (Oka, 1988).

Causes of F1 sterility of *sativa* intervarietal crosses are complex. The partial F1 sterility in *japonica-indica* crosses has been attributed to genic differences and different models can be applied to different situations (Oka, 1986). According to Oka (1974), the genetic basis of this phenomenon is gametophytic and controlled by a number of sets of duplicate genes governing gametic development. Ikehashi

and Araki (1986) have supplied evidence supporting earlier studies (Kitamura, 1962; cited by Ikehashi and Araki, 1986) which showed that an antagonism between the heterozygous maternal tissue and the gametes carrying one of the alleles is the basis for F1 sterility (one locus sporo-gametophytic interaction). High to complete sterility has not been explained yet in *indica-japonica* crosses and experts agree that we need more experimental work to explain various aspects of the sterility (Ikehashi and Araki, 1986).

The sterility occurring in F2 and later generations (particularly in the progeny of fertile F1 plants) is sporophytic (the development of gametes is controlled by genes of the maternal tissues). Evidence for this is the establishment of true breeding semi-sterile lines in the selfed progeny of partly fertile segregants. The F1 and F2 sterilities are not always correlated among different cross-combinations (Oka, 1988).

In *indica-japonica* hybrids, segregation ratios usually deviate from the Mendelian ratio owing to an increase of genes derived from the *indica* parent. This distortion of F2 ratios was found to be related to F1 sterility, suggesting that the gametophytic sterility genes cause gametic selection and certation (Oka, 1988).

In attempting to overcome the difficulties related to hybridization between *japonica* and *indica* cultivars, Chinese scientists have used anther culture to produce

doubled-haploid (DH) plants from the first generation of *japonica-indica* crosses. Although F1 plants are often sterile or semi-sterile, their anthers can be used as material for culturing pollen plants because their pollen sterility occurs after the uninucleate stage of pollen development (Zhang, 1989). Hsu and his colleagues (*see* Chu, 1982) found that fertility was restored in more than half of the spontaneously doubled anther-derived gametoclones that they regenerated from a partially sterile hybrid between *japonica* and *indica* subspecies. According to Li *et al.* 1983 (*see* Chen, 1986), in general, the rate of seed set of *japonica/indica* F1 generation is 26-30 %, and backcrossing could enhance the rate to 45-50%. In contrast, the fertility of pollen lines derived from the same combinations is as high as 80%.

Prolonged segregation through several generations following wide hybridization may be avoided through the production of doubled haploids using anther culture of the F1 hybrid. Using this approach Chinese scientists have released new rice cultivars which are widely grown in China (Chu, 1982; Zhang, 1982; Shen *et al.*, 1983). Anther-derived doubled haploid lines from *japonica-indica* crosses are being developed in Korea (Zapata *et al.*, 1986), Taiwan (Woo and Chen, 1980), and Centro International de Agricultura Tropical (CIAT) in Colombia (Pulver, 1986; Martínez and Pulver, 1989).

However, no release of *japonica-indica* cultivars developed in this way has been reported in those countries.

Niizeki and Oono (1968) were the first to succeed in regenerating haploid plants from rice pollen through *in vitro* culture of immature anthers (*see* Raina,1989). Thereafter, owing to the significance of haploids in speeding up breeding programs, rice anther culture gained considerable importance.

There exists considerable genotypic variation in anther response (Chaleff, 1979; Miah *et al.*, 1985, Raina, 1989). As the methodology has been improved over the years, genotypic variation is now seen mostly on the extent of response, rather than in the existence of some responsive and some unresponsive genotypes. In, general, *japonica* rice cultivars are known to respond better than *indicas* (Zapata, 1985, Pulver and Jennings, 1985, Chen, 1986). The culturing ability of glutinous rice is generally the highest. It was reported that the sequence ability was as follows: glutinous > *japonica* > *japonica/indica* hybrid > hybrid rice of *indica* > *indica* (Shen *et al.*, 1982, cited by Chen, 1986). Not only anther response, but also the subsequent regeneration of green plants, is better in *japonica* types. Chen (1986), has reported that on average the frequency of green plants was higher than 10% for cultured anthers when anthers were incubated in liquid media. However, in *indica* rice, the induction frequency of pollen plants still does not reach 2% on the average.

In addition to alleviating the problems associated with wide hybridizations, AC techniques allow the breeding period to be shortened through the rapid development of completely homozygous lines and consequently the ability to select among those lines in the absence of dominance (Baenziger and Schaeffer, 1983). When compared to the rate at which homozygosity is reached in a pedigree program the "instant" homozygosity obtained from using a doubled haploid system may increase the efficiency of selection for both qualitative, and quantitative inherited traits. If desirable alleles are recessive, then only a proportion $(1/4)^n$, (where **n** is the number of loci segregating), will have the desirable allelic combination. Consequently, the probability of fixation of all desirable alleles is low even if **n** is relatively small. However with a doubled haploid population, such genotypes will be at a frequency of $(1/2)^n$. Thus the frequency of fixation in a F1-derived doubled haploid population is the square root of the probability in a F2 population. Clearly, even for small numbers of major genes, the fixation of desirable recessive alleles is greatly facilitated (Baenziger and Schaeffer, 1983; Snape, 1989).

Although doubled haploid systems facilitate selection of major gene loci, their greatest advantage is increasing the efficiency of selection of quantitative characters. In the early generations of a pedigree system, efficient selection is hindered by low additive variance, presence of dominance, within-family segregation in the F2 and F3 generations.

Doubled haploids can help alleviate these difficulties. An advantage of using doubled haploid lines is that greater additive genetic variance is expressed between the recombinant products of a cross than between the relative F2's and F3's. Also dominance variation is absent (Snape, 1989).

Since DH lines are the products of meiosis in F1 plants haploid segregants are the result of only one round of recombination between the parental genomes. If repulsion linkage of useful genes is common, especially where parents are selected with contrasting attributes, doubling haploids may miss many of the advantageous recombinants (Walsh, 1974; Riggs and Snape, 1977). This situation may be particularly true in the case of wide crosses like those between upland and irrigated rice lines (or between *japonica* wide sense and *indica* types). This problem may be alleviated by increasing the number of doubled haploids produced in order to increase the probability to obtain favorable gene combinations (Choo, 1981), or by forming doubled haploids from the F2 or F3 rather than the F1 (Jinks and Pooni, 1981; Snape and Simpson, 1981). From a practical and theoretical standpoint the latter alternative could be the best approach for producing DH lines if linkage disequilibrium is important. More advanced generations (F2, F3) undergo further rounds of recombination allowing the break up of repulsion linkages. Nevertheless, one of the putative advantages of the use of DH in wide crosses resides in alleviating the partial sterility shown in

F1 plants. A delay in the production of DH's until the F2 or F3 would not resolve this problem.

It is also of interest for plant breeders to know whether DH lines show the same potential (estimated by means and variances of agronomic characters) as lines obtained by other methods. In theory, three factors could affect the means and variances of DH lines for a quantitative character: 1) the limited possibilities of genetic recombination through one cycle of meiosis compared with inbred lines in the presence of linkage (Snape, 1976; Riggs and Snape, 1977); 2) the possible occurrence of gametophytic selection (Powell *et al.*, 1986; Guiderdoni *et al.*, 1989a,1991); and, 3) culture-induced variation (Schaeffer, 1982; Zhang, 1989).

The doubled haploid method has been extensively compared with the single seed descent (SSD) method. In theory, the means and variances for a quantitative character for lines from two populations derived by the two methods are expected to be the same in the absence of linkage. When linkage and/or additive epistasis are present, however, the methods differ due to a higher frequency of recombinant lines in the SSD population (Snape, 1976). These theoretical findings were supported by the results of computer simulation studies (Riggs and Snape, 1977).

Studies of field performance have been carried out in barley to compare *Hordeum bulbosum*-derived DH lines with lines developed by the pedigree (PD) and the SSD methods (Park *et al.*, 1976). Similar means and ranges, genetic

variances and frequencies of desirable genotypes were obtained in the populations produced by three breeding methods for grain yield, heading date and plant height. Mean grain yields of superior lines were similar for all three methods. There was no indication of deleterious effect resulting from complete homozygosity in the DH lines. In the two crosses examined, the materials generated by the DH method were as good agronomically as those produced by the PD or SSD methods (Park *et al.*, 1976). A further analysis by Choo *et al.* (1982) found that the frequency distributions of grain yield, heading date, and plant height of the *Hordeum bulbosum*-derived DH and SSD lines were very similar. These results indicated that although the SSD method had more opportunity for recombination than the DH method it did not produce recombinants which differed significantly from the DH sample. A comparison between the DH and bulk plot (BK) breeding methods, using the same barley crosses showed that the DH method produce genotypes having the same grain yield potential as those developed by the BK method (Song *et al.*, 1978). Caligari *et al.* (1987) when comparing DH versus SSD lines for five spring barley crosses found a higher frequency of desirable recombinants among the DH lines. These differences may reflect a distinction in the method of extraction, i.e., the DH are the products of one round of meiosis while the SSD lines have been exposed to several cycles of meiosis. The higher frequency of desirable

recombinants may therefore be due to an excess of coupling linkages built up by previous recombination and selection (Snape and Simpson, 1981).

Charmet and Branlard (1985), found that the production of anther-derived DH lines of triticale from F1 plants provides the same range of recombinant genotypes as inbred lines obtained by several generations of selfing. This was true even for tightly linked genes such as those controlling gliadins. Similar results were found when androgenetic DH spring barley lines were compared in field experiments with their PD-selected sister lines. Both PD- and DH-families showed similar levels of performance with regard to the characters studied, although no conscious selection had been applied in deriving DH progeny (Friedt *et al.*, 1986).

Aside from preliminary results by Pulver (1986), and, Martínez and Pulver (1989), no agronomic comparison between anther-derived DH progeny and those obtained through other breeding methods has yet been reported in rice. They compared the yielding ability of lines developed by the pedigree method and AC and concluded based on field performance, that both methods are equally capable of producing desirable homozygous lines.

Studies in rice on qualitatively inherited characters indicated that the segregation observed within anther-derived DH lines conformed to expected Mendelian ratios, suggesting that gametic selection does not occur (Chen *et al.*, 1982; Chen *et al.*, 1983). These findings are in contrast with those

of Guiderdoni *et al.* (1989a,b), who analyzed the segregation pattern of 12 isozyme genes among DH lines derived from a *japonica* x *indica* cross. In their study, four of 12 isozyme markers showed distorted segregation. When compared to F2 plants derived from the same cross, they found two instances in which the distortion occurred specifically in AC-derived materials. This suggested that "embryogenic" microsporal populations did not represent a random gametic array. The apparent discrepancy between these results and the random assortment patterns observed by Chen and co-workers (Chen *et al.*, 1982; 1983), could be explained by the use of intra-*japonica* crosses and that only two morphological markers were employed in their studies. The existence of segregation distortion among microspore-derived plants have been also reported in other crops like barley (Foroughi-Wehr and Friedt, 1984; Powell *et al.*, 1986), broccoli (Orton and Browers, 1985), and pearl millet (Ha and Pernes, 1982).

Two other instances of aberrant segregation in DH derived lines were detected where distortion was also observed in the F2 progeny of the same cross, suggesting that it was not directly related to the mechanisms of androgenesis (Guiderdoni *et al.* 1989a,b). The cause was more likely related to the gametic selection and certation which occur in hybrids between distantly related cultivars. This observation was confirmed subsequently, when studying the segregation patterns of five other *japonica/indica* crosses.

In general, Guiderdoni (1991) found no significant differences among the allelic frequencies calculated from the F2 and the nonmorphogenic calli and AC populations derived from the same cross. This suggested that the mechanisms of microspore selection occurring during gametogenesis, was responsible for the skewed segregation in the F2 progeny.

Nevertheless, in one cross, segregation patterns of five isozyme loci that deviated in the F2 did fit the expected 1:1 ratio in the AC-derived plants. A similar response was observed in a previous study involving another cross (Guiderdoni, 1989). In this instance, the F2 progenies also displayed distortions at five isozyme loci, while the segregation of the AC-derived lines from the same cross did fit the expected 1:1 ratio at all loci surveyed. This suggests that AC may, in some situations, bypass a phenomenon that appears related to sterility observed in F1's from distantly related parents which hinders the conventional exploitation of rice in wide crosses.

Another factor that will affect the genetic variation exhibited by DH lines derived from distantly related parents is changes induced by the technique or protocol. In such progenies the *in vitro* culture phase may induce genetic variation over and above that derived from recombination and segregation of the parental genomes. Results from evaluation of the phenotypic performance of AC-derived lines in barley (Powell *et al.*, 1984) and in wheat (Baenziger *et al.*, 1983, 1989) suggest that AC does induce "gametoclonal variation".

It is, genetic variability brought about by *in vitro* culture of gametic tissue (Evans *et al.*, 1984). Such variation usually appears to be detrimental to performance for yield traits and does not appear to be useful for breeding purposes. It is important to establish whether the AC system in rice likewise induces such genetic variation in DH progenies. Two blast resistant variants were obtained through AC by Zheng and associates in China, in 1985 (Zhang, 1989). Likewise gametoclonal variation in rice resulted in short stature variants (Schaeffer, 1982), and lines with increased seed storage proteins (Schaeffer *et al.* (1984), and grain chalkiness (Schaeffer *et al*, 1986).

MATERIALS AND METHODS

Two upland-dwarf rice lines: WC 5103 and WC 5121, obtained from the Centro Internacional de Agricultura Tropical (CIAT)'s upland rice program, were crossed to each of two irrigated rice cultivars : CICA-8 and Oryzica-1. The four single crosses generated are identified by the letters **E**, **F**, **G**, and **H** (Appendix Table 1).

WC 5103 and WC 5121 are improved upland lines with glabrous leaves, early maturity, and low grain amylose content. WC 5103 has a moderate response to anther culture (AC), while WC 5121 is the most responsive genotype that CIAT posses to date. CICA-8 and Oryzica-1 are modern, high yielding cultivars with pubescent leaves, intermediate maturity, intermediate amylose content, and very low response to AC.

Five sets of progenies denoted as: **F1DH**, **F2DH-R**, **F2DH-S**, **SSD-R**, and **SSD-S**, were generated from each cross. **F1DH**, **F2DH-R**, and **F2DH-S** were composed of doubled haploid (DH) lines generated by *in vitro* AC, using the methodology developed at CIAT (Pulver, 1986). **F1DH** progeny was derived from the F1 of each cross, while, **F2DH-R** and **F2DH-S** were obtained from the F2 generation. **F2DH-R** DH lines originated from F2 plants collected at random from each cross, with the **F2DH-S** DH lines obtained from F2 plants selected for early maturity, leaf pubescence and a semidwarf plant type with erected leaves and high tillering ability. **SSD-R** and **SSD-S**

progenies were constituted from F4 lines developed using a single seed descend (SSD) procedure (Brim, 1966). SSD-R lines were derived from unselected (random) F2 plants. The **SSD-S** were composed of F4 lines arising from F2 plants selected for earliness, presence of leaf pubescence and plant type. The procedure used to generate these five populations is provided in Appendix Figure 1.

Selection of female plants and hybridization was initiated in September 1988, at CIAT in Palmira, Colombia. In order to ensure the genetic purity of the original parental material, only one plant per line or cultivar was chosen to be used as a parent. These plants were protected with glycine bags before flowering to avoid cross pollination. At the time of panicle emergence, panicle-bearing tillers were excised from the selected female plants, and their leaves removed to reduce respiration. The panicles were then emasculated after removing apical and basal spikelets. The prepared panicles were enclosed in glycine bags and placed in fresh water for 1-2 days, pollinated with freshly collected pollen, rebagged, and placed back in fresh water until maturity. The hybrid seed produced was then harvested and stored in envelopes until planting. Some panicles were allowed to self pollinate in order to maintain a seed source of the parental plants.

Spikelet sterility of 10 F1 plants was evaluated for each cross. The panicle of the main culm of each plant was harvested and placed in a separate envelope shortly before

physiological maturity to minimize shattering. Percentage of spikelet sterility was calculated on the basis of the percentage of empty spikelets on the total number of spikelets per panicle.

Production of doubled haploid lines.

Seed dormancy was broken by maintaining seed at 50°C for 5 days. Staggered planting of the F1 was done from October through December 1988 to obtain a longer period of flowering. This permitted continous processing of harvested anthers without exceeding the capacity of the anther culture laboratory. The F1 seed was planted in sterilized soil in plastic trays in the screen house. After 30 to 35 days the F1 seedlings were then transplanted to the field using a spacing of 30 cm. between plants and 40 cm. between rows. This spacing allowed for optimum growth and facilitated the selection and harvesting of young panicles for anther culture.

Harvesting of panicles for anther culture started when the plants reached the booting stage, about 60-70 days after transplanting. A distance of 4-8 cm. between the auricles of the two last leaves was used as a criterion to select panicles. This morphological criterion allowed for harvesting of panicles in the field which had anthers with pollen grains in the late-uninucleate-to-early-binucleate stage. This has been found to be the most responsive stage of

pollen development for anther inoculation (Nuñez *et al.*, 1989). An additional morphological criterion used was the color (greenish yellow) and firmness (fragile) of flowers before the plating of anthers.

After harvesting, panicles still within their sheath leaves were sterilized in 70% alcohol for 1 minute. Sheaths were then removed and the top and bottom portions of the panicle were discarded leaving approximatedly 25 florets per panicle. Prior to plating of anthers, panicles were again sterilized in a 2% solution of calcium hypochlorite with Tween 80 (3 drops per 100 ml. of solution), as a dispersant. They were allowed to soak for 3 minutes and then washed in a series of 4-5 deionized, distilled, sterile water baths. Florets were then removed from the panicles at their base. Anthers were transferred to the induction medium by tapping cut flowers on the perimeter of the induction flask containing 10 ml. of potato extract liquid medium (Chuang *et al.*, 1978; Chen, 1986), which included 4 ppm of naphtaleneacetic acid (NAA) and 1 ppm. of kinetin as exogenous hormones (Appendix, Table 2). All procedures were conducted under a laminar flow hood using sterile conditions. Approximately 200-250 anthers were planted per flask, sealed and placed in the dark at 8°C for 10-15 days, then incubated at 25°C in the dark for about 40-45 days. Under these conditions microspores multiplication occurred leading to microcalli formation. Such formations started to appear about 20 days after culture planting, and proliferated for

35-45 days reaching the optimal size to be transferred to the regeneration medium at about 40-45 days.

Calli were transferred to a Gelrite-solidified MS regenerative medium (Murashige and Skoog, 1962), containing 1 ppm NAA + 4 ppm kinetin, and incubated under 16 hour photoperiod time (2500 lux, provided by Sylvania® cool-white fluorescent lights), for approximately 30 days. Thereafter, green pollen plantlets having developed roots and shoots were removed manually from the flasks, washed with water to remove calli and medium residues. Their roots were then submerged in water for 1-2 days. Subsequently, the regenerated green plantlets were taken to the screen house where they were tranferred to plastic trays containing water-saturated sterilized soil. No treatment was applied to double the chromosome number of haploid individuals. After 8 days plantlets were fertilized with 15 cc of a mixture containing 30% N, 7% P_2O_5, and 6% K, diluted in 4 g/l of water. After two or three weeks, depending on their growth, the plantlets were transplanted to the field were they received the standard agronomic practices for irrigated rice (*see* Tascón and García, 1985). The production of doubled haploid plants in this work followed the anther culture methodology developed in the Rice Program at CIAT to handle a reasonably high number of crosses required in a rice breeding program (Pulver, 1986). Further details of this methodology have been described elsewhere (Nuñez *et al.*, 1989).

Each regenerated R1 plant that reached maturity in the field was assigned an identification number. At full maturity, data of the number of haploid, spontaneous doubled haploids, and fully sterile plants were taken. Doubled haploid R1 plants were then harvested individually. The seed of each R1 plant, became a R2 line. Doubled haploid seed was increased during the 1989-1990 season in Vista Florida Agricultural Station at Chiclayo, Perú. R3 seed harvested there constituted the **F1DH** population. Sixty one **F1DH** lines were generated from cross **E**, 104 from cross **F**, 81 from **G**, and 117 from **H**.

F2 seed from the selfed F1 plants was planted in plastic trays in Palmira in April 1989, and transplanted the following month in 10 m.-rows with 0.40 m. between rows, with 0.30 m. between plants. At the proper stage of pollen development, young panicles were taken from about 500 F2 plants per cross. Each panicle was identified as to the original mother plant, and processed following the AC procedures described above. R1 plants and eventually R2 doubled haploid lines developed this way formed the **F2DH-R** population. Ninety five DH lines were developed from cross **E**, 41 from cross **F**, 66 from cross **G**, and 69 from cross **H**.

Approximately 800 F2 plants per cross were seeded in the screen house in May 1989 in Palmira, and transplanted one month later in the field in 10 m. rows with 0.30 m. between plants and 0.40 m. between rows. **F2DH-S** lines were developed through AC from about 500 hundred F2 plants selected for

early maturity, leaf pubescence, and plant type. Eighty three **F2DH-S** lines were produced from cross **G**, and 105 from cross **H**. No **F2DH-S** lines were produced from crosses **E** and **F** due to their low response to anther culture.

Production of SSD lines.

About 1500 F2 plants per cross were planted in May 1989, at Palmira. Transplanted plants were arranged in 30 rows per cross with spacings of 0.40 m. between rows and 0.30 m. between plants. One panicle was harvested from about 600 plants chosen at random from the first 15 rows (unselected group). One panicle per plant was also harvested from about 300 plants selected from the remaining 15 rows for earliness, presence of leaf pubescence and plant type (selected group). At the CIAT seed laboratory in Palmira, one seed per panicle was taken to form two F3 seed bulks, one per group, to be planted the next season.

To obtain the F4 generation, both F3 bulks were planted under irrigated conditions in November 1989, in 'Vista Florida' Experimental Station at Chiclayo (Perú). At maturity each plant was harvested individually. Four hundred F4 lines per cross were chosen at random from the unselected group to form the **SSD-R** progeny from each cross. Two hundred F4 lines derived from selected F2 plants originated the **SSD-S** population of each cross. No **SSD-S** lines were produced from cross **E**.

Experimental design.

Progenies (treatments), developed by the procedures described above, were arranged in a generalized randomized block design (Wilk, 1955; Steel and Torrie, 1981) with 27 blocks. A random sample of lines from each progeny was randomly allotted to the plots of each block. The number of lines per progeny per block was proportional to the total number of lines per progeny (unequal but proportional sub-class numbers). Thus, each block was a replication of the whole experiment. Each block consisted of 125 plots (experimental units) of three rows with seven plants per row, having 0.30 m. between plants and between rows.

This experiment was planted in two locations: Huarangopampa Experimental Station at Bagua (Perú), and Palmira (Colombia).

Huarangopampa Experimental Station is located in the Province of Bagua Chica, Department of Amazonas (Marañón's North-eastern Region). It is located in an important irrigated rice growing area in the eastern slopes of the Peruvian Andes. It is geographically situated at longitude 78°22'W, and latitude 5°35'S, at an elevation of 500 m. above sea level. The annual temperature mean is about 27°C, with an annual rainfall of approximately 1200 mm. A summary of climatic data on a per month basis is given in the Appendix Table 3.

Palmira is located in the Department of Valle del Cauca, a very important agricultural region between the western and central ranges of the Colombian Andes. It is situated at 3°32' latitude N, and 76°16' longitude W ; at an elevation of 1030 m. above sea level. The annual rainfall is about 1000 mm., and 24-26°C is the annual mean temperature. Climatic data collected during the growing season in this location is provided in Appendix Table 3.

In Huarangopampa, the experimental materials were planted in seedbeds in July 13, 1990. All entries were transplanted on August 22, following the experimental design described above. Plots were hand-weeded during the season. Fertilization and management practices were those recommended for irrigated rice at this location. Plants were harvested in January 1991.

The experimental material was planted July 9, 1990 and transplanted August 13 at the Palmira site. Agronomic practices were the standard for irrigated rice. Plants were harvested in December 1990.

Traits measured.

Leaf pubescence was evaluated by detecting with the fingers the presence of leaf hairs in the upper side of the leaf. The total number of plants per plot was evaluated for the presence (pubescent) or absence (glabrous) of hairs on the

leaf and expressed as the ratio of number of pubescent plants/total number of plants.

Days to maturity was obtained calculating the number of days from planting to the maturity date. Maturity date was recorded when more than 80% of plants within the plot had reached physiological maturity as evidenced by the color and hardness of the grain. At this stage most grains have turned yellow and hard, and the grains in the lower portion of the panicle are in the hard dough stage.

Number of panicles per square meter was determined by counting the number of panicles on each of five plants in the center of the plot.

Number of grains per panicle was determined on 10 randomly chosen panicles from the five plants in the center of the plot, and expressed as an average.

Spikelet sterility was determined from the ratio of number of empty grains to the total number of grains per panicle, multiplied by 100, and taken as the average of 10 panicles per plot.

1000-grain weight was determined by weighing a random sample of 1000 grains from 10 harvested panicles.

Plant height was obtained by measuring the length of the tallest tiller, from the base of the culm to the tip of panicle, on five plants located in the center of the plot. This trait was evaluated only at the Huarangopampa site.

Endosperm dispersion (gelatinization temperature) was estimated by the extent of the spreading and clearing of milled rice treated with a 1.7% solution of potassium hydroxide for 23 hours at 30°C. A numerical scale of seven digits was used to visually rate the appearance and disintegration of the endosperm, from non-affected kernels (rated 1) to completely dispersed and intermingled kernels (rated 7). A rating of spreading of 1-3 is classified as high, 4-5 as intermediate, and 6-7 as low gelatinization temperature. Gelatinization temperature is partly associated with the amylose content of the starch, the major determinant of cooking quality (Jennings *et al.*, 1979). This trait was evaluated only at Palmira.

Statistical analysis.

An analysis of variance was employed to test if significant differences existed among locations and progenies for the traits studied. The size of the two-locations matrix of data was too big to be analyzed through an appropriate statistical procedure (e.g. SAS's GLM) by a conventional mainframe computer. Therefore, a combined analysis of

variance was performed using the SAS's ANOVA procedure which, even though is not well suited for unbalanced designs, allowed us to estimate location x progeny interactions.

A second analysis of variance was computed for each location to test if significant differences existed among crosses, generations (F1- vs F2-derived progenies), methods (DH vs SSD, in F2-derived progenies), and selection procedures (progenies derived from selected vs non-selected F2 plants). A general linear model (GLM) procedure was used to perform the analysis of variance for each trait. This procedure allows the data to be analyzed in unbalanced situations, i.e., models where there are unequal number of observations for different combinations of variables (Bancroft, 1968; SAS, 1987).

Furthermore, the following set of contrasts was used to test specific comparisons of interest:

1. F1DH vs F2DH-R;
2. F2DH-R vs F2DH-S;
3. F2DH-R vs SSD-R;
4. SSD-R vs SSD-S;
5. F1DH vs SSD-R.

Contrast 1 used data from all four crosses. Contrasts 2 to 5 were obtained with progenies derived from crosses G and H.

The tests of hypothesis used the Type III mean square (see SAS, 1987) for Block x Progeny as an error term. Even though there were unequal sub-class numbers, this error term

was nearly the same as the appropriate error term obtained by the Satterthwaite's method (Satterthwaite, 1946; Snedecor and Cochran, 1980). This may be accounted for the proportional distribution of the number of lines per progeny (sub-class numbers) in each block in this experiment.

Homogeneity of variances between progenies was determined using the Levene's test (Levene,1960). As a measure of the variation within a class, the Levene's test uses the average of the absolute deviations instead of the mean square of the deviations. This avoids squaring and makes the criterion much less sensitive to long tailed distributions (Snedecor and Cochran, 1980).

Chi-square tests were used to estimate differences in frequency of desirable recombinants among the populations studied in this research.

RESULTS

Base experimental populations from which the doubled haploid and single seed descent lines were derived consisted of six parents and the resulting F1's. In Table 1, the percentage of spikelet sterility of the parents and F1's are provided. High levels of spikelet sterility were observed for the F1's and their respective parents. Cross H had 48.9 % sterile spikelets in contrast to cross G with only 33.9 % sterility. Among the parents, both upland cultivars (WC 5103 and WC 5121) had more sterility than the irrigated cultivars [CICA-8 (E), Oryzica-1 (F), CICA-8 (G), and Oryzica-1 (H)].

Results from field trials which involved parents and 16 derived populations of four upland/irrigated rice crosses obtained through either DH or SSD breeding methods and planted in two locations, Bagua (Perú) and Palmira (Colombia) will be presented in four sections: (1) combined analysis of variance for spikelet sterility, leaf pubescence, days to maturity, number of grains per panicle, number of panicles per square meter, and thousand grain weight; (2) analysis of variance for each location, addressing the question of the effect of cross, generation, method, and selection on the performance of the populations for the traits studied; (3) contrasts comparing specific populations of interest across crosses; and (4) comparison of the proportion of recombinant lines produced from DH and SSD populations.

Table 1. Mean percentages and standard deviations for spikelet sterility of the F1 and respective parents of four upland/irrigated crosses planted in Palmira (Colombia), 1989.

Genotypes	$\bar{x}$	s.d.
F1 's		
WC 5103/CICA-8(E)	39.3	9.6
WC 5103/Oryzica-1(F)	34.2	8.1
WC 5121/CICA-8(G)	33.9	7.9
WC 5121/Oryzica-1(H)	48.9	17.6
Parents		
CICA-8(E)	11.1	4.3
Oryzica-1(F)	13.1	5.2
CICA-8(G)	12.2	4.6
Oryzica-1(H)	14.3	6.5
WC 5103	22.3	7.9
WC 5121	17.3	5.2

Combined analysis of variance.

Evaluation of parents. Observed mean squares among the parents for the traits measured are provided in Appendix Table 4. Differences were found for all the traits. No significant genotype x location interaction was found for leaf pubescence, days to maturity and number of grains per panicle. A comparison of the parental mean values for these traits is presented in Table 2. CICA-8 (G), an irrigated cultivar, was the latest maturing parent, and WC 5121, an upland rice line, was the earliest. Irrigated rice genotypes (both CICA-8's and both Oryzica-1's) showed higher number of grains per panicle than the upland types. Significant genotype x location interactions were found for spikelet sterility, number of panicles per square meter, and 1000-gr. weight (Appendix Table 4). As noted in Table 3, high levels of spikelet sterility were detected for the parents, with WC 5103 having the highest average percentage for the two locations. Irrigated cultivars yielded a higher number of panicles per square meter than the upland types at both locations. Both CICA-8's (E and G) had the lowest thousand grain weight, regardless of location.

Evaluation of populations. Observed mean squares for six traits from the combined analyses of variance for the 16 populations grown at two locations are given in Appendix

Table 2. Combined mean values for leaf pubescence, days to maturity and number of grains per panicle, of six parents grown at Bagua (Perú), and Palmira (Colombia), 1990.

Parent	Means		
	Leaf pubescence¶	Days to maturity	Grains/ panicle
CICA-8 (E)	1.0 **a**†	150.7 **n**	136.2 **w**
CICA-8 (G)	1.0 **a**	152.7 **m**	124.7 **x**
Oryzica-1 (F)	1.0 **a**	148.7 **p**	123.1 **x**
Oryzica-1 (H)	1.0 **a**	149.7 **np**	118.5 **x**
WC 5121	0.0 **b**	140.9 **q**	93.2 **z**
WC 5103	0.0 **b**	149.7 **np**	110.0 **y**

¶ proportion of pubescent plants in the plot.
† Fisher's (protected) LSD test; means with a common letter in the same column are not significantly different at the 0.05 probability level.

Table 3. Mean values for spikelet sterility (%), number of panicles per square meter and 1000-gr. weight of six parents grown at Palmira (Colombia), and Bagua (Perú), 1990.

Parents	Means								
	Spikelet sterility (%)			Panicles per m^2			1000-gr. weight		
	Palmira	Bagua	$\bar{x}$	Palmira	Bagua	$\bar{x}$	Palmira	Bagua	$\bar{x}$
CICA-8 (E)	15.7	13.2	14.4 d†	222.7	172.6	197.6 m	23.0	23.0	23.0 z
CICA-8 (G)	16.8	15.3	16.1 cd	203.0	181.5	192.3 mn	23.4	22.9	23.2 z
Oryzica-1 (F)	16.2	21.6	19.5 b	209.1	152.7	173.7 p	25.6	26.5	26.2 x
Oryzica-1 (H)	18.2	18.4	18.3 bc	192.6	172.4	182.3 np	24.5	25.6	25.1 y
WC 5121	22.6	13.9	18.3 bc	157.3	129.1	142.7 q	26.4	26.3	26.4 x
WC 5103	36.5	20.1	28.0 a	175.0	136.8	155.5 q	24.8	27.4	26.1 x

† Fisher's (protected) LSD test; means with a common letter in the same column are not significantly different at the 0.05 probability level.

Table 5. Differences were observed among populations for all the traits. Differences were detected between locations for spikelet sterility, number of grains per panicle and number of panicles per square meter. Progeny x location interactions were found to be significant for days to maturity, number of grains per panicle and 1000-gr. weight.

Coefficients of variation were low for leaf pubescence (near to zero), days to maturity (1.7), and 1000-gr. weight (4.0); intermediate for panicles per square meter (15.2) and high for number of grains per panicle (22.9), and spikelet sterility (33.7).

Single location analyses of variance.

Observed mean squares for the traits measured in Bagua and Palmira are presented in Appendix Tables 6 and 7, respectively. Results for the traits plant height and endosperm dispersion, which were evaluated only at one location, are also included in their respective Tables. Differences were found among populations for all traits at both locations. The population effect was partitioned by cross, generation, method, and selection effects. In Bagua, blocking was found to be effective in accounting for field variability for all traits, except leaf pubescence. Blocking was also effective in reducing the variability for most traits under evaluation in Palmira, except for spikelet sterility, leaf pubescence, and number of grains per panicle.

Percentage of spikelet sterility. Mean values and standard deviations for percentage of spikelet sterility at Bagua and Palmira locations, are provided in Appendix Tables 8 and 9, respectively. Differences were found among crosses, but not between generations, at either location (Appendix Tables 6 and 7). Cross x generation interactions were significant only at Palmira (Table 4). A significant difference between methods was observed only at Bagua (Appendix Table 6). Cross x method interaction was detected at both locations. Doubled haploid-derived populations tended to have a higher percentage of spikelet sterility than SSD populations at both locations. The one exception was the cross F at Palmira (Table 5). The response to selection of the populations studied was significant. No cross x selection interaction was observed at either site (Appendix Tables 6 and 7). At both locations, populations derived from selected F2 plants had a lower percentage of spikelet sterility than those derived from randomly selected F2 plants (Table 6).

Proportion of pubescent plants. Mean values and standard deviations of populations within crosses for pubescence are provided in Appendix Tables 10 and 11, for Bagua and Palmira, respectively. There were no significant differences among crosses for this trait, but differences among generations were noted at both locations (Appendix Tables 6 and 7). Cross x generation interaction was non-significant at both sites, making it possible to assess the overall effect of

Table 4. Mean values and standard deviations for percentage of spikelet sterility of rice progenies derived from two generations, planted at Bagua (Perú), and Palmira, (Colombia), 1990.

	Crosses[1,2]							
Generation	E		F		G		H	
Bagua	27.7	(13.2)	27.8	(11.4)	26.2	(12.3)	29.9	(14.3)
	ab[†]		**ab**		**b**		**a**	
Palmira								
F1-derived	39.0	(15.8)	44.7	(19.0)	38.8	(23.6)	46.0	(21.0)
F2-derived	47.0	(20.7)	39.2	(18.6)	42.1	(20.9)	42.7	(20.5)
sgn[††]	*		*		ns		ns	

[1] E = WC 5103/CICA-8(E); F = WC 5103/Oryzica-1(F); G = WC 5121/CICA-8(G); H = WC 5121/Oryzica-1(H).

[2] differences among crosses were significant at the 0.01 probability level, at both locations; cross x generation interaction was significant at the 0.01 probability only at Palmira.

[†] means with a common letter are not significantly different at the 0.05 probability level.

[††] significance of difference between means within the same column.

Table 5. Mean values and standard deviations for percentage of spikelet sterility of rice progenies derived from four crosses and developed through either doubled haploid (DH) or single seed descent (SSD) breeding methods, planted at two locations, Bagua and Palmira, 1990.

	Crosses[1,2]							
Method	E		F		G		H	
Bagua								
DH	31.3	(16.6)	30.6	(13.4)	28.9	(13.1)	34.6	(16.5)
SSD	27.0	(12.0)	27.0	(11.0)	25.3	(11.6)	28.1	(12.7)
sgn[†]	**		ns		**		**	
Palmira								
DH	47.0	(20.7)	38.6	(18.1)	45.5	(23.0)	44.8	(23.1)
SSD	--.--		39.3	(18.7)	41.3	(20.3)	42.1	(19.7)
sgn			ns		*		ns	

[1] E = WC 5103 / CICA-8(E); F = WC 5103 / Oryzica-1(F); G = WC 5121 / CICA-8(G); H = WC 5121 / Oryzica-1(H).

[2] difference between methods was significant at the 0.01 probability level at Bagua; cross x method interaction was significant at the 0.05 probability level at both locations.

[†] significance of difference between means within the same column.

generation on population performance for this trait. At both locations, F2-derived populations had a higher proportion of pubescent plants than F1-derived populations (Table 7). Differences were also noted for breeding method and cross x method interactions (Appendix Tables 6 and 7). There was a higher proportion of pubescent plants in SSD than in DH populations from crosses G and H, both having the same female parent (Table 8). In Table 9, comparisons of the effect of selection and selection x cross interaction are made for proportion of pubescent plants at Bagua and Palmira. Selection was found to be significant at both locations, while cross x selection interaction was only significant at Palmira (Appendix Tables 6 and 7). Populations derived from selected F2-plants had higher proportion of pubescent plants (0.70) than populations derived from non-selected F2-plants (0.63) at Bagua. Same tendency was observed in Palmira, although differences were significant only for cross G (Table 9).

Days to maturity. Mean values and standard deviations for this trait are provided in Appendix Tables 12 and 13, for Bagua and Palmira, respectively. As previously mentioned, a highly significant population x location interaction was found for this trait (Appendix Table 5). In Bagua, no statistical difference was detected for crosses and cross x generation interaction; however, differences between generations were found to be significant (Appendix Table 6).

Table 6. Mean values and standard deviations for percentage of spikelet sterility of rice progenies derived either from random or selected F2 plants, grown at two locations, Bagua and Palmira, 1990.

	Bagua		Palmira	
	$\bar{x}$	s.d.	$\bar{x}$	s.d.
Random	28.1	12.7	42.6	20.0
Selected	27.3	13.0	40.0	19.0

selection effect was significant at the 0.05 and 0.01 probability level at Bagua and Palmira, respectively.

Table 7. Mean values and standard deviations for the proportion of pubescent plants of rice progenies derived from two filial generations and planted at two locations, Bagua (Perú), and Palmira (Colombia), 1990.

	Bagua		Palmira	
Progenies	$\bar{x}$	s.d.	$\bar{x}$	s.d.
F1-derived	0.53	0.47	0.52	0.48
F2-derived	0.65	0.42	0.65	0.43

mean differences between generations were significant at the 0.01 probability level, at both locations.

Table 8. Mean values and standard deviations for the proportion of pubescent plants of rice progenies derived from the F2 of four upland/irrigated crosses either through doubled haploid (DH) or single seed descent (SSD) breeding methods, planted at two locations, Bagua (Perú), and Palmira (Colombia), 1990.

	Crosses[1,2]							
Method	E		F		G		H	
Bagua								
DH	0.57	(0.48)	0.68	(0.44)	0.53	(0.49)	0.48	(0.48)
SSD	0.66	(0.41)	0.62	(0.42)	0.73	(0.38)	0.69	(0.38)
sgn[†]	*		ns		**		**	
Palmira								
DH	0.56	(0.48)	0.68	(0.45)	0.53	(0.49)	0.48	(0.47)
SSD	-.-		0.61	(0.43)	0.76	(0.38)	0.66	(0.41)
sgn			ns		**		**	

[1] E = WC 5103 / CICA-8(E); F = WC 5103 / Oryzica-1(F); G = WC 5121 / CICA-8(G); H = WC 5121 / Oryzica-1(H).

[2] differences between methods and cross x method interaction were significant at the 0.01 probability level, at both locations.

[†] significance of difference between means within the same column.

Table 9. Mean values and standard deviations for proportion of pubescent plants of rice progenies derived from either random or selected F2-plants, planted at Palmira (Colombia), 1990.

	Crosses[1,2]			
	E	F	G	H
Random	0.56 (0.48)	0.62 (0.43)	0.66 (0.43)	0.60 (0.43)
Selected	-.-	0.59 (0.43)	0.80 (0.36)	0.65 (0.42)
sgn[†]		ns	**	ns

[1] E = WC 5103 / CICA-8(E); F = WC 5103 / Oryzica-1(F); G = WC 5121/CICA-8(G); H = WC 5121 / Oryzica-1(H).

[2] selection effect was significant at the 0.05 and 0.01 probability level at Bagua and Palmira, respectively. Cross x selection interaction was significant at the 0.05 probability level, at Palmira.

[†] significance of difference between means within the same column.

F2-derived populations had a shorter maturation period (146.8 days) than F1-derived populations (147.5 days). In Palmira, differences were noted for crosses and cross x generation interaction (Appendix Table 7). In cross E, F2-derived populations matured earlier than the F1-derived populations (there were only two populations from this cross and both were DH), while in cross F, F1-derived had an earlier maturation period than the F2-derived populations. Populations from crosses G and H showed no differences among generations at this location (Table 10). There were no significant differences in maturity due to method, or cross x method interaction among populations planted at Bagua and Palmira (Appendix Tables 6 and 7). A significant cross x selection interaction for maturity was observed at Bagua (Appendix Table 6). In cross H, populations derived from selected F2 plants matured earlier than those derived from randomly selected F2 plants. However, no significant differences were observed in crosses F and G (Table 11). Nevertheless, in Palmira, where the effect of selection was found to be significant (Appendix Table 7), populations derived from random F2 plants matured earlier (146.8 d.) than those from selected F2 plants (147.1 d.).

Number of grains per panicle. A significant location x populations interaction for number of grains per panicle was detected (Appendix Table 5). Mean values and standard deviations for this trait are provided in Appendix Tables 14

Table 10. Mean values and standard deviations for days to maturity of rice progenies for two generations, planted at Palmira (Colombia), 1990.

	Crosses[1,2]			
Generation	E	F	G	H
F1-derived	147.3 (7.6)	145.8 (8.4)	147.5 (7.6)	146.0 (8.6)
F2-derived	144.0 (9.1)	147.8 (7.5)	147.9 (7.4)	145.7 (8.3)
sgn†	**	**	ns	ns

[1] E = WC 5103 / CICA-8(E); F = WC 5103 / Oryzica-1(F); G = WC 5121 / CICA-8(G); H = WC 5121 / Oryzica-1(H).

[2] differences between generations were significant at the 0.05 probability level at Bagua; cross effect and cross x generation interaction were significant at the 0.01 probability level at Palmira.

† significance of difference between means within the same column.

Table 11. Mean values and standard deviations for days to maturity of rice progenies derived either from random or selected F2 plants, planted at Bagua (Perú), 1990.

	Crosses[1,2]			
	E	F	G	H
Random	147.7 (6.6)	148.7 (6.7)	146.5 (6.1)	146.7 (5.9)
Selected	--.--	147.2 (5.8)	146.7 (6.6)	145.7 (6.3)
sgn†		ns	ns	*

[1] E = WC 5103 / CICA-8(E); F = WC 5103 / Oryzica-1(F); G = WC 5121 / CICA-8(G); H = WC 5121 / Oryzica-1(H).

[2] cross x selection interaction was significant at the 0.05 probability level at Bagua; selection was significant at the 0.05 probability level at Palmira.

† significance of difference between means within the same column.

and 15, for Bagua and Palmira, respectively. Differences among crosses, generations, and for the cross x generation interaction were significant at Bagua (Appendix Table 6). F2-derived populations tend to have higher mean values than F1-derived populations. This tendency was noted for all crosses except cross E in Bagua (Table 12). However, in Palmira, where only the cross x generation interaction was found to be significant, no differences were found between generations. The exception was the cross E where F1-derived DH lines showed higher number of grains per panicle than F2-derived populations (Table 12). The effect of breeding methods was found to be significant at Bagua, while cross x method interaction was detected at Palmira (Appendix Tables 6 and 7). Single seed descent populations had higher number of grains per panicle (102.6) than DH populations (90.0), at Bagua. However, in Palmira this response was detected only for cross G (Table 13). No selection effect was found at Bagua, while cross x selection interaction was detected at Palmira (Appendix Table 6 and 7). In cross F, progenies derived from selected F2 plants had higher mean values than those derived from random F2 plants. No significant differences were found for crosses G and H (Table 14).

Thousand-grain weight. The population x location interaction was significant for this trait (Appendix Table 5). Mean values and standard deviations of the population performance

Table 12. Mean values and standard deviations for number of grains per panicle of rice progenies of four crosses derived for two generations, grown at Bagua and Palmira, 1990.

	Crosses[1,2]			
Generation	E	F	G	H
Bagua				
F1-derived	101.5 (25.7)	95.9 (27.1)	88.7 (27.2)	88.1 (32.4)
F2-derived	106.8 (28.9)	103.3 (26.1)	101.7 (28.0)	97.3 (29.0)
sgn[†]	ns	*	**	**
Palmira				
F1-derived	79.2 (28.8)	69.4 (25.9)	78.4 (40.8)	71.2 (33.6)
F2-derived	61.9 (28.8)	74.3 (29.0)	71.9 (32.0)	70.6 (31.7)
sgn	**	ns	ns	ns

[1] E = WC 5103 / CICA-8(E); F = WC 5103 / Oryzica-1(F); G = WC 5121 / CICA-8(G); H = WC 5121 / Oryzica-1(H).

[2] generation effect and cross x generation interaction were significant at the 0.05 probability level at Bagua; cross x generation interaction was significant at the 0.01 probability level at Palmira.

[†] significance of difference between means within the same column.

Table 13. Mean values and standard deviations for number of grains per panicle of rice progenies of four upland/irrigated rice crosses derived either from doubled haploid (DH) or single seed descent (SSD) methods, grown at Palmira, 1990.

	Crosses[1,2]			
Method	E	F	G	H
DH	61.9 (28.8)	80.4 (29.5)	66.9 (35.3)	68.9 (34.8)
SSD	--.--	73.8 (28.9)	73.1 (31.1)	71.1 (30.7)
sgn[†]		ns	*	ns

[1] E = WC 5103 / CICA-8(E); F = WC 5103 / Oryzica-1(F); G = WC 5121 / CICA-8(G); H = WC 5121 / Oryzica-1(H).

[2] difference between methods was significant at the 0.01 probability level at Bagua; cross x method interaction was significant at the 0.01 probability level at Palmira.

[†] significance of difference between means within the same column.

for this trait can be found in Appendix Tables 16 and 17, for Bagua and Palmira, respectively. Differences were noted among crosses in Bagua and Palmira (Appendix Table 6 and 7). Crosses E and G had lower 1000-gr weight than F and H crosses at Bagua (Table 15). This appears to be associated with their common parent, CICA-8, which has the lowest 1000-gr weight (Table 3). A significant cross x generation interaction was detected in Palmira (Table 15). No significant effect of generation was noted either at Bagua or Palmira (Appendix Tables 6 and 7). Differences between breeding methods were detected at both locations, while cross x method interaction was only found to be significant at Palmira (Appendix Tables 6 and 7). Higher mean values were registered for SSD populations (25.5 g.) as compared to DH populations (23.8 g.), at Bagua. The same pattern was observed in all crosses at Palmira, although significant differences were noted only for cross H (Table 16). Response to selection was not significant at either location, with method x selection interaction being significative at Bagua (Appendix Tables 6 and 7). Doubled haploid populations tended to respond to selection while SSD populations did not (Table 17). This trend was observed at both locations even though the interaction was only significant in Bagua.

Panicles per square meter. No significant location x population interaction was detected for this trait (Appendix

Table 14. Mean values and standard deviations for number of grains per panicle of rice progenies of four upland/irrigated rice crosses, derived from random or selected F2 plants, grown at Palmira, 1990.

	Crosses[1,2]			
	E	F	G	H
Random	61.9 (28.8)	73.3 (29.1)	72.3 (32.9)	68.9 (32.1)
Selected	--.--	84.1 (25.5)	71.2 (30.6)	73.3 (30.9)
sgn†		*	ns	ns

[1] E = WC 5103 / CICA-8(E); F = WC 5103 / Oryzica-1(F); G = WC 5121 / CICA-8(G); H = WC 5121 / Oryzica-1(H).

[2] cross x selection interaction was significant at the 0.01 probability level.

† significance of difference between means within the same column.

Table 15. Mean values and standard deviations for 1000-gr weight (g.) of 16 rice progenies of four upland/ irrigated crosses derived from two generations, grown at Bagua and Palmira, 1990.

Location/ Generation	Crosses[1,2]			
	E	F	G	H
Bagua	24.2 (2.3)	25.5 (2.7)	24.4 (2.7)	25.8 (3.0)
	b[†]	a	b	a
Palmira				
F1-derived	23.3 (2.5)	23.9 (3.0)	24.5 (3.1)	26.9 (3.9)
F2-derived	22.6 (2.9)	24.9 (2.5)	24.7 (2.6)	25.6 (3.5)
sgn[††]	ns	**	ns	**

[1] E = WC 5103 / CICA-8(E); F = WC 5103 / Oryzica-1(F); G = WC 5121 / CICA-8(G); H = WC 5121 / Oryzica-1(H).

[2] differences among crosses were significant at the 0.01 probability level at both locations; cross x generation interaction was significant at the 0.05 probability level, at Palmira.

[†] means with a common letter are not significantly different at the 0.05 probability level.

[††] significance of difference between means within the same column.

Table 16. Mean values and standard deviations for 1000-gr weight (g.) of 16 rice progenies of four upland/irrigated crosses derived through either doubled haploid (DH) or single seed descent (SSD) breeding methods, grown at Palmira, 1990.

	Crosses[1,2]			
Method	E	F	G	H
DH	22.6 (2.9)	23.7 (2.8)	24.4 (3.1)	24.4 (2.8)
SSD	--.--	25.0 (2.5)	24.8 (2.5)	26.0 (2.9)
sgn[†]		ns	ns	**

[1] E = WC 5103 / CICA-8(E); F = WC 5103 / Oryzica-1(F); G = WC 5121 / CICA-8(G); H = WC 5121 / Oryzica-1(H).

[2] differences between methods were significant at the 0.01 probability level at both locations; cross x method interaction was significant at the 0.01 probability level at Palmira.

[†] significance of difference between means within the same column.

Table 4). Mean values and standard deviations are provided in Appendix Tables 17 and 18, for Bagua and Palmira, respectively. Differences among crosses were detected at both Bagua and Palmira, while cross x generation interaction was observed only at Palmira (Appendix Tables 6 and 7). Crosses E and G (CICA-8 as a female parent) had the highest mean values at Bagua, as can be observed in Table 18. In Palmira, F1-derived progenies from cross G had a higher number of panicles/m2 than F2-derived populations, in contrast to cross E where a different response was observed (Table 18). There was no significant effect of breeding method on the population performance for this trait at the locations studied. Cross x method interaction was detected only at Palmira (Appendix Table 7). Mean values of DH populations were higher than SSD populations only for cross H in Palmira (Table 19). Selection effect was not significant at either location; however, a significant cross x selection interaction was observed at Palmira (Appendix Table 7). Table 20 provides the populations response to selection for number of panicles/m2 across crosses in Palmira.

Plant height. Means and standard variations for this trait are provided in Appendix Table 20. A highly significant difference was registered among crosses and between generations (Appendix Table 6). Cross H had the highest mean value; while crosses E and G, both having CICA-8 as a

Table 17. Means and standard deviations for 1000-gr weight (g.) of rice progenies of four upland/irrigated rice crosses developed either through doubled haploid or single seed descent from random and selected F2 plants, grown in Bagua and Palmira, 1990.

	Bagua[1]		Palmira	
	DH	SSD	DH	SSD
Random	23.4 (3.1)	25.4 (2.6)	23.5 (2.8)	25.3 (2.7)
Selected	24.3 (3.0)	25.1 (2.7)	24.6 (3.1)	25.1 (2.7)
sgn[†]	**	ns	**	ns

[1] method x selection interaction was significant at the 0.01 probability level at Bagua.

[†] significance of difference between means within the same column.

Table 18. Mean values and standard deviations for number of panicles per square meter of 16 rice progenies of four upland/irrigated crosses derived from two generations, grown at two locations, Bagua and Palmira, 1990.

Progenies	Crosses[1,2]			
	E	F	G	H
Bagua	168 (35)	159 (34)	163 (38)	158 (37)
	a[†]	**c**	**b**	**c**
Palmira				
F1-derived	186 (40)	201 (45)	210 (43)	200 (50)
F2-derived	209 (58)	193 (43)	197 (49)	197 (50)
sgn[††]	**	ns	*	ns

[1] E = WC 5103 / CICA-8(E); F = WC 5103 / Oryzica-1(F); G = WC 5121 / CICA-8(G); H = WC 5121 / Oryzica-1(H).

[2] differences among crosses were significant at the 0.01 probability level, at both locations; cross x generation interaction was significant at the 0.05 probability level at Palmira.

[†] means with a common letter are not significantly different at the 0.05 probability level.

[††] significance of difference between means within the same column.

Table 19. Mean values and standard deviations for number of panicles per square meter of 16 rice progenies of four upland/irrigated crosses derived through doubled haploid (DH) or single seed descent (SSD) breeding methods, grown at Palmira, 1990.

	Crosses[1,2]			
Method	E	F	G	H
DH	209 (58)	193 (52)	198 (52)	208 (66)
SSD	----	193 (43)	197 (48)	193 (43)
sgn[†]		ns	ns	**

[1] E = WC 5103 / CICA-8(E); F = WC 5103 / Oryzica-1(F); G = WC 5121 / CICA-8(G); H = WC 5121 / Oryzica-1(H).

[2] cross x method interaction was significant at the 0.05 probability level.

[†] significance of difference between means within the same column.

Table 20. Mean values and standard deviations for number of panicles per square meter of 16 rice progenies of four upland/irrigated crosses derived through doubled haploid (DH) or single seed descent (SSD) breeding methods, grown at Palmira, 1990.

	Crosses[1,2]			
Progenies	E	F	G	H
Random	209 (57)	194 (44)	197 (48)	193 (46)
Selection	----	179 (34)	197 (49)	202 (54)
sgn[†]		*	ns	*

[1] E = WC 5103 / CICA-8(E); F = WC 5103 / Oryzica-1(F); G = WC 5121 / CICA-8(G); H = WC 5121 / Oryzica-1(H).

[2] cross x selection interaction was significant at the 0.01 probability level.

[†] significance of difference between means within the same column.

parent, had the lowest mean values (Table 21). Populations derived from the F2 were taller (99.9 cm.) than those derived from the F1 generation (93.8 cm.). Differences between breeding methods were significant, with no cross x method interaction noted (Appendix Table 6). Populations derived through SSD were taller (101.2 cm.) than those derived through AC (95.7 cm.). Significant differences due to selection were detected for this trait (Appendix Table 6). Populations derived from selected F2 plants tend to be shorter than those derived from random F2 plants (Table 22); nevertheless, a highly significant cross x selection interaction was observed for this trait (Appendix Table 6). Progenies of cross H derived from selected F2 plants, had higher mean values than those derived from non-selected F2 plants, with populations from crosses F and G showing no significant response to selection (Table 22). The response to selection seemed to be related in this case to the degree of difference between the parents of each cross (Appendix Table 20). The wider the range between the parents of a cross, the greater the selection effect. There was also a significant method x selection interaction for this trait (Appendix Table 6). Populations developed through AC showed a greater response to selection in the F2, than those developed through SSD (Table 23).

Endosperm dispersion. Mean values and standard deviations for endosperm dispersion (temperature of gelatinization) are

Table 21. Mean values and standard deviations for plant height (cm.) of 16 rice progenies of four upland/irrigated crosses, grown at Bagua, 1990.

	Crosses[1,2]			
	E	F	G	H
$\bar{x}$	97.1**b**[†]	98.5**b**	97.1**b**	102.6**a**
s.d.	11.3	10.9	13.9	15.0

[1] E = WC 5103 / CICA-8(E); F = WC 5103 / Oryzica-1(F); G = WC 5121 / CICA-8(G); H = WC 5121 / Oryzica-1(H).

[2] differences among crosses were significant at the 0.01 probability level.

[†] means with a common letter are not significantly different at the 0.05 probability level.

Table 22. Mean values and standard deviations for plant height (cm.) of 16 rice progenies of four upland/irrigated crosses derived from either random or selected F2 plants, grown at Bagua, 1990.

	Crosses[1,2]			
Progenies	E	F	G	H
Random	97.8 (11.4)	99.6 (10.5)	98.6 (13.3)	105.5 (14.1)
Selected	--.--	99.5 (8.1)	96.9 (14.2)	99.7 (15.2)
sgn[†]		ns	ns	**

[1] E = WC 5103 / CICA-8(E); F = WC 5103 / Oryzica-1(F); G = WC 5121 / CICA-8(G); H = WC 5121 / Oryzica-1(H).

[2] selection and cross x selection interaction were significant at the 0.01 probability level.

[†] significance of difference between means within the same column.

given in the Appendix Table 20. Differences were detected among crosses and between generations for this trait (Appendix Table 7). There was a significant cross x generation interaction for this trait (Appendix Table 7). F1-derived populations had a higher dispersion score than F2-derived populations, in crosses G and H, while no significant differences were detected in crosses E and F (Table 24). There were no significant differences due to the effect of breeding method or selection for this trait.

Contrasts for populations across crosses.

Observed mean squares of five contrasts between different combinations of populations for the seven traits evaluated in Bagua are found in Appendix Table 22. Differences were noted for spikelet sterility, panicles per square meter, 1000-gr. weight and plant height when F1DH populations were contrasted to F2DH-R populations. When comparing F2DH-R and F2DH-S populations, differences were detected for number of grains per panicle, number of panicles per square meter and plant height. Contrasting F2DH-R with SSD-R populations showed highly significant differences for all traits, except panicles per square meter, were noted. When SSD-R populations were compared to SSD-S populations, differences were noted only for leaf pubescence. Contrasts between F1DH and SSD-R populations, were significant for all

Table 23. Means and standard deviations for plant height (cm.) of 16 rice progenies of four upland/irrigated rice crosses developed either through doubled haploid or single seed descent from random and selected F2 plants, grown in Bagua, 1990.

	Method[1]	
	DH	SSD
Random	97.3 (14.8)	100.9 (12.3)
Selection	93.4 (16.0)	100.5 (13.5)
sgn[†]	**	ns

[1] difference between methods and the method x selection interaction were significant at the 0.01 and 0.05 probability level, respectively.

[†] significance of difference between means within the same column.

Table 24. Mean values and standard deviations for endosperm dispersion of 16 rice progenies of four upland/irrigated crosses derived from F1 and F2 generations, grown at Palmira, 1990.

	Crosses[1,2]			
Progenies	E	F	G	H
F1-derived	3.8 (1.7)	4.7 (2.0)	3.2 (1.6)	4.0 (1.8)
F2-derived	3.4 (1.6)	4.5 (1.8)	3.8 (1.6)	4.7 (1.8)
sgn[†]	ns	ns	**	**

[1] E = WC 5103 / CICA-8(E); F = WC 5103 / Oryzica-1(F); G = WC 5121 / CICA-8(G); H = WC 5121 / Oryzica-1(H).

[2] differences among crosses, between generations and the cross x generation interaction were significant at the 0.01, 0.01, and 0.05 probability level, respectively.

[†] significance of difference between means within the same column.

traits, with days to maturity and panicles per square meter being the exceptions.

Observed mean squares of the same contrasts for the seven traits evaluated in Palmira, are presented in Appendix Table 23. Differences between F1DH and F2DH-R populations were noted for spikelet sterility and 1000-grain weight. Differences were observed for leaf pubescence, spikelet sterility, and 1000-grain weight, when F2DH-R populations were compared to SSD-R populations. Differences for leaf pubescence and number of panicles per square meter were detected in the contrast between F1DH and SSD-R populations. When F2DH-R populations were contrasted to F2DH-S populations, differences were detected for leaf pubescence and spikelet sterility. Leaf pubescence, and spikelet sterility, in addition to number of grains per panicle, also differed between SSD-R and SSD-S populations.

Spikelet sterility. Means and mean deviations for spikelet sterility of the contrasts under study are presented in Table 25. Significant differences between mean deviations, which estimate the variability within populations, are also provided using Levene's test of homogeneity of variances. F2DH-R populations had higher percentage of spikelet sterility than F1DH and SSD-R populations, at both locations. Similarly, F2DH-R populations had higher spikelet sterility than F2DH-S populations, although this difference was significant only at Palmira. SSD-R populations had higher

mean values than SSD-S populations at both locations, but it was again only significant at Palmira. Higher percentage of spikelet sterility was observed in F1DH populations as compared to SSD-R populations, at both locations, with significance only detected at the Bagua site.

Since significant differences between F1DH and F2DH-R were detected at both locations, these populations were compared to their mid-parental values to explore the presence of epistasis for this trait. As can be observed in Tables 26 and 27, both DH populations showed higher percentage of spikelet sterility than the mid-parental values of their respective crosses, at both locations.

Proportion of pubescent plants. Mean and mean deviations for proportion of pubescent plants in the contrasts of interest are provided in Table 28. No differences were detected between F1DH and F2DH-R populations at either location, however higher variability was detected within F1DH populations, at Palmira. SSD-R populations had higher mean values than F2DH-R and F1DH populations, at both locations. F2DH-S had higher proportion of pubescent plants than F2DH-R at both locations; but, it was only significant at Palmira. Selection was found to be significant in SSD populations at both sites.

Table 25. Number (N), means ($\bar{x}$) and mean deviations (d) for percentage of spikelet sterility for five contrasts between progenies derived from four upland/ irrigated rice crosses grown at Bagua and Palmira, 1990.

	Bagua		sgn†	Palmira		sgn
	F1DH	**F2DH-R**		**FIDH**	**F2DH-R**	
N	360	266		341	258	
$\bar{x}$	29.7	32.3	*	42.8	46.8	*
d††	8.4	9.6		13.3	12.4	
	F2DH-R	**F2DH-S**		**F2DH-R**	**F2DH-S**	
N	134	186		130	181	
$\bar{x}$	33.5	30.9		48.9	42.4	**
d	9.8	8.7		15.2	15.2	
	F2DH-R	**SSD-R**		**F2DH-R**	**SSD-R**	
N	266	1585		167	1163	
$\bar{x}$	32.3	27.1	***	46.7	41.7	*
d	9.6	8.9		12.9	15.5	***
	SSD-R	**SSD-S**		**SSD-R**	**SSD-S**	
N	1188	447		1163	435	
$\bar{x}$	27.2	25.8		41.7	39.1	**
d	8.83	8.54	**	15.5	13.4	***
	F1DH	**SSD-R**		**F1DH**	**SSD-R**	
N	360	1585		282	1163	
$\bar{x}$	29.7	27.1	*	43.6	41.7	
d	8.4	8.9		14.42	15.5	

† significance of difference between contrasted means ($\bar{x}$), and contrasted mean deviations (d), as resulted from analyses of variance and Levene's test of homogeneity of variances, respectively.

†† average of the absolute deviations = $\Sigma |x_{ij} - \bar{x}_{i.}| / n$

* significant at the 0.05 probability level.
** significant at the 0.01 probability level.
*** significant at the 0.001 probability level.

Table 26. Means ($\bar{x}$), standard deviations (s.d.), and range for spikelet sterility of parents and doubled haploid (DH) populations of four upland/irrigated rice crosses at Bagua (Perú), 1990.

Cross	N	$\bar{x}$†	s.d.	range
WC 5103 x CICA-8 (E)				
Parents	54	16.6	5.8	5.1 - 23.9
F1DH	60	27.6 **	13.7	4.1 - 85.6
F2DH-R	93	31.1 **	16.6	2.5 - 75.6
WC 5103 x Oryzica-1 (F)				
Parents	54	20.8	6.5	9.3 - 44.1
F1DH	102	30.2 **	11.6	4.1 - 54.3
F2DH-R	39	30.6 **	13.4	7.4 - 68.9
WC 5121 x CICA-8 (G)				
Parents	54	14.6	7.9	2.9 - 32.0
F1DH	81	27.5 **	14.4	1.6 - 69.3
F2DH-R	66	28.6 **	13.3	4.8 - 65.5
WC 5121 x Oryzica-1 (H)				
Parents	54	16.1	7.8	2.9 - 37.3
F1DH	117	31.7 **	16.3	2.8 - 84.6
F2DH-R	68	38.4 **	18.4	2.3 - 94.1

† mid-parental value
** significant at the 0.01 probability level.

Table 27. Means ($\bar{x}$), standard deviations (s.d.), and range for spikelet sterility of parents and doubled haploid (DH) populations of four upland/irrigated rice crosses grown at Palmira (Colombia), 1990.

Cross	N	$\bar{x}$†	s.d.	range
WC 5103 x CICA-8 (E)				
Parents	52	26.3	12.4	7.7 - 53.2
F1DH	60	39.4 **	16.1	7.8 - 89.9
F2DH-R	92	46.9 **	20.6	11.2 - 92.0
WC 5103 x Oryzica-1 (F)				
Parents	43	28.5	12.1	6.5 - 53.2
F1DH	99	44.4 **	19.4	5.4 - 85.7
F2DH-R	38	38.3 **	18.7	8.4 - 85.9
WC 5121 x CICA-8 (G)				
Parents	50	19.7	7.2	7.6 - 41.8
F1DH	78	38.8 **	23.6	4.6 - 95.8
F2DH-R	62	45.9 **	23.6	5.5 - 98.2
WC 5121 x Oryzica-1 (H)				
Parents	51	20.4	8.2	8.9 - 41.8
F1DH	108	45.5 **	21.0	7.9 - 93.5
F2DH-R	69	51.7 **	25.7	5.8 - 99.9

† mid-parental value
** significant at the 0.01 probability level.

Days to maturity. Table 29 shows the means and mean deviations of the contrasted combinations for days to maturity. F1DH populations had higher mean values and variability than F2DH-R populations at Bagua. The same tendency was observed at Palmira, but no significant difference was detected at this site. No differences were detected between F2DH-R and F2DH-S populations at either location. SSD-R populations matured later than F2DH-R populations at Bagua. SSD-R populations presented higher intra-population variability than either F2DH-R or SSD-S populations, at both locations. However, the mean value of SSD-R populations was no different from that of SSD-S populations. No differences were detected between F1DH and SSD-R populations at either location.

Number of grains per panicle. Mean and mean deviations for grains per panicle are provided in Table 30. No differences were found between F1DH and F2DH-R populations in either location. F2DH-R populations showed higher mean and mean deviation values than F2DH-S populations in Bagua. No differences were detected at Palmira. SSD-R populations showed higher mean values when contrasted to either F1DH or F2DH-R populations, at Bagua only. SSD-S populations showed higher number of grains per panicle than SSD-R populations at Palmira.

Table 28. Number (N), means ($\bar{x}$) and mean deviations (d) for proportion of pubescent plants for five contrasts between progenies derived from four upland/ irrigated rice crosses, grown at two locations, Bagua and Palmira, 1990.

	Bagua		sgn†	Palmira		sgn
	F1DH	**F2DH-R**		**FIDH**	**F2DH-R**	
N	361	266		346	255	
$\bar{x}$	0.53	0.53		0.53	0.51	
d††	0.36	0.34		0.38	0.32	***
	F2DH-R	**F2DH-S**		**F2DH-R**	**F2DH-S**	
N	135	187		130	177	
$\bar{x}$	0.45	0.54		0.43	0.56	**
d	0.33	0.37		0.31	0.34	
	F2DH-R	**SSD-R**		**F2DH-R**	**SSD-R**	
N	266	1594		167	1164	
$\bar{x}$	0.53	0.66	***	0.49	0.65	**
d	0.34	0.34		0.29	0.37	***
	SSD-R	**SSD-S**		**SSD-R**	**SSD-S**	
N	1196	446		1164	434	
$\bar{x}$	0.66	0.77	**	0.65	0.78	**
d	0.34	0.21	***	0.37	0.23	***
	F1DH	**SSD-R**		**F1DH**	**SSD-R**	
N	361	1594		287	1164	
$\bar{x}$	0.53	0.66	***	0.53	0.65	***
d	0.36	0.34		0.40	0.37	*

† significance of difference between contrasted means ($\bar{x}$), and contrasted mean deviations (d), as resulted from analyses of variance and Levene's test of homogeneity of variances, respectively.

†† average of the absolute deviations = $\Sigma|x_{ij} - \bar{x}_{i.}|/n$

* significant at the 0.05 probability level.
** significant at the 0.01 probability level.
*** significant at the 0.001 probability level.

Table 29. Number (N), means ($\bar{x}$) and mean deviations (d) for days to maturity for five contrasts between progenies derived from four upland/irrigated rice crosses, grown at two locations, Bagua and Palmira, 1990.

	Bagua		sgn†	Palmira		sgn
	F1DH	**F2DH-R**		**FIDH**	**F2DH-R**	
N	361	268		346	254	
$\bar{x}$	147.5	145.9	*	146.5	145.7	
d††	4.9	3.9	***	4.4	3.9	
	F2DH-R	**F2DH-S**		**F2DH-R**	**F2DH-S**	
N	135	187		129	181	
$\bar{x}$	146.5	146.1		146.0	146.4	
d	4.3	4.3		4.1	5.1	
	F2DH-R	**SSD-R**		**F2DH-R**	**SSD-R**	
N	268	1592		165	1179	
$\bar{x}$	145.9	147.6	***	146.7	147.0	
d	4.0	4.9	***	3.4	4.9	***
	SSD-R	**SSD-S**		**SSD-R**	**SSD-S**	
N	1195	447		1179	434	
$\bar{x}$	147.3	146.4		147.0	147.4	
d	4.9	4.0	***	4.9	4.4	***
	F1DH	**SSD-R**		**F1DH**	**SSD-R**	
N	361	1592		287	1179	
$\bar{x}$	147.5	147.6		146.3	147.0	
d	4.9	4.9		4.6	4.9	

† significance of difference between contrasted means ($\bar{x}$), and contrasted mean deviations (d), as resulted from analyses of variance and Levene's test of homogeneity of variances, respectively.

†† average of the absolute deviations = $\Sigma |x_{ij} - \bar{x}_{i.}| / n$

* significant at the 0.05 probability level.
** significant at the 0.01 probability level.
*** significant at the 0.001 probability level.

Thousand grain weight. Means and mean deviations for 1000-grain weight are presented in Table 31. F1DH populations showed higher mean values than F2DH-R populations at both locations. In addition, higher variability was found within F1DH populations, although it was only significant at Palmira. Higher mean and mean deviations values were found in SSD-R populations than in F2DH-R populations at both locations. SSD-R populations presented higher 1000-gr. weight than F1DH populations only at Bagua. Higher 1000-gr. weight was observed in F2DH-S populations than in F2DH-R populations, at Bagua. Higher variability was found within F2DH-S as compared to F2DH-R populations, at both locations. No differences between mean values were noticed when SSD-R and SSD-S populations were contrasted. However, higher variability was found within SSD-R populations at both locations.

Since DH populations differed at both locations, they were compared to their respective MP values (Tables 32 and 33). F1DH from cross E showed lower mean values than its MP value at Bagua, while the F2DH from the same cross showed lower 1000-gr. weight than the mean performance of their parents at both locations. In cross F, both populations showed lower mean values than the MP for this trait at both locations. In DH populations derived from cross G, only F2DH-R at Bagua showed lower 1000-gr. weight than its MP value. F1DH from cross H presented higher 1000-gr. weight than its respective MP value, at Palmira. F2DH-R population

Table 30. Number (N), means ($\bar{x}$) and mean deviations (d) for number of grains per panicle for five contrasts between progenies derived from four upland/ irrigated rice crosses, grown at Bagua, and Palmira, 1990.

	Bagua		sgn†	Palmira		sgn
	F1DH	**F2DH-R**		**FIDH**	**F2DH-R**	
N	360	266		341	258	
$\bar{x}$	92.7	93.4		73.7	67.9	
d††	19.7	20.2		21.7	19.4	*
	F2DH-R	**F2DH-S**		**F2DH-R**	**F2DH-S**	
N	134	186		130	181	
$\bar{x}$	93.6	85.4	*	68.5	67.6	
d	23.8	19.2	**	25.3	21.9	
	F2DH-R	**SSD-R**		**F2DH-R**	**SSD-R**	
N	266	1587		167	1163	
$\bar{x}$	93.4	104.8	***	71.1	71.5	
d	20.2	19.8		20.9	23.9	***
	SSD-R	**SSD-S**		**SSD-R**	**SSD-S**	
N	1190	447		1163	435	
$\bar{x}$	102.9	102.0		71.5	75.5	**
d	19.8	19.6		23.9	21.0	***
	F1DH	**SSD-R**		**F1DH**	**SSD-R**	
N	360	1587		282	1163	
$\bar{x}$	92.7	104.8	***	72.6	71.5	
d	19.6	19.8		22.9	23.9	

† significance of difference between contrasted means ($\bar{x}$), and contrasted mean deviations (d), as resulted from analyses of variance and Levene's test of homogeneity of variances, respectively.

†† average of the absolute deviations = $\Sigma |x_{ij} - \bar{x}_{i.}| / n$

* significant at the 0.05 probability level.
** significant at the 0.01 probability level.
*** significant at the 0.001 probability level.

Table 31. Number (N), means ($\bar{x}$) and mean deviations (d) for 1000-grain weight for five contrasts between progenies derived from four irrigated/upland rice crosses, grown at two locations, Bagua and Palmira, 1990.

	Bagua		sgn†	Palmira		sgn
	F1DH	**F2DH-R**		**FIDH**	**F2DH-R**	
N	361	266		342	258	
$\bar{x}$	24.9	23.4	***	24.9	23.5	***
d††	1.9	1.6		2.1	1.5	***
	F2DH-R	**F2DH-S**		**F2DH-R**	**F2DH-S**	
N	134	187		130	181	
$\bar{x}$	23.5	24.3	*	24.1	24.6	
d	1.8	2.1	*	1.6	2.1	**
	F2DH-R	**SSD-R**		**F2DH-R**	**SSD-R**	
N	266	1589		167	1163	
$\bar{x}$	23.4	25.4	***	24.1	25.3	***
d	1.6	1.9	**	1.4	2.0	***
	SSD-R	**SSD-S**		**SSD-R**	**SSD-S**	
N	1192	445		1163	435	
$\bar{x}$	25.6	25.1		25.3	25.1	
d	2.0	1.8	***	2.0	1.8	***
	F1DH	**SSD-R**		**F1DH**	**SSD-R**	
N	361	1589		283	1163	
$\bar{x}$	25.0	25.4	***	25.2	25.3	
d	1.9	1.9		2.3	2.0	**

† significance of difference between contrasted means ($\bar{x}$), and contrasted mean deviations (d), as resulted from analyses of variance and Levene's test of homogeneity of variances, respectively.

†† average of the absolute deviations = $\Sigma |x_{ij} - \bar{x}_{i.}| / n$

* significant at the 0.05 probability level.
** significant at the 0.01 probability level.
*** significant at the 0.001 probability level.

Table 32. Means ($\bar{x}$), standard deviations (s.d.), and range for 1000-gr. weight of the parentas and doubled haploid (DH) populations of four upland/irrigated crosses at Bagua (Perú), 1990.

Cross	N	$\bar{x}$†	s.d.	range
WC 5103 x CICA-8 (E)				
Parents	54	25.2	2.5	21.5 - 28.9
F1DH	60	23.9 **	2.2	19.6 - 28.8
F2DH-R	93	22.9 **	2.4	16.4 - 28.5
WC 5103 x Oryzica-1 (F)				
Parents	54	26.9	0.8	25.3 - 28.9
F1DH	103	24.8 **	2.5	17.6 - 35.6
F2DH-R	39	24.1 **	3.6	15.8 - 31.0
WC 5121 x CICA-8 (G)				
Parents	54	24.6	2.3	20.0 - 29.9
F1DH	81	24.3	3.2	17.7 - 33.3
F2DH-R	66	23.0 **	2.8	17.2 - 35.0
WC 5121 x Oryzica-1 (H)				
Parents	54	25.9	1.5	20.0 - 29.9
F1DH	117	26.1	3.3	15.9 - 34.5
F2DH-R	68	24.0 **	3.6	13.1 - 33.2

† mid-parental value
** significant at the 0.01 probability level.

Table 33. Means ($\bar{x}$), standard deviations (s.d.), and range for 1000-gr. weight of the parents and doubled haploid (DH) populations of four upland/irrigated crosses at Palmira (Colombia), 1990.

Cross	N	$\bar{x}$†	s.d.	range
WC 5103 x CICA-8 (E)				
Parents	52	23.9	1.6	21.1 - 27.3
F1DH	60	23.3	2.5	18.1 - 30.4
F2DH-R	92	22.5 **	2.9	14.5 - 35.5
WC 5103 x Oryzica-1 (F)				
Parents	43	25.1	1.4	22.2 - 27.4
F1DH	99	23.9 **	2.9	17.6 - 35.3
F2DH-R	38	23.8 *	2.8	18.6 - 29.7
WC 5121 x CICA-8 (G)				
Parents	50	24.9	2.1	21.7 - 28.8
F1DH	78	24.5	3.2	15.0 - 31.0
F2DH-R	62	24.2	2.6	19.2 - 32.5
WC 5103 x Oryzica-1 (H)				
Parents	51	25.4	2.0	22.6 - 28.8
F1DH	109	26.9 **	3.9	19.9 - 35.8
F2DH-R	69	24.2 **	2.6	17.2 - 30.5

† mid-parental value
* significant at the 0.05 probability level.
** significant at the 0.01 probability level.

from this cross showed lower mean values than the MP for this trait, at both locations.

Number of panicles per square meter. Means and mean deviations for number of panicles per square meter are provided in Table 34. Higher mean values in F1DH populations over F2DH-R populations were detected only at Bagua. At this site, higher mean and mean deviations values were noted in F2DH-S populations as compared to F2DH-R populations. SSD-R populations showed a higher variability than either F2DH-R or SSD-S populations at the Bagua site. At both locations, no differences between mean values for this trait were observed when SSD-R populations were compared to either F2DH-R, or SSD-S populations. F1DH populations had higher number of panicles per square meter than SSD-R populations, at Palmira.

Plant height. Mean and mean deviations values for plant height are presented in Table 35. F2DH-R populations showed higher mean values when contrasted to either F1DH or F2DH-S populations, but had lower mean values than SSD-R populations. F2DH-R populations presented lower variability than SSD-R populations. No difference in mean values was detected when SSD-R populations were compared to SSD-S populations. However, higher variability was detected within the former population. SSD-R populations were taller on average than F1DH populations.

Table 34. Number, (N), means ($\bar{x}$) and mean deviations (d) for number of panicles per square meter for five contrasts between progenies derived from four upland/irrigated rice crosses, grown at Bagua and Palmira, 1990.

	Bagua		sgn†	Palmira		sgn
	F1DH	**F2DH-R**		**FIDH**	**F2DH-R**	
N	361	268		346	257	
$\bar{x}$	163.4	161.1	*	200.3	204.5	
d††	23.2	22.8		27.6	33.6	*
	F2DH-R	**F2DH-S**		**F2DH-R**	**F2DH-S**	
N	135	188		131	178	
$\bar{x}$	155.0	164.5	*	204.8	202.7	
d	22.7	29.4	**	38.0	37.8	
	F2DH-R	**SSD-R**		**F2DH-R**	**SSD-R**	
N	268	1592		167	1173	
$\bar{x}$	161.1	161.3		202.3	194.0	
d	22.8	25.3	**	34.4	31.6	
	SSD-R	**SSD-S**		**SSD-R**	**SSD-S**	
N	1194	447		1173	434	
$\bar{x}$	159.8	159.9		194.0	196.0	
d	25.4	23.7	**	31.6	31.7	*
	F1DH	**SSD-R**		**F1DH**	**SSD-R**	
N	361	1592		287	1173	
$\bar{x}$	163.4	161.3		203.2	194.0	**
d	23.2	25.3		28.7	31.6	

† significance of difference between contrasted means ($\bar{x}$), and contrasted mean deviations (d), as resulted from analyses of variance and Levene's test of homogeneity of variances, respectively.

†† average of the absolute deviations = $\Sigma |x_{ij} - \bar{x}_{i.}| / n$

* significant at the 0.05 probability level.
** significant at the 0.01 probability level.
*** significant at the 0.001 probability level.

Endosperm dispersion. Mean and mean deviation values for endosperm dispersion are presented in Table 35. No differences between mean values were detected when F2DH-R populations were contrasted to either F1DH or F2DH-S or SSD-R populations. However, differences between mean deviations were detected in these comparisons, with all three populations, F1DH, F2DH-S, and SSD-R, having higher within-population variability than F2DH-R populations. No differences for this trait were detected when SSD-R populations were compared to SSD-S populations. However, SSD-R populations showed higher endosperm dispersion variability than SSD-S populations. SSD-R populations had higher dispersion scores and higher variability than F1DH populations.

Proportion of recombinant lines.

Proportion of recombinant lines with 0.75 or higher proportion of pubescent plants, lesser number of days to maturity than the mid-parent (MP) value, and exceeding the MP values for all other traits are presented, for DH and SSD-R populations, in Tables 36 to 43. Only those traits in which a significant difference was detected between the parents were considered in the respective cross and location.

Chi-square tests indicated that there was no difference between the proportion of favorable recombinant DH lines produced from the F1 and F2 generation of cross E at Bagua

Table 35. Number (N), means ($\bar{x}$) and mean deviations (d) for plant height and endosperm dispersion for five contrasts between progenies derived from four irrigated/upland rice crosses, grown at Bagua and Palmira, respectively, 1990.

	BAGUA Plant height (cm.)		sgn†	PALMIRA Endosperm dispersion††		sgn
	F1DH	**F2DH-R**		**FIDH**	**F2DH-R**	
N	361	268		346	250	
$\bar{x}$	93.8	97.3	***	3.97	4.03	
d†††	8.7	8.1		1.19	0.88	***
	F2DH-R	**F2DH-S**		**F2DH-R**	**F2DH-S**	
N	135	187		126	175	
$\bar{x}$	99.6	93.4	***	4.18	4.41	
d	10.3	11.2		1.04	1.36	**
	F2DH-R	**SSD-R**		**F2DH-R**	**SSD-R**	
N	268	1593		164	1165	
$\bar{x}$	97.3	100.9	**	4.99	4.33	
d	8.1	9.0	**	0.89	1.35	***
	SSD-R	**SSD-S**		**SSD-R**	**SSD-S**	
N	1195	447		1165	432	
$\bar{x}$	101.5	100.5		4.33	4.34	
d	9.3	8.9	***	1.35	1.17	***
	F1DH	**SSD-R**		**F1DH**	**SSD-R**	
N	361	1593		288	1165	
$\bar{x}$	93.8	100.9	***	4.01	4.33	**
d	8.7	9.0		1.26	1.35	*

† significance of difference between contrasted means ($\bar{x}$), and between contrasted mean deviations (d), as resulted from contrast's analyses of variance and Levene's test of homogeneity of variances, respectively.

†† spreading of milled rice treated with a 1.7% solution of KOH; 1-7 scale, where 1= non affected grain, 7= completely dispersed and intermingled kernels.

††† average of the absolute deviations = $\Sigma |x_{ij} - \bar{x}_{i.}| / n$

(Table 36). Nevertheless, the F1DH failed to produce favorable recombinants in 4 out of 7 combination of traits. The SSD-R presented higher proportion of desirable recombinants than F1DH and F2DH-R. However, there were only few significant differences. No differences in proportion of favorable lines were detected either between F1DH and F2DH-R when this cross was grown in Palmira (Table 37).

No differences were found for the proportion of desirable lines for all comparisons made between any pair of populations of cross F at Bagua (Table 38). When these populations were compared in Palmira, a significant difference was detected between F1DH and F2DH-R when the proportion of lines fulfilling the given standards for pubescence and endosperm dispersion was taken into consideration (Table 39). The apparent advantage of F2DH-R over F1DH in this instance was ephemeral and failed to produce any desirable recombinant for two other combination of traits. Consistent with the findings in Bagua, no significant differences were detected between DH populations and SSD-R in any combination of traits at Palmira (Tables 38 and 39). However, SSD-R populations of this cross were able to produce favorable recombinants in all combinations of traits, while F1DH and F2DH-R failed to present any desirable lines for one and two combinations of traits, respectively.

The proportion of favorable recombinant lines of F1DH and F2DH-R populations of cross G showed no difference at Bagua (Table 40) and Palmira (Table 41). When these

Table 36. Proportion of lines performing above the mid-parent value for different combination of traits from doubled haploid (DH) and single seed descent (SSD) derived progenies from cross WC 5103 x CICA-8 (E), at Bagua, 1990.

		trait combinations†						
	N	1	1+2	1+2+3	1+2+4	1+2+ 3+4	1+2+ 4+5	1+2+ 3+4+5
F1DH	60	0.55	0.03	0.02	0.0	0.0	0.0	0.0
F2DH-R	92	0.57	0.04	0.01	0.03	0.01	0.02	0.01
SSD-R	398	0.66	0.14	0.06	0.08	0.05	0.03	0.02
SSD-R(A)††	85	0.65	0.22	0.11	0.13	0.07	0.05	0.01
		χ_1^2						
F1DH vs F2DH-R		0.03	0.09	0.28	1.99	0.66	1.32	0.66
F1DH vs SSD-R		2.54	5.26*	2.07	5.36*	2.82	1.86	0.92
F2DH-R vs SSD-R		2.66	6.33*	4.01*	2.78	2.37	0.21	0.09

† 1 = pubescence (proportion of plants having ≥ 75% of pubescent plants); 2 = grains/panicle; 3 = % spikelet sterility; 4 = panicles/square meter; 5 = 1000-grain weight.
†† random sample taken from SSD-R.
* significant at the 0.05 probability level.

Table 37. Proportion of lines performing over the mid-parent value for different combinations of traits of doubled haploid (DH) progenies derived from cross WC 5103 x CICA-8 (E), at Palmira, 1990.

		trait combinations†						
	N	1	1+6	1+2	1+2+6	1+2+3	1+2+ +3+6	1+2+4
F1DH	58	0.53	0.28	0.07	0.07	0.05	0.05	00
F2DH-R	84	0.58	0.19	0.04	0.02	0.01	0.01	00
		χ^2_1						
F1DH vs F2DH-R		0.33	1.43	0.81	1.73	1.99	1.99	00

† 1 = pubescence (proportion of plants having ≥ 75% of pubescent plants);
2 = grains/panicle; 3 = % spikelet sterility;
4 = panicles/square meter; 6 = endosperm dispersion.

Table 38. Proportion of lines performing over the mid-parent value for different combinations of traits of doubled haploid (DH) and single seed descent (SSD) progenies derived from cross WC 5103 x Oryzica-1 (F), at Bagua, 1990.

		trait combinations†		
	N	1	1+2	1+2+4
F1DH	101	0.50	0.10	0.06
F2DH-R	39	0.67	0.18	0.05
SSD-R	396	0.60	0.15	0.08
SSD-R(A)††	85	0.62	0.13	0.09
		χ^2_1		
F1DH vs F2DH-R		2.97	1.71	0.03
F1DH vs SSD-R		3.39	1.68	0.42
F2DH-R vs SSD-R		0.55	0.26	0.37

† 1 = pubescence (proportion of plants having ≥ 75% of pubescent plants);
2 = grains/panicle; 4 = panicles/square meter.

†† random sample taken from SSD-R.

Table 39. Proportion of lines performing over the mid-parent value for different combinations of traits of doubled haploid (DH) and single seed descent (SSD) progenies derived from cross WC 5103 x Oryzica-1 (F), at Palmira, 1990.

		trait combinations†					
	N	1	1+6	1+2	1+2+6	1+2+4	1+2+4+6
F1DH	99	0.52	0.28	0.03	0.01	0.01	0.0
F2DH-R	37	0.68	0.49	0.11	0.03	0.0	0.0
SSD-R	376	0.56	0.32	0.05	0.03	0.01	0.01
SSD-R(A)††	85	0.52	0.30	0.05	0.04	0.03	0.01
		χ^2_1					
F1DH vs F2DH-R		2.82	4.99*	3.34	0.53	0.38	0.0
F1DH vs SSD-R		1.45	0.98	1.07	1.17	0.19	1.33
F2DH-R vs SSD-R		1.21	3.40	1.61	0.0	0.60	0.50

† 1 = pubescence (proportion of plants having ≥ 75% of pubescent plants);
2 = grains/panicle; 4 = panicles/square meter;
6 = endosperm dispersion.

†† random sample taken from SSD-R.

populations were compared to SSD-R populations, significant differences were only detected for the number of favorable lines for the trait pubescence, at both locations. SSD-R populations consistently produced desirable recombinants for all combinations of traits, while F1DH and F2DH-R were unable to produce favorable lines for some combinations of traits at both locations (Table 40 and 41).

Chi-square tests indicated no difference for the proportion of favorable lines between F1DH and F2DH-R (Tables 42 and 43). SSD-R showed a higher proportion of favorable lines than either, F1DH (for two combinations of traits) and F2DH-R (for three combinationss of traits), at Bagua (Table 42). SSD-R also resulted in a number of desirable lines than both F1DH and F2DH-R, for one combinations of traits, at Palmira (Table 43). No differences were found between all other combinations of traits.

Random samples of 85 lines were taken from each SSD-R population, in order to observe the performance of SSD populations when lesser number of individuals were involved. These samples produced desirable recombinants for all combinations of the traits considered in all crosses at both locations, except for a combination of five traits in cross G at Palmira (Tables 36 to 43).

Doubled haploid lines derived from cross H were used to estimate the effect of selection in the F2 prior to AC in increasing the frequency of desirable recombinants in AC-derived populations. Cross H had contrasting parents for

Table 40. Proportion of lines performing over the mid-parent value for different combinations of traits of doubled haploid (DH) and single seed descent (SSD) progenies derived from cross WC 5121 x CICA-8 (G), at Bagua, 1990.

		trait combinations†				
	N	1	1+7	1+2+7	1+2 4+7	1+2+ 4+5+7
F1DH	81	0.53	0.28	0.04	0.01	0.0
F2DH-R	66	0.50	0.33	0.08	0.03	0.0
SSD-R	398	0.66	0.38	0.11	0.06	0.03
SSD-R(A)††	85	0.65	0.35	0.08	0.06	0.02
		χ_1^2				
F1DH vs F2DH-R		0.14	0.42	1.06	0.59	0.0
F1DH vs SSD-R		4.53*	2.92	3.71	2.71	2.29
F2DH-R vs SSD-R		5.92*	0.63	0.55	0.72	1.87

† 1 = pubescence (proportion of plants having ≥ 75% of pubescent plants);
2 = grains/panicle; 4 = panicles/square meter;
5 = 1000-grain weight; 7 = days to maturity.
†† random sample taken from SSD-R.
* significant at the 0.05 probability level;

Table 41. Proportion of lines performing over the mid-parent value for different combinations of traits of doubled haploid (DH) and single seed descent (SSD) progenies derived from cross WC 5121 x CICA-8 (G) at Palmira, 1990.

		combination of traits†								
	N	1	1+7	1+6+7	1+2+7	1+2+ 4+7	1+2+4 +5+7	1+2 +6+7	1+2+4 +6+7	1+2+4 5+6+7
F1DH	77	0.55	0.23	0.05	0.04	0.04	0.03	0.0	0.0	0.0
F2DH-R	60	0.50	0.22	0.15	0.0	0.0	0.0	0.0	0.0	0.0
SSD-R	379	0.68	0.21	0.08	0.05	0.03	0.02	0.03	0.02	0.01
SSD-R(A)††	85	0.65	0.20	0.06	0.07	0.04	0.02	0.02	0.01	0.0
		χ^2_1								
F1DH vs F2DH-R		0.28	0.06	3.78	2.39	2.39	1.58	0.0	0.0	0.0
F1DH vs SSD-R		4.78*	0.15	0.58	0.17	0.04	2.62	2.08	1.65	0.61
F2DH-R vs SSD-R		11.95**	0.09	3.54	3.14	2.12	0.96	1.62	1.29	0.48

† 1 = pubescence (proportion of plants having ≥ 75% of pubescent plants);
2 = grains/panicle; 4 = panicles/square meter; 5 = 1000-grain weight;
6 = endosperm dispersion; 7 = days to maturity.
†† random sample taken from SSD-R.
* significant at the 0.05 probability level;
** significant at the 0.01 probability level.

Table 42. Proportion of lines performing over the mid-parent value for different combinations of traits of doubled haploid (DH) and single seed descent (SSD) progenies derived from cross WC 5121 x Oryzica-1 (H), at Bagua, 1990.

		trait combinations†			
	N	1	1+7	1+2+7	1+2 +4+7
F1DH	117	0.49	0.21	0.03	0.03
F2DH-R	69	0.41	0.16	0.01	0.0
SSD-R	399	0.64	0.39	0.08	0.05
SSD-R(A)††	85	0.64	0.45	0.12	0.05
		χ^2_1			
F1DH vs F2DH-R		1.16	0.82	0.64	2.41
F1DH vs SSD-R		9.35**	11.85**	3.20	0.67
F2DH-R vs SSD-R		14.03**	13.23**	4.06*	3.80

† 1 = pubescence (proportion of plants having ≥ 75% of pubescent plants);
2 = grains/panicle; 3 = % spikelet sterility;
4 = panicles/square meter; 5 = 1000-grain weight;
6 = endosperm dispersion; 7 = days to maturity.
†† random sample taken from SSD-R.
* significant at the 0.05 probability level;
** significant at the 0.01 probability level.

Table 43. Proportion of lines performing over the mid-parent value for different combinations of traits of doubled haploid (DH) and single seed descent (SSD) progenies derived from cross WC 5121 x Oryzica-1(H), at Palmira, 1990.

		combination of traits†						
	N	1	1+7	1+6+7	1+2+7	1+2+ 4+7	1+2 +6+7	1+2+4 +6+7
F1DH	108	0.51	0.17	0.05	0.02	0.02	0.02	0.02
F2DH-R	65	0.37	0.14	0.06	0.01	0.0	0.0	0.0
SSD-R	386	0.59	0.23	0.14	0.04	0.03	0.03	0.02
SSD-R(A)††	85	0.54	0.25	0.16	0.04	0.04	0.02	0.02
		χ^2_1						
F1DH vs F2DH-R		3.21	0.25	0.03	0.02	1.22	1.22	1.22
F1DH vs SSD-R		2.29	1.88	5.63*	1.05	0.19	0.33	0.05
F2DH-R vs SSD-R		11.06**	2.64	3.05	0.89	1.72	1.90	1.02

† 1 = pubescence (proportion of plants having ≥ 75% of pubescent plants);
2 = grains/panicle; 4 = panicles/square meter; 6 = endosperm dispersion;
7 = days to maturity.
†† random sample taken from SSD-R.
* significant at the 0.05 probability level;
** significant at the 0.01 probability level.

proportion of pubescent plants, days to maturity, and plant height which were the criteria for selection in the F2. Proportion of recombinant lines of DH populations with 0.75 or higher proportion of pubescent plants, lesser number of days to maturity than the MP value, plant height within the range of both parents $[(P_1 - 1.0\ \sigma) \leq x \leq (P_2 + 1.0\ \sigma)]$, and higher values than the MP value for grains/panicle, and panicles/m2, are presented in Table 44. Chi-square tests indicated that F2DH-S had higher frequency of desirable lines for all three traits that were selected for than F2DH-R. No differences were detected when F1DH and F2DH-S populations were compared.

Table 44. Proportion of desirable lines for different combinations of traits of doubled haploid (DH) progenies derived from cross WC 5121 x Oryzica-1 (H), at Bagua, 1990.

		trait combinations†				
	N	1	1+8	1+7+8	1+2+ 7+8	1+2 +4+7+8
F1DH	117	0.49	0.29	0.15	0.02	0.02
F2DH-R	69	0.41	0.22	0.03	0.0	0.0
F2DH-S	104	0.53	0.28	0.17	0.02	0.0
		χ^2_1				
F1DH vs F2DH-S		0.38	0.04	0.15	0.01	1.79
F2DH-R vs F2DH-S		2.52	0.83	8.42**	1.34	0.0

† 1 = pubescence (proportion of plants having ≥ 75% of pubescent plants);
2 = grains/panicle; 4 = panicles/square meter;
7 = days to maturity; 8 = plant height.

** significant at the 0.01 probability level.

DISCUSSION

Most Latin American irrigated rice cultivars share a narrow genetic base which limits their ability to deal with the potential appearance of new pathogens or new physiological races of existing pathogens. This further restricts their use in favored-upland areas being sown to irrigated cultivars.

Upland-dwarf cultivars, developed from African and Brazilian germplasm at the Centro Internacional de Agricultura Tropical (CIAT, Colombia), posses an array of desirable traits, i.e. earliness, deep rooting patterns, resistance to rice blast, brown spot, etc. If such germplasm could be incorporated into irrigated cultivars the total genetic variability could be enhanced. Breeders at CIAT have started to hybridize these two gene pools, but problems related to wide hybridization, such a F1 partial sterility and which extends segregating generations, are often found in the resulting progeny of these crosses.

Anther culture, which reportedly alleviates the F1 partial sterility problem in *japonica-indica* crosses. This procedure also decreases the time needed to produce homozygous lines. It may provide a bridge to incorporate desired traits from upland into an irrigated rice germplasm. However, several considerations must be taken into account

before anther culture becomes an established breeding method in using wide hybridization for rice improvement.

Two-locations were used to compare AC and SSD breeding methods for spikelet sterility and proportion of recombinant lines for a number of traits in four upland/irrigated crosses. Doubled haploid lines obtained from both F1 and F2 generations were also compared for a number of traits to determine the most appropriate generation for extracting anthers for in vitro AC. The response to selection in the F2 prior to the production of DH lines was also explored by comparing DH lines obtained from random (non selected) versus selected F2 plants for earliness, presence of pubescence and semidwarf plant type.

Doubled Haploids generated from F1 and F2 generations.

Following hybridization in self-pollinated crops, different generations can be chosen as parental material for producing doubled haploids, although the most common strategy is to use the F1 generation. Thus the products resulting from segregation and recombination between the two parents can be stabilized at the earliest possible occasion. The resulting haploid lines generated are products of meiosis in the F1 and represent the result of only one opportunity for recombination between the parental genomes. If linkage of useful genes is an important component of the genetic variation, doubled haploid lines generated from the F1 may

not be able to achieve some desirable recombinants. In such circumstances, production of doubled haploids from the F2, instead of the F1, would allow for one additional cycle of meiosis and the additional opportunity of breaking undesirable coupling or repulsion linkages (Snape, 1989). Field comparisons between dihaploid lines generated from different filial generations have been made in barley using the *H. bulbosum* technique (Snape and Simpson, 1981; Bjørnstad, 1987). This investigation is the first to undertake such a comparison with dihaploid lines derived from anther culture, using several crosses and more than one location.

In this study, based on the overall means of the F1DH and F2DH-R populations grown at the two locations, differences were detected only for spikelet sterility and 1000-grain weight.

Differences in means and variances are indicative of the presence of linkage if epistasis between the linked genes is present (Choo, 1981). Even though no individual comparisons in terms of means and variances were made per cross, a tentative analysis can be made from the results of this study to determine the presence or absence of linkage in the crosses utilized. The significant difference between the overall means of DH populations is suggestive that linkage for spikelet sterility is involved in most crosses. In fact, F2DH-R populations from all crosses, except cross F, had a higher percentage of spikelet sterility than F1DH progenies.

The different response of cross F (WC 5103/Oryzica-1) seems to be related to the small number of lines that composed the F2DH-R population of this cross, which may not represent a random sample of lines.

According to Snape and Simpson (1981), a test for epistasis is provided by a comparison of the midparental value with that of a DH population. In this investigation, all DH populations showed a higher percentage of spikelet sterility than their respective mid parental value, suggesting the presence of epistasis in all crosses investigated. This observation seems to reinforce the linkage possibility. Nevertheless, no differences were detected between the overall variances of these two populations failing to confirm the presence of linkage for spikelet sterility. Ranges exhibited by DH progenies derived from both F1 and F2, showed evidence of transgressive segregation in all crosses and may also indicate that no close linkages were associated with this trait. In this study, recombination seems to have led to an increase of percentage of sterility in three of the four crosses investigated.

F1DH populations showed higher overall mean values for 1000-grain weight than F2DH-R populations. When DH progenies of each cross were compared to their respective mid-parental values, only the F1DH from cross F (WC 5103/Oryzica-1) showed consistent differences at both locations. However, F2DH-R progenies from all crosses, except cross G (WC 5121/CICA-8), showed consistent differences with their mid-parental value,

which may be indicative of the presence of epistasis for this trait. While the F1DH progenies had higher overall variances than F2DH-R progenies, which is characteristic of a preponderance of coupling linkages with no epistatic effects (Snape and Simpson, 1981). This seems to have been the case with crosses E (WC 5103/CICA-8) and G (WC 5121/CICA-8), where both had contrasting parents for 1000-grain weight.

The frequency of inbred lines extractable from a cross which transgress a given standard is of primary concern for the breeder. In comparing the proportion of favorable recombinants produced from both DH populations, a common pattern did not appear for all crosses. In some crosses, either one, F1DH or F2DH-R was unable to produce any favorable recombinant at all for some combination of traits. However this appears to be related to relative population sizes rather than to number of meiosis cycles. The population that failed to produce any recombinants was invariably the one of smaller size. These observations highlight the importance that adequate population sizes has for the success of anther culture as a breeding method.

It does not seem that linkage played a mayor role in the genetic variability for the traits investigated in these crosses. However, recombination seems to have resulted in a decrease in mean and variance for 1000-grain weight of most crosses. This probably is the result of the breakage of desirable coupling linkages. Derivation of DH lines from the F1 appears to be a more practical approach if there is a

predominance of desirable coupling linkages. The F2 would be the generation of choice if undesirable coupling or repulsion linkages are predominant in the parental generation. Rice breeding programs interested in bringing in a few new upland traits into an irrigated rice background should probably select the F1 generation for deriving dihaploids if contrasting parents are used for the traits of interest. However, other considerations should be also taken, such as the chance of selecting for highly heritable traits in the F2 prior to the production of dihaploids.

Effect of selection in the F2 before the production of homozygous lines.

It has been postulated that one of the advantages of using F2 plants for producing haploids is the chance of selecting for major gene characters and for quantitatively inherited traits of high heritability (Choo *et al.*, 1985). Using simulation methods, Yonezawa *et al.* (1987), found that DH lines produced from the best 10 plants in the F2 were always superior to DH lines derived from the F1 in keeping desirable genotypes. Nevertheless, no field comparison has been reported so far to estimate the usefulness of selecting in the F2 prior to the production of DH plants.

In this research, selection in the F2 prior to anther culture was effective in increasing the proportion of pubescent plants and decreasing the plant height of DH

derived progenies. No consistent response to selection was observed for days to maturity. These results seem to confirm expectations that selection in the F2 could be effective in increasing the frequency of favorable genotypes in the DH population for simple inherited traits and quantitative traits of high heritability.

Selection in the F2 was also effective in increasing the mean value of SSD populations for proportion of pubescent plants, but not for plant height. In the comparisons between SSD-R and SSD-S based on three crosses, only one cross involved parents diverging significatively for plant height. This may account for the lack of overall response to selection in the SSD for this trait.

Selection practiced in the F2 for pubescent plants of shorter maturation period and semidwarf plant type did not significatively affect the populations performance for 1000-grain weight, number of panicles/m2, and number of grains/panicle, suggesting that no tight linkages existed between those traits and traits of economic importance.

Given these considerations it seems wise to practice selection in the F2 for traits of high heritability. This will increase the frequency of alleles for these traits, and hence the proportion of recombinant lines carrying all the desirable traits. Only desirable lines will then be used for doubled haploid production ensuring a higher frequency of lines with the desired levels of performance and, therefore,

smaller population sizes would be necessary for genetic advance.

Nevertheless, even though the frequency of desirable recombinants increased in the F2DH-S population of cross H as compared to unselected F2DH population, F2DH-S did not produce a higher proportion of desirable lines than the F1DH of the same cross. This might be due to the presence of favorable coupling linkages in the F1 which were then broken in the F2. Selection in the F2 was effective in recovering favorable recombinants but it could not surpass the performance of the F1DH progenies.

Doubled haploid versus SSD breeding.

Single seed descent and DH are both methods that generally reduce the time to develop homozygous lines. In theory, the mean and variances of DH lines are identical to those of pure-breeding lines derived from the SSD method in absence of linkage and linkage disequilibrium (Jinks and Pooni, 1981). Computer simulation studies confirmed these theoretical assumptions and have shown that SSD lines were superior to DH in obtaining a higher frequency of recombinants when linkage was either in coupling or in repulsion (Riggs and Snape, 1977). Field experiments have failed to confirm these findings. Doubled haploidy has been extensively compared to SSD in field performance trials in barley. Choo *et al.* (1981), and Park *et al.* (1976) found no

differences between these two methods in frequency distributions, means, variances, and frequency distributions. Powell and colleagues (Powell *et al.*, 1986, Caligari *et al.* 1987), found that DH was more efficient than SSD in preserving desirable coupling linkages. Similar comparisons have been made in triticale (Charmet and Branlard, 1985), and Brussels sprouts (Kubba, *et al.*, 1989), indicating that DH lines perform comparably to an equivalent sample of inbred lines produced by SSD. No such a comparison has been reported so far in rice.

In this investigation, SSD progenies had higher mean values than DH progenies involving pubescent plants. They also showed a tendency for having lower percentage of spikelet sterility, higher 1000-grain weight and were taller than DH progenies.

At least part of the differences between SSD's and DH's progenies can be explained in terms of residual heterozygosity and hence dominance effects in the SSD progenies. This could be true in this study as pubescence is dominant over non-pubescence, and tallness is dominant over short stature (Aquino and Jennings, 1966). Nevertheless, dominance alone can not explain the large differences detected for proportion of pubescent plants. The expected frequency of pubescent plants in the F4 for this trait, which is controlled by a single major gene, is 0.5625, while mean values observed in the SSD populations ranged from 0.61 to 0.69. Only frequencies observed in cross F (WC 5103/Oryzica-

1) fit the expected mendelian expectations for this trait. Theoretically, the difference between these two populations can be explained only by the presence of linkage disequilibrium between pairs of loci displaying non-allelic interactions. Nevertheless, differential survival or selection during the production of the DH or SSD lines may also explain these differences. Natural selection favoring individuals with traits conferring adaptation to irrigated conditions may have increased the frequency of pubescent lines in most crosses. Recombination did not appear to have contributed to this increased proportion of pubescent plants in the SSD. Single seed descent lines had only one additional cycle of meiosis as compared to F2DH, which did not show a significant increase for this trait compared to F1DH in most crosses. Moreover, recombination is theoretically higher in the F2 than in the F3 (Choo, 1981).

When SSD-R progenies were compared to DH progenies in terms of the proportion of desirable traits, SSD-R was able to provide desirable recombinants for all the combinations of traits considered in all crosses at both locations. However, the DH technique failed to provide desirable lines for some combination of traits, especially in populations with less than 70 DH lines. The higher number of lines generally associated with the SSD method could be a factor in its apparent advantage over DH. Nevertheless, SSD was still superior to DH populations even when a random sample of 85

lines was taken from each SSD population and analyzed for the proportion of desirable recombinants.

In a rice breeding program many more traits than those analyzed here must be taken into account. Screening for traits such as disease resistance (to blast, Hoja Blanca virus, etc.), tolerance to soil stresses (Fe and Al tolerance), and grain quality (amylose content), will decrease sharply the probability of obtaining individuals carrying all the desirable traits. The success of a breeding program using the DH method will rely heavily on producing large enough numbers of dihaploids by efficient and simple techniques from a wide array of genotypes.

In spite of the important improvements reached on the rice AC technique during the past years, most irrigated germplasm remains poorly responsive to AC. Chinese scientists have estimated that a population of 150 dihaploid lines is equivalent to a population of 4000 - 5000 F2 plants (Shen *et al.*, 1983). Scientists at the Centro Internacional de Agricultura Tropical considered that 125 dihaploid lines per cross is a reasonable target (Pulver and Jennings, 1985). Actual numbers will vary according to the genetic diversity between parents. Few breeding programs would probably dedicate the effort necessary to generate an adequate number of dihaploids lines from crosses involving poorly responsive genotypes like those included in this study.

In irrigated rice breeding programs, anther culture will probably play a more important role in germplasm enhancement

rather than the direct production of commercial cultivars. A few fixed lines carrying one or two upland traits within an irrigated background could be rapidly produced through AC and serve as donors of these traits for further improvement.

Gametic selection in rice anther culture.

Evidence of gametic selection in AC has been reported in several crops. Shen *et al.* (1981), reported in rice that most of the green plantlets developed by AC from an *indica-japonica* cross were *japonica* and intermediate type, indicating that there may be a mechanism favoring the development of *japonica* rice pollens. This observation was corroborated by Oono (1981), when DH progenies were compared to seed-derived F2 progeny which showed by contrast an equal distribution of *japonica* and *indica* types. In DH progenies from barley crosses involving the highly responsive to AC cultivar Blenheim, Thompson *et al.* (1991), found evidence suggesting that genetic response factors from Blenheim are linked to four genetic markers. Selection during androgenesis due to a change in medium composition has been also observed in barley anther culture (Kao and Horn, 1988).

Response to anther culture is under genetic control. This has been illustrated for rice (Miah *et al.*, 1985), and for a number of other crops, including barley (Knudsen *et al.*, 1989; Finnie *et al.*, 1989; Powell, 1988), wheat (Bullock *et al.*, 1982), and broccoli (Orton and Browers, 1985).

Japonica genotypes are generally more responsive to anther culture than *indica* genotypes (Chen Ying, 1986). At the Centro Internacional de Agricultura Tropical the most responsive genotypes are found within the upland germplasm, even though many upland lines are not responsive. Poor response, or no response, are characteristic of the irrigated germplasm. In this investigation four crosses were used each involving an AC responsive upland cultivar and a poorly responsive (sometimes no-responsive) irrigated cultivar. If genetic factors controlling AC responsiveness were linked to loci controlling traits studied in this investigation it should have been detected.

It was observed that earliness from the upland line WC 5121 (the most responsive genotype in the Latin American germplasm) combined rather easily to traits found in irrigated types such a leaf pubescence, high number of grains/panicle[1] and high number of panicles/m2 which were contributed from CICA-8 (a poorly responsive genotype). About half of the DH lines produced from this cross were pubescent, and about half of these were earlier than the midparent value. This suggests that factors controlling androgenesis in rice are not linked to traits of economic importance.

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[1]high number of grains/panicle as a consequence of large panicles are usually regarded as typical of upland cultivars. In this investigation, however, upland lines presented high spikelet sterility and hence lower number of grains/ panicle, probably as a result of being grown under irrigated conditions.

Application of AC in rice wide hybridizations.

Wide hybridization as a rice breeding strategy to enhance genetic variability, either between subspecies or between ecotypes, is difficulted because of the partial sterility of the F1 hybrids, undesirable segregation patterns for several generations, and differentiation of characters into two extremes. Chinese scientists have reported that AC can alleviate the partial sterility problem commonly found in the F1 of wide crosses in rice (Zhang, 1982; Shen *et al.*, 1983). Li *et al.*, 1983, reported that a rate of seed set of 80 % could be achieved in AC lines derived from *indica/japonica* crosses as compared to backcrossing that only could reach 40-50 % with the same combinations.

In this investigation that involved four crosses between upland and irrigated rice genotypes with F1 partial sterility, it was observed that SSD progenies tended to have less spikelet sterility than AC derived DH progenies. Since SSD lines were grown over more seasons than DH progenies, some selection may have taken place favoring the survival of the more fertile plants, and given a slight advantage to SSD progenies in most crosses. However, there were no consistent differences detected between the variances of these two populations. Aside from the higher percentage of sterility shown by the F2DH progenies when compared to F1DH-R, as previously discussed, AC does not seem to have affected the performance of DH lines in the crosses studied.

Based on these observations AC does not seem to have any additional advantage over conventional breeding, in upland x irrigated crosses, other than accelerated generation advance.

SUMMARY AND CONCLUSIONS

Two breeding methods, doubled haploid (DH), and single seed descent (SSD), were compared to determine which would provide the highest percentage of desired progeny from crosses between *japonica* and *indica* rice gene pools.

The experimental populations were derived from four upland (*japonica sensu lato*) x irrigated (*indica*) rice single crosses. Doubled haploid lines were produced via anther culture (AC). Single seed descent lines were advanced and evaluated in the F4 generation. Field performance of the populations was compared at two locations, Bagua (Perú), and Palmira (Colombia). To determine the optimum generation, DH populations were obtained from F1 and F2 and their field performance compared for an array of traits. The putative advantage of selecting in the F2 prior to the production of DH plants was further explored by comparing DH populations derived from both random and selected F2 plants for earliness, semidwarf plant type, and presence of leaf pubescence.

All comparisons between populations were made in terms of means, variances and proportion of favorable recombinant lines for a set of traits which included: spikelet sterility, days to maturity, proportion of pubescent plants, number of grains/panicle, number of panicles/m2, 1000-gr. weight, plant height, and endosperm dispersion.

The following conclusions were drawn based on the

results obtained in this investigation:

1. Single seed descent progenies had higher mean values than DH progenies for proportion of pubescent plants. They also had higher 1000-grain weight and were taller than DH progenies. Residual heterozygosity in SSD progenies may account for part of these differences.

2. Doubled haploid progenies tended to have higher spikelet sterility than SSD progenies. This may be due to the effect of natural selection favoring the survival of fertile plants in the SSD or to some carryover deleterious effect resulting from the anther culture technique.

3. Single seed descent provided desirable recombinants for a higher number of desirable traits than DH populations at both locations.

4. Doubled haploid progenies derived from the F2 had a higher percentage of spikelet sterility and lower 1000-grain weight than those derived from the F1.

5. Selection in the F2 prior to developing doubled haploids was effective in increasing the proportion of pubescent plants and decreasing plant height compared to DH populations derived from non-selected F2 plants. This suggests that selection for simple inherited traits and quantitative traits with high heritabilities can effectively be made in this generation.

6. Selection in the F2 prior to anther culturing did not increase the proportion lines carrying all the desirable attributes for pubescence, days to maturity, number of

panicles/m2, plant height and grains/panicle when compared to F1-derived DH progenies. This may be explained by the breakage of desirable coupling linkages in F2-derived progenies, and suggests that the F1 may be superior in deriving DH in rice irrigated programs trying to incorporate a few traits from upland rice.

7. Desirable traits contributed from irrigated cultivar CICA-8 (poorly responsive to AC technique) combined with earliness contributed from upland cultivar WC 5121 (highly responsive to AC) in doubled haploid lines, suggesting that factors controlling AC response are not linked to these traits of economic importance in rice.

8. Dihaploid populations with less than 70 lines per cross provided recombinants for lesser number of desirable traits than populations with greater number of individuals. The success of AC as a breeding method will rely heavily on obtaining an adequate number of individuals to ensure a wide array of genetic combinations.

9. Performance of the two breeding methods gave consistent results across locations. This indicates that results obtained in this research may have a broad application.

10. Anther culture allows the rapid fixation of traits into completely homozygous lines, and may play an important role in a breeding scheme oriented to enhance the genetic variability of irrigated rice. By using AC, a few (one or two) upland traits can be rapidly fixed within an irrigated background from the F1 of upland x irrigated crosses. Derived

DH lines may be then used as donors of traits found in upland rice for further improvement using conventional breeding methods.

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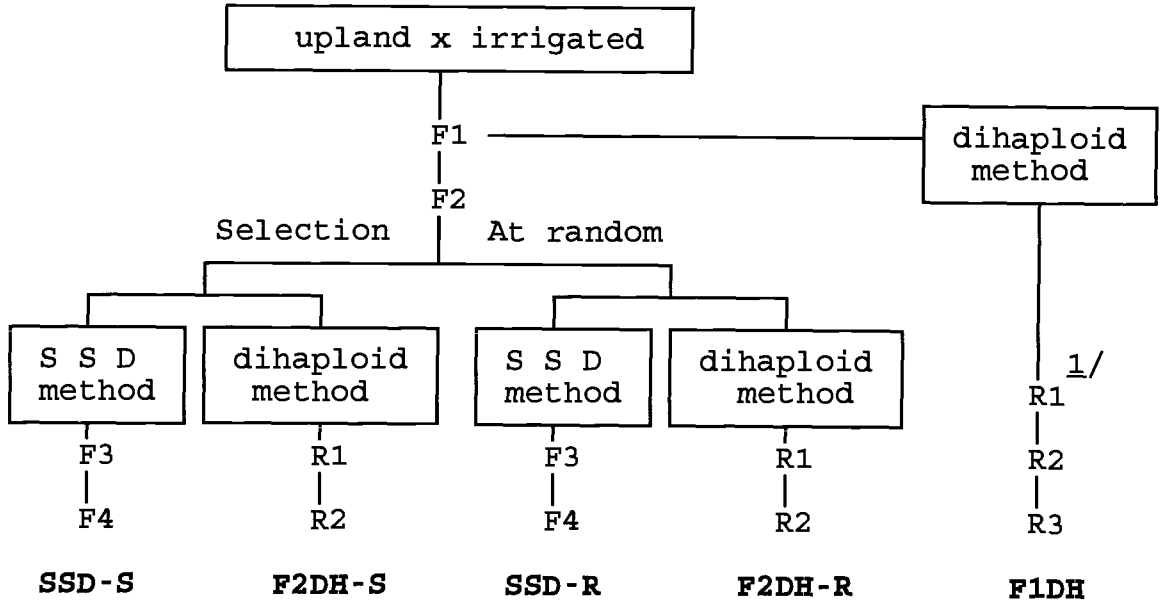
Appendix Table A1. Four single upland x irrigated rice crosses used in this study.

Cross	Pedigree[1]	CIAT cross N°
E	WC 5103 x CICA-8	CT 10366
F	WC 5103 x Oryzica-1	CT 10369
G	WC 5121 x CICA-8	CT 9585
H	WC 5121 x Oryzica-1	CT 9586

[1] WC 5103 = CT 6516-23-10-1-2-2 = TOX 1010-49-1 / IRAT 121 // Col.1 x M312A
WC 5121 = CT 6241-17-1-5-1 = Ngovie / Taipei 309 // Col.1 x M312A



1/ the regenerated plantlet from the regenerative medium and later transplanted to the field is R1; seed harvested out of this R1 plant is called R2 seed. Planting of this R2 seed will produce the R2 plants and, ultimately R3 seeds.

Appendix Figure 1. Diagrammatic description of the five sets of progenies compared in this research.

Appendix Table A2. Composition of media employed for rice anther culture.

Constituens	Potato media (mg/l)	Murashige & Skoog's media (mg/l)
NH_4NO_3	-.-	1,650
$(NH_4)_2SO_4$	100	-.-
KNO_3	1,000	1,900
KH_2PO_4	200	170
$MgSO_4.7H_2O$	126	370
$CaCl_2.2H_2O$	-.-	440
$Ca(NO_3)_2.4H_2O$	100	-.-
KCL	36	-.-
H_3BO_3	-.-	6.2
$MnSO_4.4H_2O$	-.-	22.3
$ZnSO_4.7H_2O$	-.-	8.6
$Na_2MoO_4.2H_2O$	-.-	0.25
$CuSO_4.5H_2O$	-.-	0.025
$CoCl_2.6H_2O$	-.-	0.025
KI	-.-	0.83
Na_2EDTA	37.3	37.3
$FeSO_4.7H_2O$	27.8	27.8
Myo-Inositol	-.-	100
Thiamine-HCL	1	0.1
Nicotinic acid	-.-	0.5
Pyridoxine	-.-	0.5
Glycine	-.-	2
NAA	4	1
Kinetin	1	4
Sucrose	50,000	30,000
Potato extract	100,000	-.-
Gel-rite	-.-	1,500

Appendix Table A3. Summary of climatic data on a per month basis for Huarangopampa Research Station (Bagua, Perú) and Palmira (Valle del Cauca, Colombia), July December 1990.

Location	Month	Rainfall (mm)	Temperature (°C)	
			Average Min.	Average Max.
Bagua	July	49.1	19.6	31.4
	August	11.9	20.3	32.3
	September	13.3	21.3	33.3
	October	54.8	21.9	33.4
	November	98.5	21.9	32.4
	December	71.7	20.6	31.6
Palmira	July	38.6	18.5	30.0
	August	5.8	19.1	30.9
	September	4.3	19.7	31.0
	October	142.7	19.1	28.2
	November	88.6	19.6	28.8
	December	47.9	19.3	28.7

Appendix Table A4. Observed mean squares for six agronomic traits of six parents planted at Palmira (Colombia) and Bagua (Perú), 1990.

Source of variation	df	Mean squares					
		Leaf pubes-cence¶	Days to maturity	Spikelet sterility (%)	Grains/ panicle	Panicles per m2	1000-gr. weight (g.)
Location†	1	0.003	143.85	1344.12**	28608.5**	95749.1**	46.11**
Block(Location)	52	0.005	52.88	47.11	573.8	1866.2	2.34
Genotypes††	5	14.061**	897.68**	1223.86**	11322.5**	24195.9**	123.00**
Location x Genotype††	5	0.000	11.42	732.00**	730.2	2335.9*	14.28**
Error	260	0.000	17.94	49.98	392.1	828.5	1.52
C.V.		0.0	2.8	37.1	16.8	16.5	4.9

¶ proportion of pubescent plants in the plot.
† test of hypothesis using the ANOVA MS for Block(Location) as an error term.
†† test of hypothesis using the ANOVA MS for Error as the error term.
* significant at the 0.05 probability level.
** significant at the 0.01 probability level.

Appendix Table A5. Observed mean squares for six agronomic traits from a combined analysis of variance of 16 populations derived from four rice crosses involving either doubled haploid or single seed descent breeding methods, at two locations, Bagua (Perú) and Palmira (Colombia), 1990.

Source of variation	df	Mean squares					
		Leaf pubes-cence¶	Days to maturity	Spikelet sterility (%)	Grains/ panicle	Panicles per m2	1000-gr. weight (g.)
Location = LOC†	1	0.011	15.29	2328.2**	886931.8**	1592022**	18.51
Block(LOC)	52	0.210	822.62	8.3	1862.7	15544	26.34
Population††	15	3.326**	304.12**	23.0**	6397.1**	6706**	265.10**
LOC x Population††	15	0.044	132.85**	4.2	2793.6**	3232	20.75**
Block x Population(LOC)	779	0.167	44.08	3.0	901.6	1788	7.61
Line(Population)	2444	0.301**	67.10**	3.6**	1154.9**	2187**	11.38**
Error	1525	1.0×10^{-9}	6.12	1.4	383.5	743	1.02
C.V. (%)		0.0	1.68	33.7	22.9	15.2	4.0

¶ proportion of pubescent plants in the plot.
† test of hypothesis using the ANOVA mean square for Block(Location) as an error term.
†† test of hypothesis using the ANOVA mean square for Block x Population(Location) as an error term.
* significant at the 0.05 probability level.
** significant at the 0.01 probability level.

Appendix Table A6. Observed mean squares for seven agronomic traits from an analysis of variance of 16 progenies derived from four rice crosses involving either doubled haploid or single seed descent breeding methods, grown at Bagua (Perú), 1990.

Source of variation	df	Mean squares†						
		Leaf pubescence¶	Days to maturity	Spikelet sterility (%)	Grains/ panicle	Panicles per m2	1000-gr. weight (g.)	Plant height (cm.)
Block	26	0.228	201.5**	519.2**	1250.7*	8134**	17.19**	453.6**
Population	15	1.510**	206.3**	1187.1**	6685.9**	3524**	148.06**	3286.1**
Cross	3	0.219	92.0	1925.1**	3815.7**	8872**	185.89**	4472.8**
Generation = Genr	1	1.863**	263.7*	178.9	4083.0*	1232	11.05	4798.9**
Cross x Genr	3	0.069	28.8	133.3	2288.4*	1047	21.42	347.1
Method(Genr)	1	4.766**	108.5	4222.6**	21111.5**	1232	431.00**	2939.8**
Cross x Method(Genr)	2	1.231**	69.3	756.2*	1239.1	1943	7.80	310.4
Selection(Genr)	1	1.053*	45.9	1325.0*	1269.8	2748	17.51	2403.6
Cross x Selection(Genr)	2	0.375	181.5*	353.4	1095.4	1464	14.47	1339.9**
Method x Selection(Genr)	1	0.027	3.2	210.7	2026.8	4474	110.80**	1126.6*
Cross x Met x Sel(Genr)	1	0.780*	327.2**	703.3	182.6	419	0.01	1284.1**
Population x Block	390	0.167	39.9	207.3	838.23	1300	8.68	186.5
Error	2026	0.175	39.2	153.2	750.65	1212	7.15	164.1
$\bar{x}$		0.636	146.94	28.13	98.85	160.86	25.10	99.1
C.V. (%)		64.3	4.3	44.0	27.7	21.5	10.6	12.9

† tests of hypothesis using the type III mean square for Population x Block as an error term.
¶ proportion of pubescent plants in the plot.
* significant at the 0.05 probability level.
** significant at the 0.01 probability level.

Appendix Table A7. Observed mean squares for seven agronomic traits from an analysis of variance of 16 populations derived from four rice crosses involving either doubled haploid or single seed descent breeding methods, grown at Palmira (Colombia), 1990.

Source of variation	df	Mean squares†						
		Leaf pubes-cence¶	Days to maturity	Spikelet sterility (%)	Grains/ panicle	Panicles per m2	1000-gr. weight (g.)	Grain disper-sion¶¶
Block	26	0.209	803.7**	594.4	711.6	12211**	20.89**	9.898**
Population	15	1.735**	224.9**	1608.4**	3018.9**	6517**	127.01**	33.495**
Cross	3	0.410	341.1**	1777.4**	2484.6	9084**	242.73**	71.836**
Generation = Genr	1	1.905**	6.4	191.4	1251.1	5917	26.21*	25.617**
Cross x Genr	3	0.068	202.8**	2103.9**	6690.9**	7850*	72.61**	8.978*
Method(Genr)	1	4.781**	2.6	1429.1	346.8	822	192.34**	1.567
Cross x Method(Genr)	2	1.302**	100.5	1416.7*	3806.9**	8961*	36.22**	9.195
Selection(Genr)	1	1.881**	198.8*	5720.7**	3714.4	4697	0.30	5.915
Cross x Selection(Genr)	2	0.515*	42.7	1166.7	3379.0**	13516**	2.25	0.999
Method x Selection(Genr)	1	0.003	11.6	674.9	892.8	2478	23.71	1.580
Cross x Met x Sel(Genr)	1	0.368	91.7	1317.1	961.3	4956	0.62	2.295
Population x Block	390	0.169	48.5	411.4	982.41	2278	6.69	3.491
Error	1965	0.188	47.4	407.1	991.85	2126	7.87	2.842
$\bar{x}$		0.632	146.83	41.99	71.85	197.04	24.99	4.251
C.V. (%)		68.6	4.7	48.1	43.8	23.4	11.2	39.7

† tests of hypothesis using the type III mean square for Population x Block as an error term.
¶ proportion of pubescent plants in the plot.
¶¶ spreading of milled rice treated with a 1.7% solution of KOH for 23 hours at 30°C ; 1-7 scale, where 1= non affected grain, 7= completely dispersed and intermingled kernels.
* significant at the 0.05 probability level.
** significant at the 0.01 probability level.

Appendix Table A8. Means ($\bar{x}$) and standard deviations (s.d) for percentage of sterility of 17 populations developed from four rice crosses involving either doubled haploid or single seed descent breeding methods, grown at Bagua (Perú), 1990.

Populations[2]		Crosses[1]			
		E	F	G	H
F1DH	N	60	103	81	117
	$\bar{x}$	27.6	30.3	27.5	31.7
	s.d	13.7	11.6	14.4	8.4
F2DH-R	N	94	39	66	69
	$\bar{x}$	31.3	30.6	28.6	38.1
	s.d	16.6	13.4	13.3	18.4
F2DH-S	N	-.-	-.-	83	104
	$\bar{x}$	-.-	-.-	29.1	32.3
	s.d	-.-	-.-	12.9	14.8
SSD-R	N	397	397	399	399
	$\bar{x}$	27.0	27.4	25.8	28.4
	s.d	12.0	11.1	11.3	12.8
SSD-S	N	-.-	49	199	199
	$\bar{x}$	-.-	23.8	24.4	27.7
	s.d	-.-	9.3	12.2	12.6
P1	N	27	27	27	27
	$\bar{x}$	20.0	20.0	13.9	13.9
	s.d	4.3	4.3	7.1	7.1
P2	N	27	27	27	27
	$\bar{x}$	13.2	21.6	15.3	18.4
	s.d	6.2	8.8	8.8	8.0

[1] E = WC5103/CICA-8; F = WC5103/ORYZICA-1;
G = WC5121/CICA-8; H = WC5121/ORYZICA-1.

[2] F1DH = doubled haploid (DH) lines derived from the F1;
F2DH-R = DH lines derived from random F2 plants;
F2DH-S = DH lines derived from selected F2 plants for leaf pubescence, early maturity, and desirable plant type;
SSD-R = F4 lines developed by single seed descent (SSD) from a sample of unselected F2 plants;
SSD-S = F4 lines developed by SSD from selected F2 plants for leaf pubescence, early maturity, and desirable plant type.

Appendix Table A9. Means ($\bar{x}$) and standard deviations (s.d) for percentage of sterility of 16 populations developed from four rice crosses involving either doubled haploid or single seed descent breeding methods, grown at Palmira (Colombia), 1990.

Populations[2]		Crosses[1]			
		E	F	G	H
F1DH	N	59	98	79	110
	$\bar{x}$	39.0	44.7	38.8	46.0
	s.d	15.9	19.0	23.6	21.0
F2DH-R	N	89	36	63	66
	$\bar{x}$	47.0	38.6	45.6	52.1
	s.d	20.7	18.1	23.6	25.7
F2DH-S	N	-.-	-.-	79	102
	$\bar{x}$	-.-	-.-	45.4	40.2
	s.d	-.-	-.-	22.8	19.9
SSD-R	N	-.-	390	395	394
	$\bar{x}$	-.-	39.7	42.2	43.3
	s.d	-.-	18.9	20.7	19.9
SSD-S	N	-.-	47	192	195
	$\bar{x}$	-.-	35.9	39.4	39.5
	s.d	-.-	16.6	19.3	19.1
P1	N	26	26	25	25
	$\bar{x}$	36.5	36.5	22.6	22.6
	s.d	7.8	7.8	7.9	7.9
P2	N	27	18	27	27
	$\bar{x}$	15.4	16.3	16.8	18.2
	s.d	5.6	4.8	5.2	7.9

[1] E = WC5103/CICA-8; F = WC5103/ORYZICA-1;
G = WC5121/CICA-8; H = WC5121/ORYZICA-1.

[2] F1DH = doubled haploid (DH) lines derived from the F1;
F2DH-R = DH lines derived from random F2 plants;
F2DH-S = DH lines derived from selected F2 plants for leaf pubescence, early maturity, and desirable plant type;
SSD-R = F4 lines developed by single seed descent (SSD) from a sample of unselected F2 plants;
SSD-S = F4 lines developed by SSD from selected F2 plants for leaf pubescence, early maturity, and desirable plant type.

Appendix Table A10. Means ($\bar{x}$) and standard deviations (s.d) for proportion of pubescent plants of 17 populations developed from four rice crosses involving either doubled haploid or single seed descent breeding methods, grown at Bagua (Perú), 1990.

Populations[2]		Crosses[1]			
		E	F	G	H
F1DH	N	60	103	81	117
	$\bar{x}$	0.55	0.53	0.54	0.53
	s.d	0.49	0.46	0.47	0.46
F2DH-R	N	94	39	66	69
	$\bar{x}$	0.57	0.68	0.50	0.41
	s.d	0.48	0.44	0.49	0.48
F2DH-S	N	-.-	-.-	83	104
	$\bar{x}$	-.-	-.-	0.56	0.53
	s.d	-.-	-.-	0.48	0.48
SSD-R	N	397	397	399	399
	$\bar{x}$	0.66	0.62	0.67	0.68
	s.d	0.42	0.41	0.41	0.38
SSD-S	N	-.-	49	199	199
	$\bar{x}$	-.-	0.60	0.87	0.71
	s.d	-.-	0.42	0.28	0.37
P1	N	27	27	27	27
	$\bar{x}$	0	0	0	0
	s.d	0	0	0	0
P2	N	27	27	27	27
	$\bar{x}$	1.0	1.0	1.0	1.0
	s.d	0	0	0	0

[1] E = WC5103/CICA-8; F = WC5103/ORYZICA-1;
G = WC5121/CICA-8; H = WC5121/ORYZICA-1.

[2] F1DH = doubled haploid (DH) lines derived from the F1;
F2DH-R = DH lines derived from random F2 plants;
F2DH-S = DH lines derived from selected F2 plants for leaf pubescence, early maturity, and desirable plant type;
SSD-R = F4 lines developed by single seed descent (SSD) from a sample of unselected F2 plants;
SSD-S = F4 lines developed by SSD from selected F2 plants for leaf pubescence, early maturity, and desirable plant type.

Appendix Table A11. Means ($\bar{x}$) and standard deviations (s.d) for pubescence of 16 populations developed from four rice crosses involving either doubled haploid or single seed descent breeding methods, grown at Palmira (Colombia), 1990.

Populations[2]		Crosses[1]			
		E	F	G	H
F1DH	N	59	98	79	110
	$\bar{x}$	0.52	0.53	0.54	0.51
	s.d	0.48	0.49	0.49	0.48
F2DH-R	N	89	36	63	66
	$\bar{x}$	0.56	0.68	0.47	0.39
	s.d	0.48	0.45	0.49	0.45
F2DH-S	N	-.-	-.-	79	102
	$\bar{x}$	-.-	-.-	0.58	0.54
	s.d	-.-	-.-	0.48	0.47
SSD-R	N	-.-	390	395	394
	$\bar{x}$	-.-	0.61	0.69	0.63
	s.d	-.-	0.43	0.42	0.42
SSD-S	N	-.-	47	192	195
	$\bar{x}$	-.-	0.59	0.90	0.71
	s.d	-.-	0.43	0.25	0.39
P1	N	26	26	27	27
	$\bar{x}$	0	0	0	0
	s.d	0	0	0	0
P2	N	27	19	27	27
	$\bar{x}$	1.0	1.0	1.0	1.0
	s.d	0	0	0	0

[1] E = WC5103/CICA-8; F = WC5103/ORYZICA-1;
G = WC5121/CICA-8; H = WC5121/ORYZICA-1.

[2] F1DH = doubled haploid (DH) lines derived from the F1;
F2DH-R = DH lines derived from random F2 plants;
F2DH-S = DH lines derived from selected F2 plants for leaf pubescence, early maturity, and desirable plant type;
SSD-R = F4 lines developed by single seed descent (SSD) from a sample of unselected F2 plants;
SSD-S = F4 lines developed by SSD from selected F2 plants for leaf pubescence, early maturity, and desirable plant type.

Appendix Table A12. Means ($\bar{x}$) and standard deviations (s.d) for days to maturity of 17 populations developed from four rice crosses involving either doubled haploid or single seed descent breeding methods, grown at Bagua (Perú), 1990.

Populations[2]		Crosses[1]			
		E	F	G	H
F1DH	N	60	103	81	117
	$\bar{x}$	146.8	147.8	147.1	147.8
	s.d	7.2	7.7	6.9	7.2
F2DH-R	N	94	39	66	69
	$\bar{x}$	144.6	147.2	145.6	147.3
	s.d	6.7	5.7	6.0	6.7
F2DH-S	N	-.-	-.-	83	104
	$\bar{x}$	-.-	-.-	147.8	144.7
	s.d	-.-	-.-	7.3	6.6
SSD-R	N	397	397	399	399
	$\bar{x}$	148.4	148.8	146.6	146.6
	s.d	6.4	6.8	6.2	5.8
SSD-S	N	-.-	49	199	199
	$\bar{x}$	-.-	147.2	146.3	146.3
	s.d	-.-	5.8	6.2	6.1
P1	N	27	27	27	27
	$\bar{x}$	150.1	150.1	141.7	141.7
	s.d	3.3	3.3	3.8	3.8
P2	N	27	27	27	27
	$\bar{x}$	150.6	149.7	153.5	150.6
	s.d	2.1	3.9	2.2	4.9

[1] E = WC5103/CICA-8; F = WC5103/ORYZICA-1;
G = WC5121/CICA-8; H = WC5121/ORYZICA-1.

[2] F1DH = doubled haploid (DH) lines derived from the F1;
F2DH-R = DH lines derived from random F2 plants;
F2DH-S = DH lines derived from selected F2 plants for leaf pubescence, early maturity, and desirable plant type;
SSD-R = F4 lines developed by single seed descent (SSD) from a sample of unselected F2 plants;
SSD-S = F4 lines developed by SSD from selected F2 plants for leaf pubescence, early maturity, and desirable plant type.

Appendix Table A13. Means ($\bar{x}$) and standard deviations (s.d) for days to maturity of 16 populations developed from four rice crosses involving either doubled haploid or single seed descent breeding methods, grown at Palmira (Colombia), 1990.

Populations[2]		Crosses[1]			
		E	F	G	H
F1DH	N	59	98	79	110
	$\bar{x}$	147.3	145.8	147.5	145.9
	s.d	7.6	8.4	7.6	8.6
F2DH-R	N	89	36	63	66
	$\bar{x}$	143.9	149.2	146.5	145.5
	s.d	9.1	5.6	8.1	8.4
F2DH-S	N	-.-	-.-	79	102
	$\bar{x}$	-.-	-.-	147.8	145.3
	s.d	-.-	-.-	7.4	9.2
SSD-R	N	-.-	390	395	394
	$\bar{x}$	-.-	147.4	148.0	145.6
	s.d	-.-	7.8	7.4	8.1
SSD-S	N	-.-	47	192	195
	$\bar{x}$	-.-	149.4	147.9	146.4
	s.d	-.-	6.4	7.3	8.3
P1	N	26	26	27	27
	$\bar{x}$	149.3	149.3	140.1	140.1
	s.d	5.0	5.0	10.0	10.0
P2	N	27	19	27	27
	$\bar{x}$	151.0	147.1	151.8	148.6
	s.d	3.3	5.3	3.3	6.1

[1] E = WC5103/CICA-8; F = WC5103/ORYZICA-1;
G = WC5121/CICA-8; H = WC5121/ORYZICA-1.

[2] F1DH = doubled haploid (DH) lines derived from the F1;
F2DH-R = DH lines derived from random F2 plants;
F2DH-S = DH lines derived from selected F2 plants for leaf pubescence, early maturity, and desirable plant type;
SSD-R = F4 lines developed by single seed descent (SSD) from a sample of unselected F2 plants;
SSD-S = F4 lines developed by SSD from selected F2 plants for leaf pubescence, early maturity, and desirable plant type.

Appendix Table A14. Means ($\bar{x}$) and standard deviations (s.d) for number of grains/panicle of 17 populations developed from four rice crosses involving either doubled haploid or single seed descent breeding methods, grown at Bagua (Perú), 1990.

Populations[2]		Crosses[1]			
		E	F	G	H
F1DH	N	60	103	81	117
	$\bar{x}$	101.5	95.9	88.7	88.1
	s.d	25.7	27.1	27.2	32.4
F2DH-R	N	94	39	66	69
	$\bar{x}$	89.7	101.3	95.3	92.0
	s.d	31.8	30.8	33.9	39.8
F2DH-S	N	-.-	-.-	83	104
	$\bar{x}$	-.-	-.-	87.8	83.6
	s.d	-.-	-.-	28.5	29.9
SSD-R	N	397	397	399	399
	$\bar{x}$	110.7	103.0	105.0	100.6
	s.d	26.8	25.7	26.3	26.6
SSD-S	N	-.-	49	199	199
	$\bar{x}$	-.-	107.8	102.9	99.6
	s.d	-.-	24.6	27.2	27.0
P1	N	27	27	27	27
	$\bar{x}$	119.4	119.4	104.0	104.0
	s.d	12.5	12.5	12.9	12.9
P2	N	27	27	27	27
	$\bar{x}$	149.5	124.7	136.1	127.0
	s.d	27.1	23.4	17.1	25.9

[1] E = WC5103/CICA-8; F = WC5103/ORYZICA-1;
G = WC5121/CICA-8; H = WC5121/ORYZICA-1.

[2] F1DH = doubled haploid (DH) lines derived from the F1;
F2DH-R = DH lines derived from random F2 plants;
F2DH-S = DH lines derived from selected F2 plants for leaf pubescence, early maturity, and desirable plant type;
SSD-R = F4 lines developed by single seed descent (SSD) from a sample of unselected F2 plants;
SSD-S = F4 lines developed by SSD from selected F2 plants for leaf pubescence, early maturity, and desirable plant type.

Appendix Table A15. Means ($\bar{x}$) and standard deviations (s.d) for number of grains per panicle of 16 populations developed from four rice crosses involving either doubled haploid or single seed descent breeding methods, grown at Palmira (Colombia), 1990.

Populations[2]		Crosses[1]			
		E	F	G	H
F1DH	N	59	98	79	110
	$\bar{x}$	79.2	69.4	78.4	71.2
	s.d	28.8	25.9	40.8	33.6
F2DH-R	N	89	36	63	66
	$\bar{x}$	61.9	80.4	71.4	65.7
	s.d	28.8	29.5	38.8	39.0
F2DH-S	N	-.-	-.-	79	102
	$\bar{x}$	-.-	-.-	63.3	70.9
	s.d	-.-	-.-	32.0	31.9
SSD-R	N	-.-	390	395	394
	$\bar{x}$	-.-	72.6	72.4	69.5
	s.d	-.-	29.1	31.9	30.8
SSD-S	N	-.-	47	192	195
	$\bar{x}$	-.-	84.1	74.4	74.5
	s.d	-.-	25.5	29.4	30.4
P1	N	26	26	25	25
	$\bar{x}$	100.2	100.2	81.6	81.6
	s.d	14.0	14.0	23.1	23.1
P2	N	27	19	27	27
	$\bar{x}$	123.7	121.6	113.3	109.2
	s.d	17.4	21.5	20.5	26.5

[1] E = WC5103/CICA-8; F = WC5103/ORYZICA-1;
G = WC5121/CICA-8; H = WC5121/ORYZICA-1.

[2] F1DH = doubled haploid (DH) lines derived from the F1;
F2DH-R = DH lines derived from random F2 plants;
F2DH-S = DH lines derived from selected F2 plants for leaf pubescence, early maturity, and desirable plant type;
SSD-R = F4 lines developed by single seed descent (SSD) from a sample of unselected F2 plants;
SSD-S = F4 lines developed by SSD from selected F2 plants for leaf pubescence, early maturity, and desirable plant type.

Appendix Table A16. Means ($\bar{x}$) and standard deviations (s.d) for 1000-grain weight (g.) of 17 populations developed from four rice crosses involving either doubled haploid or single seed descent breeding methods, at Bagua (Perú), 1990.

Populations[2]		Crosses[1]			
		E	F	G	H
F1DH	N	60	103	81	117
	$\bar{x}$	24.0	24.8	24.3	26.1
	s.d	2.2	2.5	3.2	3.3
F2DH-R	N	94	39	66	69
	$\bar{x}$	23.0	24.1	23.0	24.0
	s.d	2.4	3.6	2.8	3.6
F2DH-S	N	-.-	-.-	83	104
	$\bar{x}$	-.-	-.-	23.7	24.8
	s.d	-.-	-.-	3.0	2.9
SSD-R	N	397	397	399	399
	$\bar{x}$	24.5	25.7	24.9	26.3
	s.d	2.2	2.6	2.5	2.8
SSD-S	N	-.-	49	199	199
	$\bar{x}$	-.-	26.0	24.3	25.7
	s.d	-.-	2.6	2.4	2.7
P1	N	27	27	27	27
	$\bar{x}$	26.7	26.7	26.3	26.3
	s.d	0.8	0.8	1.9	1.9
P2	N	27	27	27	27
	$\bar{x}$	23.2	26.5	22.9	25.6
	s.d	1.5	0.7	0.9	0.6

[1] E = WC5103/CICA-8; F = WC5103/ORYZICA-1;
G = WC5121/CICA-8; H = WC5121/ORYZICA-1.

[2] F1DH = doubled haploid (DH) lines derived from the F1;
F2DH-R = DH lines derived from random F2 plants;
F2DH-S = DH lines derived from selected F2 plants for leaf pubescence, early maturity, and desirable plant type;
SSD-R = F4 lines developed by single seed descent (SSD) from a sample of unselected F2 plants;
SSD-S = F4 lines developed by SSD from selected F2 plants for leaf pubescence, early maturity, and desirable plant type.

Appendix Table A17. Means ($\bar{x}$) and standard deviations (s.d) for 1000-grain weight of 16 populations developed from four rice crosses involving either doubled haploid or single seed descent breeding methods, at Palmira (Colombia), 1990.

Populations[2]		Crosses[1]			
		E	F	G	H
F1DH	N	59	98	79	110
	$\bar{x}$	23.3	23.9	24.5	26.9
	s.d	2.5	2.9	3.1	3.9
F2DH-R	N	89	36	63	66
	$\bar{x}$	22.6	23.7	24.1	24.1
	s.d	2.9	2.8	2.6	2.6
F2DH-S	N	-.-	-.-	79	102
	$\bar{x}$	-.-	-.-	24.7	24.5
	s.d	-.-	-.-	3.4	2.9
SSD-R	N	-.-	390	395	394
	$\bar{x}$	-.-	25.0	24.9	26.1
	s.d	-.-	2.5	2.6	2.8
SSD-S	N	-.-	47	192	195
	$\bar{x}$	-.-	25.0	24.6	25.7
	s.d	-.-	2.5	2.3	3.0
P1	N	26	26	25	25
	$\bar{x}$	24.8	24.8	26.4	26.4
	s.d	1.7	1.7	1.9	1.9
P2	N	27	19	27	27
	$\bar{x}$	23.0	25.6	23.4	24.5
	s.d	0.9	0.8	0.9	1.7

[1] E = WC5103/CICA-8; F = WC5103/ORYZICA-1;
G = WC5121/CICA-8; H = WC5121/ORYZICA-1.

[2] F1DH = doubled haploid (DH) lines derived from the F1;
F2DH-R = DH lines derived from random F2 plants;
F2DH-S = DH lines derived from selected F2 plants for leaf pubescence, early maturity, and desirable plant type;
SSD-R = F4 lines developed by single seed descent (SSD) from a sample of unselected F2 plants;
SSD-S = F4 lines developed by SSD from selected F2 plants for leaf pubescence, early maturity, and desirable plant type.

Appendix Table A18. Means ($\bar{x}$) and standard deviations (s.d) for number of panicles per square meter of 17 populations developed from four rice crosses involving either doubled haploid or single seed descent breeding methods, at Bagua (Perú), 1990.

Populations[2]		Crosses[1]			
		E	F	G	H
F1DH	N	60	103	81	117
	$\bar{x}$	173.0	162.7	164.4	159.0
	s.d	34.9	34.3	37.6	40.1
F2DH-R	N	94	39	66	69
	$\bar{x}$	172.4	155.3	158.4	151.8
	s.d	37.8	40.2	41.9	36.8
F2DH-S	N	-.-	-.-	83	104
	$\bar{x}$	-.-	-.-	161.0	167.2
	s.d	-.-	-.-	42.1	50.2
SSD-R	N	397	397	399	399
	$\bar{x}$	166	158.8	164.3	156.3
	s.d	33.6	33.4	37.9	33.5
SSD-S	N	-.-	49	199	199
	$\bar{x}$	-.-	155.9	162.3	158.5
	s.d	-.-	33.6	34.8	34.8
P1	N	27	27	27	27
	$\bar{x}$	136.8	136.8	129.1	129.1
	s.d	17.5	17.5	24.2	24.2
P2	N	27	27	27	27
	$\bar{x}$	172.6	152.7	181.5	172.4
	s.d	24.1	26.3	27.8	31.8

[1] E = WC5103/CICA-8; F = WC5103/ORYZICA-1;
G = WC5121/CICA-8; H = WC5121/ORYZICA-1.

[2] F1DH = doubled haploid (DH) lines derived from the F1;
F2DH-R = DH lines derived from random F2 plants;
F2DH-S = DH lines derived from selected F2 plants for leaf pubescence, early maturity, and desirable plant type;
SSD-R = F4 lines developed by single seed descent (SSD) from a sample of unselected F2 plants;
SSD-S = F4 lines developed by SSD from selected F2 plants for leaf pubescence, early maturity, and desirable plant type.

Appendix Table A19. Means ($\bar{x}$) and standard deviations (s.d) for panicles per square meter of 16 populations developed from four rice crosses involving either doubled haploid or single seed descent breeding methods, at Palmira (Colombia), 1990.

Populations[2]		Crosses[1]			
		E	F	G	H
F1DH	N	59	98	79	110
	$\bar{x}$	185.9	201.4	210.3	199.8
	s.d	39.6	45.2	43.1	49.9
F2DH-R	N	89	36	63	66
	$\bar{x}$	208.7	193.2	206.6	203.2
	s.d	57.5	52.3	56.0	63.0
F2DH-S	N	-.-	-.-	79	102
	$\bar{x}$	-.-	-.-	191.2	211.7
	s.d	-.-	-.-	48.4	68.5
SSD-R	N	-.-	390	395	394
	$\bar{x}$	-.-	194.5	195.9	191.5
	s.d	-.-	43.4	46.8	41.8
SSD-S	N	-.-	47	192	195
	$\bar{x}$	-.-	179.2	198.9	197.1
	s.d	-.-	34.0	49.6	45.3
P1	N	26	26	25	25
	$\bar{x}$	175.0	175.0	157.3	157.3
	s.d	41.7	41.7	26.0	26.0
P2	N	27	17	27	27
	$\bar{x}$	222.5	206.5	203.0	192.1
	s.d	27.0	65.6	34.2	28.3

[1] E = WC5103/CICA-8; F = WC5103/ORYZICA-1;
G = WC5121/CICA-8; H = WC5121/ORYZICA-1.

[2] F1DH = doubled haploid (DH) lines derived from the F1;
F2DH-R = DH lines derived from random F2 plants;
F2DH-S = DH lines derived from selected F2 plants for leaf pubescence, early maturity, and desirable plant type;
SSD-R = F4 lines developed by single seed descent (SSD) from a sample of unselected F2 plants;
SSD-S = F4 lines developed by SSD from selected F2 plants for leaf pubescence, early maturity, and desirable plant type.

Appendix Table A20. Means ($\bar{x}$) and standard deviations (s.d) for plant height (cm.) of 17 populations developed from four rice crosses involving either doubled haploid or single seed descent breeding methods, at Bagua (Perú), 1990.

Populations[2]		Crosses[1]			
		E	F	G	H
F1DH	N	60	103	81	117
	$\bar{x}$	91.6	93.3	89.6	98.2
	s.d	9.6	12.5	14.4	15.8
F2DH-R	N	94	39	66	69
	$\bar{x}$	92.6	100.7	94.3	104.6
	s.d	11.7	12.3	16.3	15.4
F2DH-S	N	-.-	-.-	83	104
	$\bar{x}$	-.-	-.-	93.2	93.6
	s.d	-.-	-.-	15.5	16.5
SSD-R	N	397	397	399	399
	$\bar{x}$	99.0	99.5	99.3	105.6
	s.d	10.9	10.3	12.6	13.8
SSD-S	N	-.-	49	199	199
	$\bar{x}$	-.-	99.5	98.4	102.9
	s.d	-.-	8.1	13.3	13.4
P1	N	27	27	27	27
	$\bar{x}$	98.5	98.5	90.4	90.4
	s.d	4.5	4.5	5.7	5.7
P2	N	27	27	27	27
	$\bar{x}$	99.4	101.2	96.7	104.6
	s.d	7.4	5.3	6.4	6.5

[1] E = WC5103/CICA-8; F = WC5103/ORYZICA-1;
G = WC5121/CICA-8; H = WC5121/ORYZICA-1.

[2] F1DH = doubled haploid (DH) lines derived from the F1;
F2DH-R = DH lines derived from random F2 plants;
F2DH-S = DH lines derived from selected F2 plants for leaf pubescence, early maturity, and desirable plant type;
SSD-R = F4 lines developed by single seed descent (SSD) from a sample of unselected F2 plants;
SSD-S = F4 lines developed by SSD from selected F2 plants for leaf pubescence, early maturity, and desirable plant type.

Appendix Table A21. Means ($\bar{x}$) and standard deviations (s.d) for endosperm dispersion of 16 populations developed from four rice crosses involving either doubled haploid or single seed descent breeding methods, at Palmira (Colombia), 1990.

Populations[2]		Crosses[1]			
		E	F	G	H
F1DH	N	58	100	77	111
	$\bar{x}$	3.8	4.7	3.2	4.0
	s.d	1.7	2.0	1.6	1.8
F2DH-R	N	86	38	62	64
	$\bar{x}$	3.4	5.0	3.8	4.5
	s.d	1.6	2.1	1.7	1.8
F2DH-S	N	-.-	-.-	77	98
	$\bar{x}$	-.-	-.-	4.1	4.7
	s.d	-.-	-.-	1.7	2.0
SSD-R	N	-.-	386	386	393
	$\bar{x}$	-.-	4.5	3.8	4.6
	s.d	-.-	1.8	1.6	1.7
SSD-S	N	-.-	47	194	191
	$\bar{x}$	-.-	4.5	3.7	4.9
	s.d	-.-	1.8	1.7	1.8
P1	N	9	9	9	9
	$\bar{x}$	3.8	3.8	2.1	2.1
	s.d	2.6	2.6	0.2	0.2
P2	N	9	9	9	9
	$\bar{x}$	4.12	6.2	3.9	7.0
	s.d	0.8	1.4	0.7	0

[1] E = WC5103/CICA-8; F = WC5103/ORYZICA-1;
G = WC5121/CICA-8; H = WC5121/ORYZICA-1.

[2] F1DH = doubled haploid (DH) lines derived from the F1;
F2DH-R = DH lines derived from random F2 plants;
F2DH-S = DH lines derived from selected F2 plants for leaf pubescence, early maturity, and desirable plant type;
SSD-R = F4 lines developed by single seed descent (SSD) from a sample of unselected F2 plants;
SSD-S = F4 lines developed by SSD from selected F2 plants for leaf pubescence, early maturity, and desirable plant type.

Appendix Table A22. Mean squares for six agronomic traits of five contrasts between progenies derived from four irrigated/upland rice crosses, grown at Bagua, 1990.

Contrasts	Leaf pubes-cence¶	Days to maturity	Spikelet sterility (%)	Grains/ panicle	Panicles per m2	1000-gr. weight (g.)	Plant height (cm.)
1. F1DH vs F2DH-R	0.02	257.9	869.7*	346	6113*	215.4***	3726.6***
2. F2DH-R vs F2DH-S	0.64	0.7	605.7	4375*	6282*	48.6	2878.4***
3. F2DH-R vs SSD-R	2.24***	545.1***	4260.8***	19375***	2202	676.7***	1301.7**
4. SSD-R vs SSD-S	1.11*	94.3	764.3	164	94	16.8	337.7
5. F1DH vs SSD-R	3.84***	21.2	1221.9*	36576***	3193	94.9***	16929.8***

¶ proportion of pubescent plants in the plot.
* significant at the 0.05 probability level.
** significant at the 0.01 probability level.
*** significant at the 0.001 probability level.

Appendix Table A23. Mean squares for six traits of five contrasts between progenies derived from four irrigated/upland rice crosses, grown at Palmira, 1990.

Contrasts	Leaf pubes-cence¶	Days to maturity	Spikelet sterility (%)	Grains/ panicle	Panicles per m2	1000-gr. weight (g.)	Grain disper-sion¶¶
1. F1DH vs F2DH-R	0.01	14.8	1610.6	2454	41	96.3***	7.06
2. F2DH-R vs F2DH-S	1.28**	51.5	3239.9**	22	488	6.5	4.26
3. F2DH-R vs SSD-R	1.84**	1.8	1584.6*	365	1972	170.8***	0.56
4. SSD-R vs SSD-S	1.61**	190.8	3265.7**	10264**	2620	9.5	1.66
5. F1DH vs SSD-R	2.77***	57.0	445.2	779	21685**	17.6	32.99**

¶ proportion of pubescent plants in the plot.
¶¶ spreading of milled rice treated with a 1.7% solution of KOH for 23 hours at 30°C ; 1-7 scale, where 1= non affected grain, 7= completely dispersed and intermingled kernels.
* significant at the 0.05 probability level.
** significant at the 0.01 probability level.
*** significant at the 0.001 probability level.