# Recent advances in breeding for improving iron utilization by plants

S.R. RODRIGUEZ DE CIANZIO

Department of Agronomy, Iowa State University, Ames, IA 50011, USA

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## Abstract

Iron-chlorosis deficiency may occur when an iron-inefficient genotype is grown on calcareous soil. One way to correct the problem is to modify the genotype by plant breeding. Cultivars have been released for oat (Avena byzantina C. Koch), sorghum [Sorghum bicolor (L.) Moench], dry bean (Phaseoulus vulgaris L.), and soybean [Glycine max (L.) Merr.]. Progress is being made in peanut (Arachis hypogea L.), forage species such as clovers (Trifolium sp.) and bluestems (Botriochloa sp.), and pepper (Capsicum annuum L.). Screening of rootstocks is done on citrus (Citrus sp.), mango (Manguifera indica L.), and avocado (Persea americana Mill.).

# Introduction

Iron-chlorosis deficiency may occur when an iron-inefficient genotype is grown on calcareous soil. One way to correct the problem is to modify the genotype by plant breeding.

Iron chlorosis is generally recognized as a real or potential problem in many crops species (Clark and Gross, 1986). Improvement in ironchlorosis deficiency by plant breeding, however, is conducted in only a few crop species (Fehr, 1984). The objective of this review is to discuss progress and current research in plant breeding for the improvement of iron utilization of cultivars. Recent published reports, information obtained from a questionnaire sent to scientists working with iron chlorosis in the United States and other countries, and current research are summarized.

Cultivars and germplasm with improved resistance to iron chlorosis have been released for oat (Avena byzantina C. Koch), sorghum [Sorghum bicolor (L.) Moench], dry bean (Phaseolus vulgaris L.), and soybean [Glycine max (L.) Merr.]. Breeders of peanut (Arachis hypogea L.), forage species such as clovers (Trifolium sp.) and bluestems (Botriochloa sp.), and pepper (Capsicum annum L.) are also engaged in improving ironchlorosis resistance. Screening and selection of rootstocks is done in fruit crops such as citrus (*Citrus* sp.), mango (*Manguifera indica* L.), and avocado (*Persea americana* Mill.).

# Oat

Breeding for iron-chlorosis resistance in oat is conducted at Texas A and M University (M.E. McDaniel, 1989, Texas A and M Univ., pers. commun.). Approximately 60% of the acreage of oat in Texas is planted on calcareous soil. Selection for iron efficiency is conducted simultaneously with selection for resistance to crown and stem rusts. Iron-deficiency symptoms in oat, as in other species, are difficult to evaluate because they disappear during plant development. Symptoms are more frequently present during cool periods. Oat is used as forage in Texas; therefore, the main concern in yield reduction is loss of foliage. Under severe conditions, McDaniel (1989, Texas A and M Univ., pers. commun.) estimates that 50% yield reduction in both foliage and grain is possible. Recently, a

cultivar, TAM 0 386, has been released having good resistance to iron chlorosis.

In the oat breeding program at Texas A and M, crosses are made between genotypes resistant to iron chlorosis and high-yielding parents, and generations are advanced by the pedigree method. Evaluation of parents and progenies are conducted in the field on calcareous soil. Attempts at using nutrient-solution culture for screening have been unsuccessful (M.E. McDaniel, 1989, Texas A and M University, pers. commun.). The program continues to use field evaluations on calcareous soils, using a rating score from 0 to 9, with 0 being the most resistant genotype. Selection of resistant plants in the field is conducted in early generations. Individual, resistant plants are harvested and seed from each is planted the next year in a head-row. Superior plants are selected within each row until uniformity of plant types and traits are observed. Selections are yield-tested in the  $F_4$  or  $F_5$  generation and reselection is done at that time if necessary.

Inheritance studies indicate that iron efficiency may be simply inherited, probably as a single gene with resistance being dominant (McDaniel and Brown, 1982).

## Sorghum

Selection for resistance is conducted in breeding programs of sorghum at Texas A and M University, Kansas State University, and the University of Nebraska-Lincoln (Clark et al., 1988) (W.B. Anderson, 1989, Texas A and M Univ., pers. commun.; P.J. Bramel-Cox, 1989, Kansas State Univ., pers. commun.). Yield losses of 32% per unit of increase in chlorosis rating have been predicted by using a scale from 1 (no yellowing) to 5 (severe yellowing) (Clark et al., 1988). Anderson (1989, Texas A and M Univ., pers. commun.) and Bramel-Cox (1989, Kansas State Univ., pers. commun.) estimate that, in farmers' fields, 20% yield loss may result in moderate cases of iron deficiency, increasing up to 100% under severe conditions.

In 1985, two bulk populations, Kansas Bulk 24 and Kansas Bulk 25, had been identified by Dan Rodgers (deceased) for superior iron-efficiency and were ready for release (P.J. Bramel-Cox, 1989, Kansas State Univ., pers. commun.). The bulks were the result of three cycles of recurrent phenotypic selection on calcareous soils. Recurrent selection is also being used at Nebraska (Williams *et al.*, 1987).

Evaluation of parents and their progeny is conducted in the field on calcareous soils at Kansas State and at Nebraska. At Texas A and M, Anderson (1989, Texas A and M Univ., pers. commun.) evaluates lines in the field on calcareous soils and in the greenhouse by using calcareous soil and nutrient solution culture. Williams *et al.* (1987), when comparing growth-chamber experiments using nutrient-solution culture and field tests, found poor correlation between ironchlorosis estimated by field and by growth-chamber environments.

Inheritance of iron efficiency in sorghum is quantitative. Broad-sense heritability, estimated from means of entries over 2 years for the average chlorosis rating was  $0.86 \pm 0.15$  (Williams *et al.*, 1987).

## Dry bean

Selection for resistance to iron chlorosis is conducted at the University of Nebraska-Lincoln as part of the ongoing breeding program that includes selection for multiple traits (D.P. Coyne, 1989, Univ. of Nebraska, pers. commun.). Research is underway to obtain estimates of yield loss due to iron inefficiency (H.Z. Zaiter, 1989, Univ. of Nebraska, pers. commun.).

Cultivars released from this program have adequate levels of resistance to iron chlorosis. Two cultivars recently released, Great Northern Valley and Neb-WM1-83-10, had mean chlorosis ratings, averaged over replications, of 1.0 after two years of field testing on calcareous soil (Zaiter *et al.*, 1987). The authors used a scale for chlorosis ratings similar to the scale used in soybean and sorghum (Cianzio and Fehr, 1980; Clark *et al.*, 1988).

Single-plant selection in segregating progenies of early generations is used with mass, pedigree, or backcross breeding methods. Routine evaluation of parents and their progeny is conducted on calcareous soils and in the greenhouse by using calcareous soil in pots (D.P. Coyne, 1989, Univ. of Nebraska, pers. commun.). Concern with identifying small differences in resistance under laboratory conditions compared with those observed in the field prompted research to evaluate chlorosis resistance in the growth chamber by using a nutrient-solution culture. Low correlations (r = 0.27) found between iron chlorosis under field conditions and in nutrient solution showed the inadequacy of using this nutrient solution (Zaiter *et al.*, 1986). The authors have not yet evaluated the nutrient solution proposed by Chaney *et al.* (1989) (D.P. Coyne, 1989,

Univ. of Nebraska, pers. commun.).

Inheritance of iron efficiency in dry beans is determined by two major genes (Coyne *et al.*, 1982). In some crosses, complementary gene action between two dominant genes has been reported (Zaiter *et al.*, 1987). Although the trait is simply inherited, environmental effects cause continuous variation (Coyne *et al.*, 1982; Zaiter *et al.*, 1988). Realized heritabilities of 0.52 and 0.49, based on the performance of  $F_3$  families from two crosses, have been obtained. Narrowsense heritabilities for 13 crosses ranging from 0.29 to 0.75 have been obtained by regressing  $F_3$ progeny means on individual  $F_2$  plants (Zaiter *et al.*, 1988).

# Soybean

Improvement of iron-chlorosis in soybean is a breeding objective of programs of private seed companies, and public institutions such as Iowa State University and the University of Minnesota.

Yield losses have been estimated to increase 20% for each unit increase of chlorosis score (Froehlich and Fehr, 1981). Orf (1989, Univ. of Minnesota, pers. commun.) estimates yield reductions in farmers' fields ranging from 1-10%, depending on the severity of symptoms.

Progress has been made in developing improved germplasm and in developing cultivars with adequate levels of resistance. Five germplasm lines have recently been released from the program at Iowa State, A11, A12, A13, A14, and A15 (Jessen *et al.*, 1988). These lines were developed by use of recurrent selection with  $S_1$  testing. Testing procedures were conducted in the field on calcareous soil, in the greenhouse using nutrient-solution culture, and in the field on calcareous soil by cutting off the main stem of plants above the unifoliolate node at stage V3 to enhance differentiation for iron-chlorosis resistance (Piper *et al.*, 1986).

In some programs, testing is conducted only under field conditions on calcareous soils, or with a combination of field and greenhouse tests using nutrient-solution culture (Jessen et al., 1986; 1988). Various nutrient solutions have been tried with soybean to identify a system that provides chlorosis ratings highly correlated with those derived from field evaluations, that minimizes cost and labor, that controls the severity of chlorosis, and that reduces environmental variability (Jessen et al., 1986; 1988). A rank correlation of 0.98 between field scores and those of the best nutrient solution were reported from a test using eight cultivars with a wide range of iron efficiency (Jessen et al., 1988). Use of growth chambers for evaluation of genotypes in potted calcareous soil has also been evaluated (Fairbanks et al., 1987), but results have been inconclusive.

Breeding methodology used depends on the program. Single-cross, three-way cross, and backcross populations are used. A study comparing single-cross with backcross populations indicated highly resistant parents could be used effectively in either single-cross or backcross populations (Hintz *et al.*, 1987). Recurrent selection with  $S_1$  testing is used at Iowa State for developing a population with superior resistance to iron-deficiency chlorosis (Fehr, 1983). This population has been a useful source of resistant germ-plasm for breeding programs (Jessen *et al.*, 1988).

Inheritance of the trait has been explained by a single major gene and modifying genes and as a quantitative character controlled by additive gene action (Cianzio and Fehr, 1980; 1982). The trait is considered quantitative for breeding purposes (Fehr, 1983).

Effectiveness of selection among single plants for iron-efficiency in a segregating soybean population in either field or nutrient solution culture tests was studied by Diers and Fehr (Diers and Fehr, 1989). Research was planned to determine

if the nutrient-solution system would increase the precision of single-plant selection compared with field selection. Six single-cross populations derived from crossing parents with high yield and moderate iron efficiency were used. Single-plant selection for iron efficiency in the field or in nutrient solution was of limited value. Heritability estimates and genetic gains for single-plant selection were small, perhaps because of the type of populations used in the study. Parents of the populations were selected for the best combination of iron efficiency and yield among cultivars, brands, and experimental lines available in 1984. The largest difference in chlorosis scores between parents was 0.4. The parents would be similar to those commonly used by breeders to develop cultivars with high yield and high iron efficiency. Similarity between field and nutrientsolution evaluation for heritabilities estimated by parent-offspring regression suggest that there was no detectable difference between evaluation methods in how well the chlorosis score of the parent predicted the chlorosis score of the progeny. Selection based on progeny tests would be recommended over single-plant selection in populations similar to those used in this research.

## Peanut

Breeding of peanut is conducted at The Volcani Center of the Agricultural Research Organization in Israel (A. Harzook, 1989, The Volcani Center, Israel, pers. commun.). Currently, no estimates of yield loss due to iron chlorosis are available. A crossing program using resistant parents is underway. Parental lines and progenies are evaluated in the field on calcareous soil and also in the greenhouse by using nutrientsolution culture.

## **Forage species**

Breeding and selection work in forages is conducted at Texas A and M University for clovers (R. Smith, 1989, Texas A and M Univ., pers. commun.). Average yield reduction of lines grown on calcareous soil has been estimated at approximately 25%. Yield reduction has not been observed when lines were grown on non-calcareous soil.

Variation among clovers for resistance to irondeficiency chlorosis has been reported from field and greenhouse evaluations, indicating that selection for resistance to iron chlorosis may be possible within these species (Gildersleeve and Ocumpaugh, 1988; 1989; Gildersleeve *et al.*, 1988).

Screening of arrowleaf clover (*Trifolium ves-iculosum*) is conducted in high-pH, water-saturated soils in the greenhouse (R. Smith, 1989, Texas A and M Univ., pers. commun.). Selected plants are transferred to cages for crossing with bees. During 1989, cycle 3 seed will be produced from this program. Inheritance of the trait in clovers seems dependent on several genes.

Screening and selection for resistant genotypes within the available germplasm of bluestem (*Botriochloa* sp.) is conducted at the Southern Plains Range Research Station, USDA-ARS, Woodward, Oklahoma (Berg *et al.*, 1986). Symptoms are temporary in bluestems and estimates of yield are not available.

Diversity found among Old World bluestems indicates that, with additional collections and screening, there is a high potential to select iron-efficient strains. Hybridization in nature and natural selection followed by strain stabilization through apomixis have resulted in a vast array of genetic diversity within the species (Berg et al., 1986). These authors consider that screening and selection at early stages of a plant breeding program would suffice, under present circumstances, to correct the iron-deficiency problem. They have pointed out, however, that artificial hybridization of genotypes followed by selection, although costly and time-consuming, will eventually be necessary to make further progress in correcting this trait.

## Pepper

Selection for resistance is conducted at The Volcani Center of the Agricultural Research Organization in Israel (C. Shifriss, 1989, The Volcani Center, Israel, pers. commun.). Inheritance studies conducted in pepper indicate that efficiency is controlled by a single dominant gene (Shifriss and Eidelman, 1983). Crosses are obtained by using resistant parents, and selection of progenies is conducted in the field.

#### Fruit crops

In citrus species, selection for resistance is done at Texas A and I University Citrus Center (D. Swietlik, 1989, Texas A and I Univ. Citrus Center, pers. commun.). Striking differences between citrus cultivars and species have been found. It is estimated that approximately 80– 90% of the citrus orchards in the Lower Rio Grande Valley of Texas are located on calcareous soils. There are no estimates concerning economic losses.

Selection of rootstocks for resistance to irondeficiency chlorosis in mango and avocado is conducted at the Eden Experimental Farm in Bet-Shean Valley, Israel (G. Fischler, 1989, Eden Experimental Farm, Israel, pers. commun.; Y. Chen, 1989, the Hebrew Univ. of Jerusalem, Israel, pers. commun.). Testing of iron-chlorosis deficiency is conducted in three steps. First, effectiveness in physiological response of genotypes to cation reduction and acidification of nutrient solution is measured. Second, genotypes classified effective are planted in the greenhouse in potted soil. Third, genotypes selected from these plantings are evaluated in the field.

#### Conclusions

Progress has been made in both basic and applied research in breeding for improved ironchlorosis deficiency. In some crop species, cultivars and germplasm have been released with resistance to iron chlorosis. For other species, screening and selection work is underway. Current research indicates that further progress will be made in the near future as new knowledge is acquired in the mechanisms responsible for iron utilization by the plant, new and improved screening techniques are developed, and appropriate breeding methods are used.

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