Research Application Summary

Enhancing resistance to angular leaf spot of common bean in Uganda

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Abstract

Angular leaf spot (ALS) is an important disease of common bean in Uganda. The presence of resistance sources with narrow adaptability coupled with pathogen's variability have greatly undermined previous breeding efforts leading to severe yield losses. This study seeks to identify new adapted resistance sources within the local germplasm and pyramid the exotic resistance sources to broaden ALS resistance. Two *Pseudocercospora griseola* isolates; 1:6 and 21:7 were used to screen 65 and 4 check genotypes. In addition, parental crosses involving three genotypes carrying *Phg 1*, *Phg 2* and Phg_{G5686A} genes were made to accumulate three resistance genes into one genetic background. Preliminary results have indicated that landraces U0053 and U0074 are resistant to both isolates. Hence the two landraces offer opportunity of being used as breeding materials for ALS resistance. However, further screening with more virulent isolates is on-going. On the other hand, fifteen parental crosses of Mexico 54, AND 277 and G5686 that are involved in gene pyramiding have been done and advanced to F_1 and F_2. Selection using molecular markers for F_2 progenies is planned.

Key words: Gene pyramiding, *Pseudocercospora griseola*, resistance, screening

Résumé

La tache angulaire de la feuille (ALS) est une maladie importante du haricot ordinaire en Ouganda. La présence de sources de résistance avec une capacité limitée d'adaptation couplée à la variabilité pathogène a grandement miné les efforts précédents de reproduction conduisant à des pertes de rendement sévères. Cette étude cherche à identifier de nouvelles sources de résistance adaptées au sein du germoplasme local et de la pyramide locale des sources de résistance exotiques afin d'élargir la résistance de l'ALS. Deux isolats *Pseudocercosporagriseola*; 1:6 et 21:07 ont été utilisés pour dépister 65 et 4 génotypes à cocher. En outre, les croix parentales impliquant trois génotypes porteurs de gènes *Phg 1, Phg 2* et

*Phg*G5686A ont été faites pour accumuler trois gènes de résistance dans un seul contexte génétique. Les résultats préliminaires ont indiqué que les variétés U0053 et U0074 sont résistantes aux deux isolats. Par conséquent, les deux variétés locales offrent la possibilité d'être utilisées comme matériaux de reproduction pour une résistance de l'ALS. Toutefois, un examen plus poussé avec des isolats plus virulents est en cours. Par contre, quinze croix parentales du Mexique 54, AND 277 et G5686 qui sont impliquées dans le cumul des gènes ont été faites et avancées à F_1 et F_2. La sélection à l'aide de marqueurs moléculaires pour les descendances de F_2 est prévue.

Mots clés: Cumul des gènes, *Pseudocercosporagriseola*, résistance, dépistage

Background

Common bean (*Phaseolus vulgaris* L.*)* is one of the most nutritionally valuable crops globally. Despite its importance yield losses of up to 80% are incurred due to angular leaf spot (ALS) caused by *Pseudocercospora griseola* (Stenglein *et al.*, 2003). Such losses affect the livelihood and nutritional well-being of millions of people who depend on beans for food. In Uganda yield loss of 50% due to ALS in released bean varieties has been reported (Opio *et al.*, 2007). This affects the protein dietary intake of about 45% of Ugandans whose dietary protein is derived majorly from beans. Genetic resistance is strategically important in managing ALS given the pathogen's high variability (Mahuku *et al.*, 2009). However, resistance sources identified elsewhere have narrow adaptability and others are not effective due to variants of the pathogen that often breaks down the resistance. This limits the use of such available sources of resistance. Therefore there is need to identify new resistance source in adapted germplasm and broaden resistance in the exotic germplasm. The study seeks to identify new sources of resistance and broaden resistance in the exotic germplasm through gene pyramiding.

Literature Summary

Angular leaf spot is a major threat to bean cultivation. Pyramiding resistance genes with additive action located on different loci can effectively be used to improve resistance to ALS (Werner *et al.*, 2005). This is because it allows combining more genes in one genotype to provide broad and durable resistance (Zheng *et al.*, 2006). Although gene pyramiding sounds to be one of the strategies to overcome ALS,it has been a big challenge due to complications associated with selecting plants with multiple resistance genes based on phenotype

(Mohler and Singrun, 2004). Hence, marker assisted selection using tightly linked gene specific markers is of great importance in improving selection efficiency during gene pyramiding to enhance the effectiveness of resistance to ALS. On the other hand, resistance on which pyramiding is based is critical. One step in achieving resistance involves testing of the available germplasm. However, the high pathogenic variability occurring in angular leaf spot (Mahuku *et al.*, 2009) dictates that in order to identify durable resistance, the reaction of available germplasm to different pathogen populations need to be established and combined to offer best resistance, hence the need to screen the available germplasm for promising resistance accessions.

Study Description

Sixty five Ugandan bean landraces were preliminarily screened with two ALS isolates (Mesoamerica; 1-6 and Andean; 21-7) obtained from International Centre for Tropical Agricultural (CIAT)-Kawanda. Thirty bean accessions which were found to be susceptible to the two isolates in the initial screening were discarded and the remaining 35 landraces, two resistant (Mexico 54 and BAT 322) and two susceptible (Kanyebwa and K131) checks were re-screened against the same isolates to confirm their level of resistance. To re-screen; bean seeds were planted in buckets laid out in a screen house in a complete random design (CRD) with three replicates. Monosporic cultures of *P. griseola* were grown on V8 agar medium for 14 days in an incubator at 24^{o}C. Inoculum was prepared by adding sterile water on the surface of culture plates and scrapping the culture surface using a sterile glass rod. Three-week-old plants were spray-inoculated with a pathogen concentration of $2x10^4$ conidia ml^{-1} until runoff using hand sprayer. The moistened plants were placed in a humid chamber at 22-28^{o}C with relative humidity of 95% for 4 days. Disease symptoms were scored 6, 12, 18 and 21 days after inoculation, using the CIAT 1–9 visual scale (Schoonhoven and Pastor-Corrales 1987), where 1= plants with no visible disease symptoms; 3 = presence of a few small nonsporulating lesions; 5 = plants with several small lesions with limited sporulation; 7 = plants with abundant and generally large sporulating lesions, and 9 = large sporulating and often coalescing lesions, and premature defoliation. Disease severity data were analysed using general ANOVA in GENSTAT (Payne *et al*., 2007).

Conversely, screening work on the same landraces using eight more ALS isolates is on-going. On the other hand, three bean genotypes AND 277, Mexico 54 and G5686 comprising of *Phg 1*, *Phg 2* and Phg_{G5686A} resistance genes were selected for pyramiding respectively. The genotypes represented varying levels and modes of resistance. Hybridisation was based on cascading pedigree pyramiding scheme (Bertrand *et al.*, 2004). Parental crosses to were made to generate F_1. These are being advanced to F_2 for progeny resistance evaluation. Disease severity data will be analysed according to Payne *et al*. 2007. Molecular markers TGA1.1, OPEO4, Pv-ag004, Pv-at007 and Pv-ctt001 will be used to identify F2 progenies with the three gene combinations.

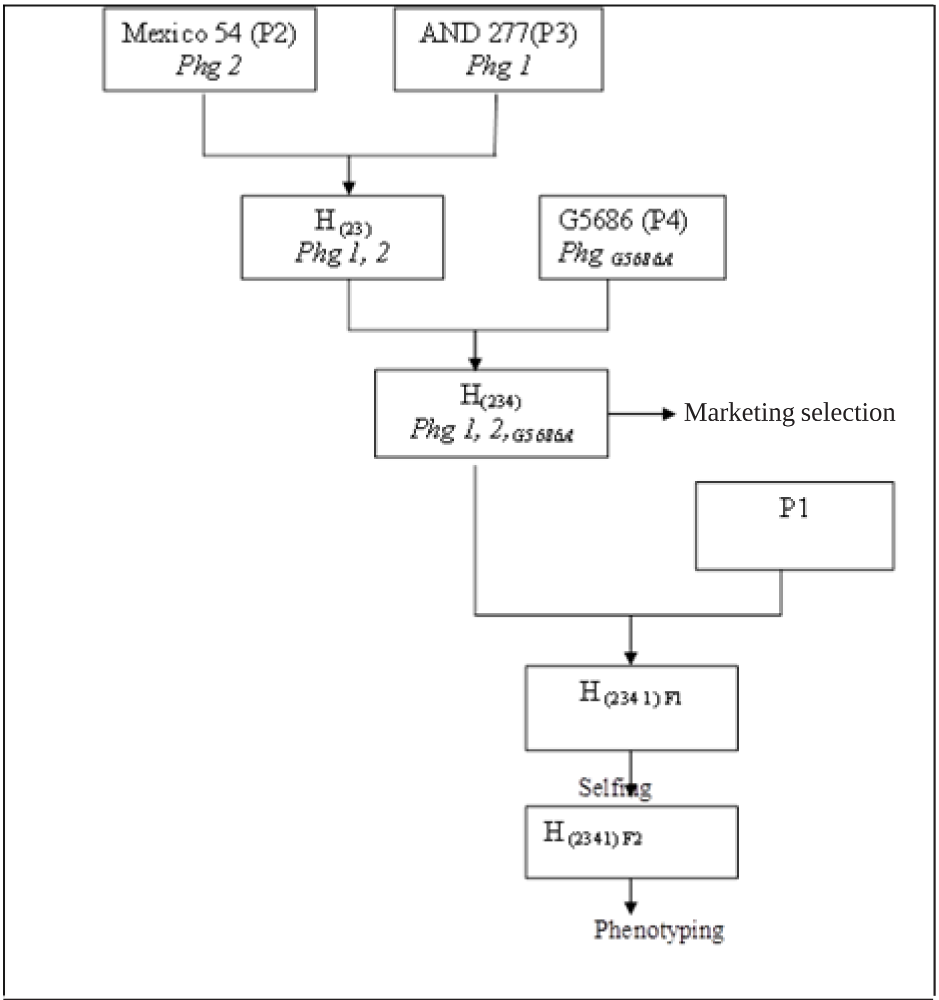

Figure 1. Pyramiding scheme used for crossing. Parents Mexico 54, AND 277 and G5686 are the donors for angular leaf spot resistance with resistant genes *Phg 2,1* and *G5686A* respectively, and P1 are the susceptible lines to be improved.

Research Application

Preliminary results indicated that for both isolates Mesoamerica (1:6) and Andean (21:7), susceptible and resistant checks had the highest and lowest ALS severity score, respectively. Fifty four percent of landraces were resistant and 11% susceptible to isolate 1:6. The high percentage of resistant landraces was possibly attributed to low virulence level of isolate 1:6. However with isolate 21:7 most landraces were moderately resistant (59%) and landraces U0074, U0053, U0083 were as equally resistant to isolate 21:7 as the two resistant checks used in the study. Nonetheless, accessions U0053 and U0074 were resistant to both isolates 1:6 and 21:7. This offers opportunity for the two landraces to be used as sources of ALS resistance to expedite the introgression of resistance into other bean germplasm.

Recommendation

Although landraces U0053 and U0074 were resistant to both races under screen house conditions but further screening with more virulent isolates and investigation under field conditions is required. Gene pyramiding is most likely to generate information on the effectiveness of gene combinations in controlling ALS which is valuable to breeders but pyramided lines need to be further tested and released as varieties by breeding programmes.

Acknowledgement

This work was supported by the Regional Universities Forum for Capacity Building in Agriculture (RUFORUM) and International Centre for Tropical Agriculture (CIAT).

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