Research Application Summary

Identification of QTL's linked for resistance to maize cob rot caused by *Fusarium graminearum* in tropical maize

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Abstract

Maize cob rots caused by *Fusarium graminearum* cause yield losses and quality as a result of produced mycotoxins. Developing varieties resistant to cob rots is an alternative strategy that is practical and provides better insurance for the small scale farmers. The subjectivity of scoring and the varying virulence responses of these pathogens to environmental conditions make selection for resistance difficult. The objectives of this study were to i) identify quantitative trait loci (QTL's) associated with resistance to *F. graminearum* and ii) evaluate the possibilities of utilising these QTL's for marker assisted selection (MAS). A major QTL was identified on chromosome 5. The additive and dominance effects ranged from -0.11 to -0.30 and all detected QTL's were more than 5 cM from the nearest molecular marker utilised in the study. Therefore, there is need to utilise the maize genomic map to identify and test several markers (< 5 cM, to the detected QTL's), in order to locate more reliable molecular markers for utilization in MAS.

Key words: Cob rot, *Fusarium graminearum*, maize, quantitative trait loci

Résumé

La pourriture d'épi de maïs causée par *Fusarium graminearum* provoque la perte de rendement et de la qualité en raison de mycotoxines produites. Le developpement des variétés résistantes à la pourriture des épis est une stratégie alternative qui est pratique et offre une meilleure assurance pour les petits agriculteurs. La subjectivité de la notation et les réponses variant de virulence de ces pathogènes aux conditions environnementales rendent difficile la sélection pour la résistance. Les objectifs de cette étude étaient : i) identifier les caractères quantitatifs de locus (QTL) associées à la résistance au *F. graminearum,* ii) évaluer les possibilités d'utilisation de ces QTL pour la sélection assistée par marqueurs (SAM). Un QTL majeur a été identifié sur le chromosome 5. Les effets additifs et dominants variaient de -0,11 à -0,30 et tous les QTL détectés ont plus de 5 cM de plus à partir du marqueur

moléculaire le plus proches qui sont utilisés dans l'étude. Par conséquent, il y a un besoin d'utiliser la carte génomique du maïs afin d'identifier et de tester plusieurs marqueurs (<5 cM, pour les QTL détectés), afin de localiser les marqueurs moléculaires les plus fiables pour l'utilisation dans les MAS.

Mots clés: épi de pourriture, *Fusarium graminearum*, le maïs, les traits quantitatifs de lotus

Background

Maize cob rots caused by *Fusarium graminearum* reduce yield and lower grain quality as a result of produced mycotoxins (Bigirwa *et al.*, 2007). Generally, with control cob rots, a combination of crop sanitation, good agronomic practices and timely harvesting has been used, but with limited success (Munkvold, 2003). Spraying with fungicide can be employed, but unfortunately, this is expensive, and in most cases, not feasible for resource-poor farmers. Developing varieties resistant to cob rots is an alternative strategy that is practical and provides better insurance for the small scale farmers.

However, the subjectivity of scoring and the varying virulence responses of the pathogens to environmental conditions make selection for resistance difficult (Bertland *et al.*, 2007). Molecular markers linked to resistant genes or QTL's can be used for MAS to enhance the accuracy of selecting the desired genotypes. In this study, simple sequence repeat (SSR) molecular markers, targeted to specific regions of the seven maize chromosomes identified by Wisser *et al.* (2006) as linked with QTL for cob and stalk rots multiple pathogens were used. The objectives of this study were to identify QTL's associated with resistance to *F. graminearum* and to analyse the possibilities of utilising the QTL's for marker assisted selection.

Literature Summary

Marker-assisted selection (MAS) is particularly effective when selecting for quantitative traits (Bertland *et al.*, 2007). It is independent of the environmental influence and may save time, resources and effort (Bertland *et al.*, 2007). In addition, molecular markers enable researchers to dissect quantitative traits into their single genetic components, allowing pyramiding of the beneficial QTL alleles (Ribaut and Raggot, 2007). Little is known about QTL molecular mapping to cob rots for *Fusarium graminearum* in tropical maize, as most research work has been reported on temperate maize. Temperate maize germplasm has a narrower genetic base (Sibov *et al.*, 2003), implying that different information can probably be obtained from mapping

with tropical maize. Working on temperate maize, Ali *et al.* (2005) detected 18 QTL's associated with resistance to *F. graminearum* and these individually accounted for 7 % to 35 % of the phenotypic evaluation. Of the 18, only one QTL (on the interval, umc0282-umc1155) on chromosome 5 was identified consistently in more than one environment.

Study Description

The mapping population consisted of 123 F_2 plants produced from advancing the cross between WL 118-10 and CZL-8 lines, resistant and susceptible respectively to *F. graminearum*. DNA for genotyping was obtained from young leaf samples two to three weeks after planting and was extracted from the powdered leaf material using the cetyltrimethylammonium bromide (CTAB) method (Hoisington *et al.,* 1994). Fourteen SSR markers which were found to be polymorphic between the two parents were used to genotype the F_2 population and the linkage map was constructed using Genstat 14 (Payne *et al.,* 2011). The phenotypic evaluation was carried out in three sites in Uganda: Burindi (1° 25¹ N, 31° 21¹ E; altitude 1140 m), Masaka (0° 20¹ S, 31° 44¹ E, altitude 1315 m) and Namulonge (0° 32¹ N, 32° 35¹ E; altitude 1150 m). The evaluation was done on 123 $F_{2:3}$ maize families.

To identify QTL's both the genotypic and phenotypic data were uploaded in the QTL cartographer software version 2.5. The analysis was then performed using composite interval mapping (CIM) which was set at the LOD score threshold of 2.5, with the walking distance and permutation time of one cM and 500, respectively. The degree of dominance for the detected QTL's was computed as d/[a], where d is the dominance gene effect and [a] is the absolute value of the additive gene effect of the putative QTL.

Research Application

Composite interval mapping identified a total of three QTL's across locations (Table 1).

The additive and dominance gene effects of associated QTL's ranged from -0.11 to -0.30. The finding on negative additive effects implies that that the substitution effect of a non favourable allele (susceptible) with a favourable allele (resistant), reduced disease severity at that locus. A major QTL (Fus_5) was identified at a distance of 39 cM from marker umc1018 and this explained 30% of the phenotypic variation.

Table 1. Identified QTL's across three locations evaluated from F_2 population associated with resistance to *F. graminearum*, derived from a cross of WL 118-10 and CZL-8.

Marker interval	*F. graminearum*						
	QTL[a]	pos. cM[b]	LOD[c]	r^{2d}	a[e]	d[f]	d/[a][g]
umc2082 - umc2280	Fus_4,1	22	3.9	20	-0.25	-0.15	-0.60
umc2280 - umc1142	Fus_4,2	18	3.7	22	-0.25	-0.11	-0.44
umc1019 - phi048	Fus_5	39	3.9	30	-0.14	-0.30	-2.14

a – QTL have been named from the trait abbreviation, followed by the chromosome number where detected. Where more than one QTL was detected on a chromosome for a particular trait, the second number is added to show the order and the closest to zero gets position 1. Example Fus_4,2- means the QTL associated with resistance to *F. graminearum* was detected on chromosome 4 and it is in the second position (in terms of distance from 0 on linkage map) to another detected QTL on the same chromosome.
b- The positions of the QTL are measured from the distance of the marker listed first for that interval.
c- Logarithm of odds likelihood equivalent to $-Log_{10}$ likelihood
d- Amount of phenotypic variance in percentage explained by the detected QTL, according to interval mapping.
e- Additive gene effect of detected QTL. f- Dominance gene effect of detected QTL. g- Degree of dominance

While this study provides useful information on approximate position and effects of putative QTL's, associated with resistance to *F. graminearum*, the linked markers are not close enough to the identified QTL to be used effectively for MAS. Therefore, the next step should be to use the maize genomic map to identify several markers in the vicinity of the detected QTL's, in order to locate nearby markers (< 5cM). In addition, sequence-based searches in the vicinity of the QTL could be an effective starting point for identifying the specific gene (s) responsible for the QTL.

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