Research Application Summary

Characterization of root development in a segregating diploid banana population as a searching basis for resistance to *Radopholus similis*

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Abstract

Plant parasitic nematodes are recognised as the most serious pests of bananas wherever they are grown. Nematodes feed, multiply and migrate inside banana roots causing necrotic and reduced root systems. Genetic resistance to nematodes provides a more sustainable management option to poor-resource farmers. Traditionally, identifying resistance/tolerance of banana varieties to nematodes depends on evaluating nematode reproduction and (histo)pathological investigations. New evidence indicates that there is an effect of root growth characteristics on resistance observations. Number and root system size seem to be the critical factors in banana tolerance to nematodes; plants with higher root numbers and/or vigour are less debilitated by nematodes. The proportion of primary, secondary and tertiary roots in bananas depends on the variety (genome group and ploidy level).

Key words: Banana, nematode resistance, QTL mapping

Résumé

Les nématodes, parasites des plantes, sont reconnus comme des parasites les plus dangereux de bananes bananes là où ils sont cultivés. Les nématodes se nourrissent, se multiplient et migrent à l'intérieur des racines des bananes, causant des nécrotiques et la réduction des systèmes racinaires. La résistance génétique aux nématodes fournit une option de gestion plus durable aux agriculteurs de maigres ressources. Traditionnellement, l'identification de la résistance et la tolérance des variétés de la banane aux nématodes dépendent de l'évaluation et de la reproduction des nématodes (histo) des enquêtes pathologiques. De nouvelles preuves indiquent qu'il existe l'effet de la croissance des racines sur les observations de la résistance. Nombre et la taille du système racine semblent être les facteurs essentiels de la tolérance aux nématodes de la banane, les plantes avec un nombre plus élevé des racines et / ou la vigueur sont moins affaiblis par les nématodes. La proportion de racines primaires, secondaires et tertiaires dans les bananes dépend de la variété (groupe de génome et le niveau de ploïdie).

Mots clés: Banane, la résistance aux nématodes, la cartographie de QTL

Background

Banana achieves its greatest importance as a food crop in the east African highlands region comprising Uganda, Rwanda, Burundi, Democratic Republic of Congo and parts of Kenya and Tanzania. Plant-parasitic nematodes are recognized as serious pests of bananas especially the burrowing nematode *Radopholus similis.* Plant resistance offers the only sustainable nematodes management approach in smallholder banana production systems, but sources of resistance to plant –parasitic nematodes in bananas are rare and the mechanisms of resistance not well known.

There are observed differences in susceptibility of banana cultivars grown in east African highlands to nematodes but the reasons for these differences are poorly understood. Banana cultivars also differ in root systems. However, it is not known whether the differences in the banana root systems affects nematode infection and damage or root system improvement can be a feasible banana breeding goal.

This study assessed differences in root development among 51 genotypes from a segregating diploid banana population and related these differences to their reaction to nematode infection and damage, with the overall aim of developing molecular markers that can be used to map the quantitative trait loci in the banana genotypes manifesting resistance/tolerance to nematodes.

Literature Summary

The search for natural sources of resistance and tolerance to *R. similis* has been carried out all over the world (Dochez *et al*., 2006, 2009; Fogain & Gowen, 1998; Pinochet & Rowe, 1979) and differences in susceptibility among banana genotypes to nematode infection and damge are documented (Speijer *et al*., 2000). However, the underlying mechanisms of host plant resistance response to nematodes penetration, development and reproduction has not been thoroughly investigated especially for migratory endoparasitic nematode such as *R. similis*. Differences in banana root characteristics have been observed in the field and hydroponic cultures (Swennen *et al*., 1988;

Blomme, 2000; Stoffellen, 2000; Rufikiri, 2000; Talwana *et al*., 2006) and it is possible that these differences, especially root numbers and root size, may play a role in banana tolerance to nematode infection and damage.

Study Description

This study was conducted from May 2009 to June 2010 at the International Institute of Tropical Agriculture (IITA), Namulonge in Uganda. The diploid banana population used in the study was derived by crossing the diploid hybrids TmB2x 6142-1 female susceptible and TmB2x 8075-7 male resistant. In order to produce uniform small plants suitable for Rhizotron analysis, the macropropagation method was used. Before planting, all roots were cleaned and cut up 10cm from the corm and planted in boxes (rhizotrons). The substrate in boxes was a sand-soil-manure mixture in a 3:1:1 ratio and was steam-sterilized before use. The experiment was laid in RCBD design and had 53 genotypes including Yangambi Km-5 and Valery as resistant and susceptible checks, respectively in three replications each during two seasons.

Root growth observations were made at weekly up to the 6th week after planting begining with the 2nd week. Root architecture, length, width, height and thickness were manually traced on transparent sheets, scanned and analysed using WinRHIZO Pro (2009a, b version) software. The 53 genotypes were inoculated with *R. similis* 7 month after planting. At 8 weeks after inoculation, two roots per genotype were sampled for scoring agronomic traits and root damage. DNA extraction and development of a molecular map were done based on SSR markers using a modified CTAB procedure. All data collected were analyzed using SAS package version 9.2 (SAS Institute, 2010). Means were separated using Least Significant Difference (LSD) at 5% level of significance.

Research Results

Based on combined data for two seasons there was a significant genotype effect on diameter, (1.07 to 1.80mm), roots length (405 to 1108cm), roots projected area (38 to 186cm2) and roots surface area (120 to 585cm2) the effect was also significant for nalyzed region area (403 to 1792cm2), analyzed region height (18 to 39cm) and tips and forks. Meaning that root as much grow, parameters increase with variability according to genetic behavior. As individual data measurement, small diameter showed high percent of root measurement (Tips 95%, Length 73%, Surface Area and Projected Area 42% each) except root volume (17%), while medium diameter has showed a relative measurement (Root Volume 33%, Surface Area and Projected Area 32% each, Length 19% and Tips 3%). High amount of root measurement was only observed on Root Volume large diameter 50%. Meaning that, fine and young roots are generally considered to have a higher activity uptake than old roots for water and nutrients from the soil also synthesis of plant growth hormones.

Indicators of plant growth and development (height, girth, standing leaves, number of healthy roots, number of total roots, plant weight, root weight showed a very high significantly different (P<0.001) among each other genotypes 14 weeks after planting which is very important for selection of agronomics traits in the breeding purpose and resistant varieties partitioned a good proportion of biomass than less resistant. Root damages parameters (dead roots, feeder roots, root knot and roots necrosis) were found with slightly necrotic lesions somehow visible on root inoculated at the harvest date. Nematodes counts (nematodes final population and reproduction ratio) showed a very high significantly different (P<.0001). However, probably the penetration of inoculum to the genotypes was not perfectly effective because for the same genotypes screened, nematodes were found in 1 or 2 replications while the remaining was unharmed. Or, some genotypes escaped to nematodes inoculation. However, our results showed a high significantly different correlation between nematodes final population and total root necrosis (R=0.6077, P<.0001), meaning that as much a number of nematodes are high the percentage of total necrosis become higher. The identification of host response to *R. similis* based on a comparison with a susceptible Valery and a resistant Yangambi km-5 eight weeks after individual root inoculation with 50 *R.similis* females as initial population, has classified genotypes according to the nematodes final population 23 resistant (R), 18 susceptible (S) and 12 partial resistant (P). Linkage of SSR markers to the phenotypic traits of 53 genotypes using mapping tools (MapManager QTL) is going on and roots phenotype traits with resistance to *R.similis* will correlate.

This study on nematode reaction is very important for selection of agronomics traits in the breeding purpose. Results showed that root traits of *Musa* horizontal radical type were influenced by the genotypic behavior and growth stages. In generally it has shown that, under optimum growing in Rhizotrons conditions, measurement of the root system can be determined. It is therefore recommended that users of image analysis systems determine this maximum length for 1st flush root (3 to 8weeks) and the 2nd (8weeks to 5months) for accurate determinations of total root length. Well adjustment of diameter intervals and increasing the scanner resolution are required to reduce this underestimation.

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