SUMMARY

Fusarium oxysporum f. sp. *cubense* (Foc), a soil-borne fungus affecting bananas (*Musa spp*.), is considered one of the most devastating pathogens in agricultural history. The fungus infects banana roots, colonises the rhizome and pseudo stem, and causes a lethal wilting disease called Fusarium wilt. Fusarium wilt can cause losses of up to 100% in banana fields planted with susceptible genotypes, without any known cure. Host plant resistance to Foc, which has been identified in the *Musa* gene pool, is widely considered the only feasible method to control the disease. However, conventional breeding to improve susceptible banana varieties is hampered by male and female sterility and the long generation period of the crop.

The inheritance of resistance in *Musa* to Foc race 1 in the 'SN8075F2' population, derived from the cross of cultivar 'Sukali Ndiizi' and the diploid banana 'TMB2X8075-7', was investigated in this study. One hundred and sixty three F2 progenies were evaluated for their response to Fusarium wilt in a screen house experiment. The test plants were inoculated by mixing loam soil with millet grains, colonized by Foc race 1, in polythene pots. One hundred and fifteen genotypes were categorized as susceptible and 48 as resistant based on rhizome discolouration. Mendelian segregation analysis for susceptible vs. resistant fitted the segregation ratio of 3:1 (X2 =1.72, P=0.81), suggesting that resistance to Fusarium wilt in the diploid line 'TMB2X8075-7' is provided by a single recessive gene. The name *pd1* (Panama disease 1) has been proposed for the recessive gene responsible for resistance to Fusarium wilt in the diploid line 'TMB2X8075-7'.

DArT markers were identified in a segregating population following a cross between the susceptible banana cultivar 'Sukali Ndiizi' and a resistant diploid banana 'TMB2X8075-7'. The markers were in qualitative linkage disequilibrium, with 13 markers linked to resistance and 88 markers associated with susceptibility to Foc race 1. Putative functions have been assigned to candidate genes through *in-silico* database analysis including Laccase-25 (LAC25), Homeobox-leucine zipper protein (HOX32), SWIM zinc finger family protein, Transcription factor MYB3, GDSL esterase/lipase EXL3 among others. The candidate markers and genes closely associated with resistance/susceptibility could also be used in genetic engineering or for marker-assisted selection (MAS) in breeding for Fusarium wilt resistance.

The Foc race 1-banana binomial interaction of three genotypes ('Sukali Ndiizi' AAB, 'Mbwazirume' AAA and 'TMB2X8075-7 AA) was investigated by deep sequencing of the root transcriptome to study Fusarium wilt resistance in bananas. A total of 299 million raw reads, each about 100-nucleotides long, were derived from cDNA libraries constructed at four time IV points: 0, 48, 96 and 192 hrs after inoculation with Foc race1. From the 10136 differentially expressed genes (DEGs), 5640 (55.7%) were uniquely up-regulated and 4496 (44.4%) uniquely down-regulated in the libraries of 'Mbwazirume', 'TMB2X28075-7' and 'Sukali Ndiizi at 48, 96 and 192 hrs post inoculation. The DEGs were annotated with Gene Ontology (GO) terms and

pathway enrichment analysis, and significant pathway categories identified included the ‘Metabolic’, ‘Ribosome’, ‘Plant–pathogen interaction’ and ‘Plant hormone signal transduction’ pathways. Salicylic acid and ethylene were stimulated in the ‘Plant hormone signal transduction’ pathways in all the three genotypes. Fifteen defence-related genes were identified as candidate genes contributing to Fusarium wilt resistance in banana. These candidate genes could be used to improve susceptible banana genotypes to enhance levels of fungal disease resistance to Foc race 1.

OPSOMMING

Fusarium oxysporum f. sp. *cubense*, ’n grondgedraagde swam wat piesangs (*Musa spp*.) affekteer, word beskou as een van die mees vernietigende siektes in die geskiedenis van landbou. Die swam infekteer piesangwortels, koloniseer die rhizoom en pseudostam, en veroorsaak ‘n dodelike verwelksiekte, genoemd Fusarium verwelk. Fusarium verwelksiekte kan verliese van tot 100% veroorsaak in plantasies wat met vatbare genotipes geplant is, sonder enige kuur. Gasheerplantweerstand teen Foc, wat in die *Musa* genepoel beskikbaar, word lank reeds beskou as die enigste haalbare manier om die siekte te beheer. Maar konvensionele teling word belemmer deur manlike en vroulike onvrugbaarheid en die lang generasie tydperk van die gewas.

Die erfenis van weerstand in *Musa* teenoorFoc ras 1 in 'SN8075F2, 'n afstammeling van die kruis tussen kultivar 'Sukali Ndiizi' en die diploïede piesang 'TMB2X8075-7’ word in hierdie studie ondersoek. Een honderd en sestig F2nasate is vir hul reaksie op Fusarium verwelking in 'n glashuis eksperiment geëvalueer. Die plante is geïnokuleer deur leemgrond te meng met millet saadwat deur Foc ras 1 gekoloniseer is, en in plastiek potte geplant is. Een honderd en vyftien (115) genotipes was vatbaar, en 48 bestand ten opsigte van die verkleuring van hul rhizoom. Mendeliese segregasie analise vir vatbaar teen bestand pas die segregasie verhouding van 3: 1 (X2 = 1,72, P = 0,81), wat daarop dui dat die weerstand teen Fusarium verwelking in diploïede lyn 'TMB2X8075-7' deur 'n enkele resessiewe geen bepaal word. Die naam *pd1* (Panama siekte 1) is voorgestel vir die resessiewe geen wat weerstand teen Fusarium verwelking in die diploïede lyn 'TMB2X8075-7 verskaf.

DArT merkers is geïdentifiseer in ‘n segregerende populasie na ‘n kruis tussen ‘Sukali Ndiizi’ en ‘n weerstandige diploid piesang ‘TMB2X8075-7’. Die merkers was onewewigtig in kwalitatiewe koppeling, met 13 merkers wat gekoppel was aan weerstand en 88 merkers aan vatbaarheid vir Foc ras 1. Funksies aan kandidaatgene toegeken deur in-silico databasis analise sluit in Laccase-25 (LAC25), Homeobox-leucine zipper proteïen (HOX32), SWIM zinc ‘finger family protein’, ‘Transcription factor MYB3’, GDSL esterase/lipase EXL3. Hierdie kandidaat merkers en gene wat nou verband hou met weerstand/vatbaarheid kan ook in die

genetiese modifikasie van piesangs, of vir merker-geassosieerde seleksie (MAS) vir die teling vir Fusarium verwelking weerstand gebruik word.

Die Foc ras 1-piesang binomiaal interaksie van drie genotipes ('Sukali Ndiizi' AAB, 'Mbwazirume' AAA en 'TMB2X8075-7 AA) was ondersoek deur analise van hul wortel transkriptoom. ‘n Totaal van 299 miljoen basispare, wat elkeen bestaan uit sowat 100 basispare, is bepaal tydens vier tydspunte: 0, 48, 96 en 192 ure na inokulasie. Van die V I 10136 gene differensieel uitgedrukte gene (DEGs) was 5640 (55,7%) uniek uitgedruk en 4496 (44,4%) uniek onderdruk in 'Mbwazirume', 'TMB2X28075-7’ en ‘Sukali Ndiizi’ teen 48, 96 en 192 uur na inokulasie. Die DEGs is met Gene Ontologie (GO) terme en pad verryking analise geannoteer. Die beduidende geenkategorieë wat geïdentifiseer is het die volgende ingesluit: 'Metaboliese', 'Ribosoom’, ‘Plant-patogeen interaksie’ en ’Plant hormoon seintransduksie'. Salisiensuur en etileen is gestimuleer in die 'Plant hormoon seintransduksie' bane in al die drie genotipes. Vyftien verdediging-verwante gene is geïdentifiseer as kandidate wat bydra tot weerstand teen Fusarium verwelking in piesangs. Hierdie kandidaatgene kan gebruik word om vatbaar genotipes te verbeter vir verhoogde weerstand teen Foc ras 1.