Identification and characterization of *Pythium* spp. at CIAT-Kawanda, Uganda

Introduction

***P**ythium* spp, cause severe bean root rots under cool and humid conditions. Poor soils and intensive cultivation compound the epidemics.

There are more than 120 species reported globally some of which are very pathogenic meanwhile others are saprophytic.

Yield losses due to *Pythium* outbreaks can be as high as 100% on susceptible varieties in the absence of control measures.

Bean root rot caused by *Pythium* is the most devastating in Eastern & Central Africa where the intensity of bean production is high.

Pathogen isolation

Uproot Wash, blot dry

CMA amended with antibiotics, incubate at 24ºC for 12 hrs

PDA plate

Preservation: PDA agar slants at 4ºC

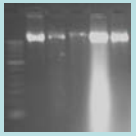
V8 juice

TWA

DNA extraction and PCR

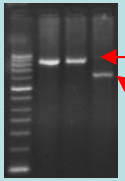
DNA is isolated from a mat of mycelia growing on V8 juice.

Maceration and DNA extraction



DNA quality and quantity

Amplification using *Pythium* specific primers



Pythium (700bp)

Others

Electrophoresis

Species identification

- PCR products of *Pythium* isolates are shipped for automated sequencing abroad.
- Chromatograms are verified and edited for correct peak scoring.
- Bioinformatics applications are used to identify the species through BLAST searches.

Pathology and screening tests

Newly identified *Pythium* species are:

1. Tested for pathogenicity using a resistant (RWR 719) and a susceptible (CAL96) varieties.
2. Used to screen Marker Assisted Selection lines to ascertain root rot resistance traits.

Procedure:

PDA, 4 days 24ºC

Sterilized finger millet/sorghum, 14 days RT

Evaluation: Uproot wash and score 21 days after planting

1-9 Scale

1 3 5 7 9

Mix 1inoculum : 8 sterilized soil and plant

Watering thrice a day in the 3rd week

This procedure is also applicable to *Fusarium spp.*