

**NEW DISEASE REPORT**

# First report of *Penicillium sclerotigenum* causing post-harvest rot of yam in Côte d'Ivoire

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Yam (*Dioscorea* spp.) is the third most produced non-cereal crop with global production of 72 million tonnes per year and is cultivated across West Africa. With a production of over 7 million tonnes, Côte d'Ivoire is the world's third largest producer and yam is the country's main non-cereal food crop (Scott *et al.*, 2020). However, the storage life of yams is greatly reduced by post-harvest rots and previous studies revealed that a range of fungi were responsible, including *Aspergillus Botryodiplodia*, *Colletotrichum*, *Mucor* and *Penicillium* species (Assiri Kouamé *et al.*, 2017).

In May and September 2021, surveys were done in two cities central to yam production and marketing in Côte d'Ivoire: Abidjan (5° 19' 0.001" N 4° 1' 59.999" W) and Bondoukou (8° 2' 27.074" N 2° 48' 5.836" W). In total 60 tubers, 30 of each variety (*Dioscorea cayennensis* subsp. *rotundata* and *D. alata*), exhibiting symptoms of dry brown rot were collected. Symptoms were characterised by slight necrosis on the pericarp of the tuber, with some tubers showing a greenish discolouration around the necrosis with a liquid that drained and gave off a foul odour (Fig. 1).

Yams displaying symptoms of brown dry rot were bisected before internal sections of diseased tissue were excised, surface sterilised, and plated onto potato dextrose agar (PDA) medium. Surface sterilisa-

tion was performed through immersion in 70% ethanol for one minute, 1% sodium hypochlorite for two minutes and rinsed in distilled water. In total 21 morphologically similar fungal isolates were obtained and purified by subculturing and single spore cultures. Morphological characteristics of a representative isolate (DAB\_PK) were assessed and confirmed to be consistent with *Penicillium* spp. The fungus produced rapid mycelial growth with a mean growth rate of 1.79 cm per day. The mycelia were green, observed from above and below, with a short and powdery mycelium often showing striations on the face (Fig. 2a). The mycelia were septate with branched conidiophores (225 to 410  $\mu$ m) with metulae (8 to 11  $\mu$ m) and phialides and produced abundant rounded, unicellular conidia, 2 to 5  $\mu$ m in size (Fig. 2b).

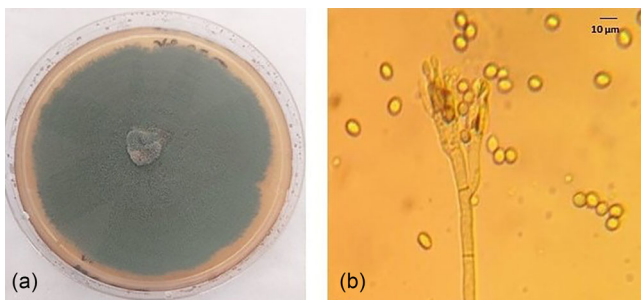
Three fungal isolates (DAB\_PK and TAN\_AS from *D. cayennensis* subsp. *rotundata*; KAT\_3 from *D. alata*) were selected for molecular identification. Genomic DNA was isolated from freeze-dried fungal mycelium as described previously (Harrison & Thompson, 2020). The ITS1-5.8S-ITS2 rDNA region was amplified by PCR using ITS1 and ITS4 primers. PCR amplicons were sequenced in both directions (Eurofins Genomics, Germany) and the sequences submitted to Genbank (Accession Nos. PP449079-PP449081 for DAB\_PK, TAN\_AS and KAT\_3, respectively). BLASTn searches showed that the sequences had

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**FIGURE 1** Yam tubers (*Dioscorea alata*) showing symptoms of dry brown rot, as observed in retail shops.

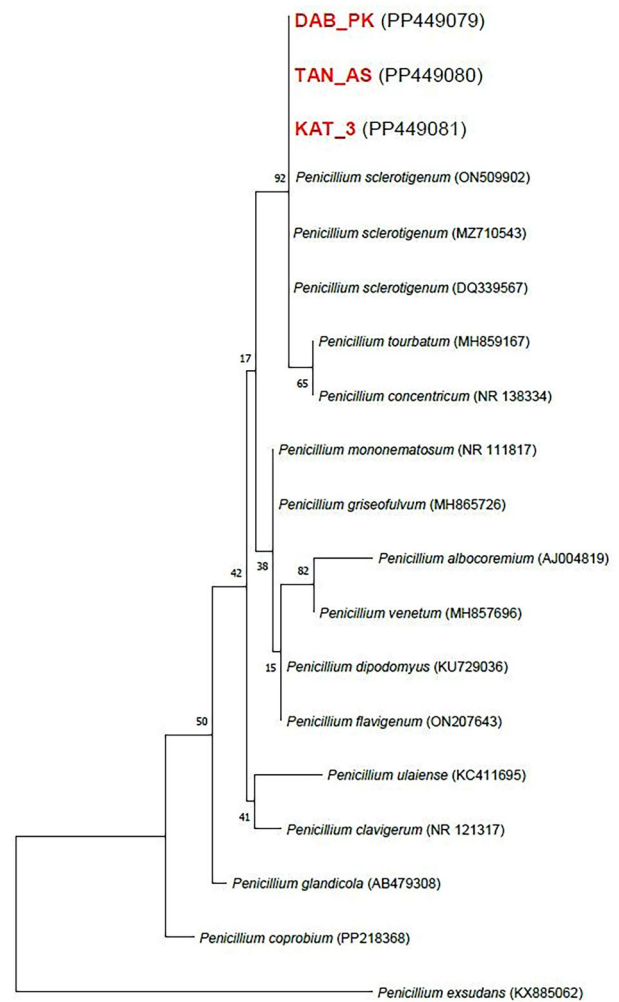


**FIGURE 2** Morphological characteristics of isolate DAB\_PK: a) cultural characteristics on potato dextrose agar after 5 days' incubation, and (b) spores and conidiophore.

>99% nucleotide identity with *P. sclerotigenum* strains NRRL 22813 (DQ339574) and YZU 21110550 (MZ710543).

Reference sequences representing key species within the *Penicillium* genus, as shown in Uy et al. (2023), were downloaded and aligned to sequenced amplicons. A maximum likelihood tree (1,000 bootstrap replicates) was constructed using a Tamura 3-parameter model, as determined by model-testing in MEGA X version 10.2.6 (Kumar et al., 2018). Phylogenetic analysis showed high bootstrap support delineating clades at the species level and placed all three sequenced isolates within the same clade as reference *P. sclerotigenum* sequences (Fig. 3).

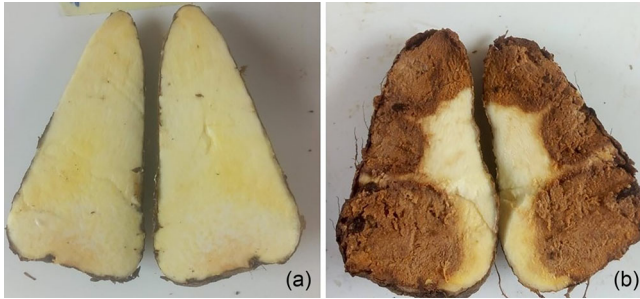
Koch's postulates were assessed using eleven isolates (NGK-B, KAT-PK1, T-PK, KM-PK, TAN\_AS, KGB-PK, DAB\_PK, BDK-AS, KAT-PK, TAN-PK and KAT\_3). Three yam tubers of each variety (*D. cayennensis* subsp. *rotundata* cv. Pkonan and *D. alata* cv. Bètè Bètè) were inoculated per isolate. The yam tubers were sterilised with 5% sodium hypochlorite, then rinsed thoroughly with distilled water and dried at room temperature for 30 minutes. Mycelial (0.5 cm diameter) plugs taken from PDA plates and placed into three holes of the same size in each



**FIGURE 3** Maximum likelihood phylogeny of the ITS region identifying isolated fungi (DAB\_PK, TAN\_AS and KAT\_3) as *Penicillium sclerotigenum*. Genbank accession numbers are displayed next to reference sequences. Node labels indicate support from 1,000 bootstrap replicates. Scale bar represents substitution sites.

tuber. The yams were wrapped with plastic film, placed in covered plastic containers and stored at room temperature ( $25 \pm 2^\circ\text{C}$ ) for 30 days. The inoculated isolates caused rots on yam tubers 13 days after inoculation. These symptoms were identical to rot symptoms on sampled yam tubers; control yams did not develop symptoms (Fig. 4). Fungi with the same morphology as the inoculated isolates were reisolated from the lesions, thus fulfilling Koch's postulates. The experiment was repeated using spore suspensions from seven-day old PDA cultures ( $1 \times 10^6$  spores per ml), rather than agar plugs. Thirty microlitres of inoculum were applied to each tuber and stored as described above. Symptoms appeared 15 days after inoculation and were identical to those observed in stored yams.

This is the first report of *P. sclerotigenum* as a causal agent of yam post-harvest disease in Côte d'Ivoire. *Penicillium sclerotigenum* was first described by Kim et al. (2008) as a pathogen responsible for yam tuber rot in south Korea. *Penicillium sclerotigenum* isolates were identified in both cultivated yam lineages, *D. alata* and *D. cayennensis*.



**FIGURE 4** Representative image of symptoms resulting from pathogenicity tests of *Penicillium sclerotigenum* isolate DAB\_PK on *Dioscorea cayennensis* subsp. *rotundata*: (a) Healthy tuber (control) and (b) inoculated tuber showing symptoms of rot.

*sis* subsp. *rotundata*. Côte d'Ivoire is the only country in West Africa where both lineages contribute equally to production (Kouakou *et al.*, 2023) and further monitoring is required to quantify losses and varietal resistance.

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