



The fungal population in oil palm plantation soil plays a diverse role that affect the palm tree productivity. Therefore, advance in fungal identification technique is crucial in order to investigate the importance of this microbial community. Fungal identification procedure is made easy by the introduction of MicroSeq® D2 LSU rDNA fungal identification sequencing system. This is an alternative molecular identification system for FIDS 1 and FIDS 3 that targets the ITS and 18S DNA regions. FIDS 2 is suitable for a large number of samples because of the rapid identification process compared to the FIDS 1 and FIDS 3. This identification system provides a precise tool to amplify and sequence the ribosomal RNA gene nuclear large subunit (LSU) expansion segment, D2 region rapidly (Sonnenberg *et al.*, 2007). Specifically, organisms can be identified until the species level based on the variation of DNA segments within the region (Sipiczki *et al.*, 2013). The identification procedure required a small quantity of a pure fungal culture for DNA amplification and sequencing. The

database provided by Applied Biosystems is based on D2 sequence and capable to identify up to more than 1000 validated fungal species. This includes filamentous fungi, yeast and oomycete.

OBJECTIVE

To precisely amplify and sequence the ribosomal RNA gene nuclear LSU expansion segment, D2 region for fungal species identification.

METHODOLOGY

The result and outline of fungal identification using Fungal Identification System 2 (FIDS 2) is presented in *Figures 1* and *2*. This sequencing kit uses Applied Biosystems BigDye® Terminator v1.1 chemistry. Dye-labelled 3'-dideoxynucleotide triphosphates (dye terminators) contained in the forward and reverse sequencing mixes are included in the sequencing kit. Four different fluorescent dyes were tagged to the dideoxy terminators (ddNTPs) with dye-terminator labelling. The extension product was terminated with the present

Project : Fungi Specimen : F S T Kit : FungalKit

Specimen	% Match	Sequence Entry	Library
F-Neg			No Libraries Searched Against
F-Pos	100.00	Saccharomyces cerevisiae (ATCC=18824)	AB_FungalLib_2011
	99.82	Saccharomyces cerevisiae (ATCC=9763)	AB_FungalLib_2011
	96.79	Saccharomyces bayanus (ATCC=76513)	AB_FungalLib_2011
	96.79	Saccharomyces pastorianus (ATCC=12752)	AB_FungalLib_2011
	95.00	Zygosaccharomyces microellipsoides (ATCC=10605)	AB_FungalLib_2011
SK8-11	99.86	Trichoderma fertile (DAOM=167162)	AB_FungalLib_2011
	99.86	Trichoderma harzianum (CBS=144.96)	AB_FungalLib_2011
	98.98	Glodadium vinde (DAOM=226717)	AB_FungalLib_2011
	98.61	Trichoderma aureoviride (DAOM=175924)	AB_FungalLib_2011
	98.61	Trichoderma harzianum (DAOM=199083)	AB_FungalLib_2011
TH	99.56	Trichoderma fertile (DAOM=167162)	AB_FungalLib_2011
	99.56	Trichoderma harzianum (CBS=144.96)	AB_FungalLib_2011
	98.86	Glodadium vinde (DAOM=226717)	AB_FungalLib_2011
	98.63	Trichoderma aureoviride (DAOM=175924)	AB_FungalLib_2011
	98.63	Trichoderma harzianum (DAOM=199083)	AB_FungalLib_2011

Figure 1. MicroSeq Sequence-Based Identification.



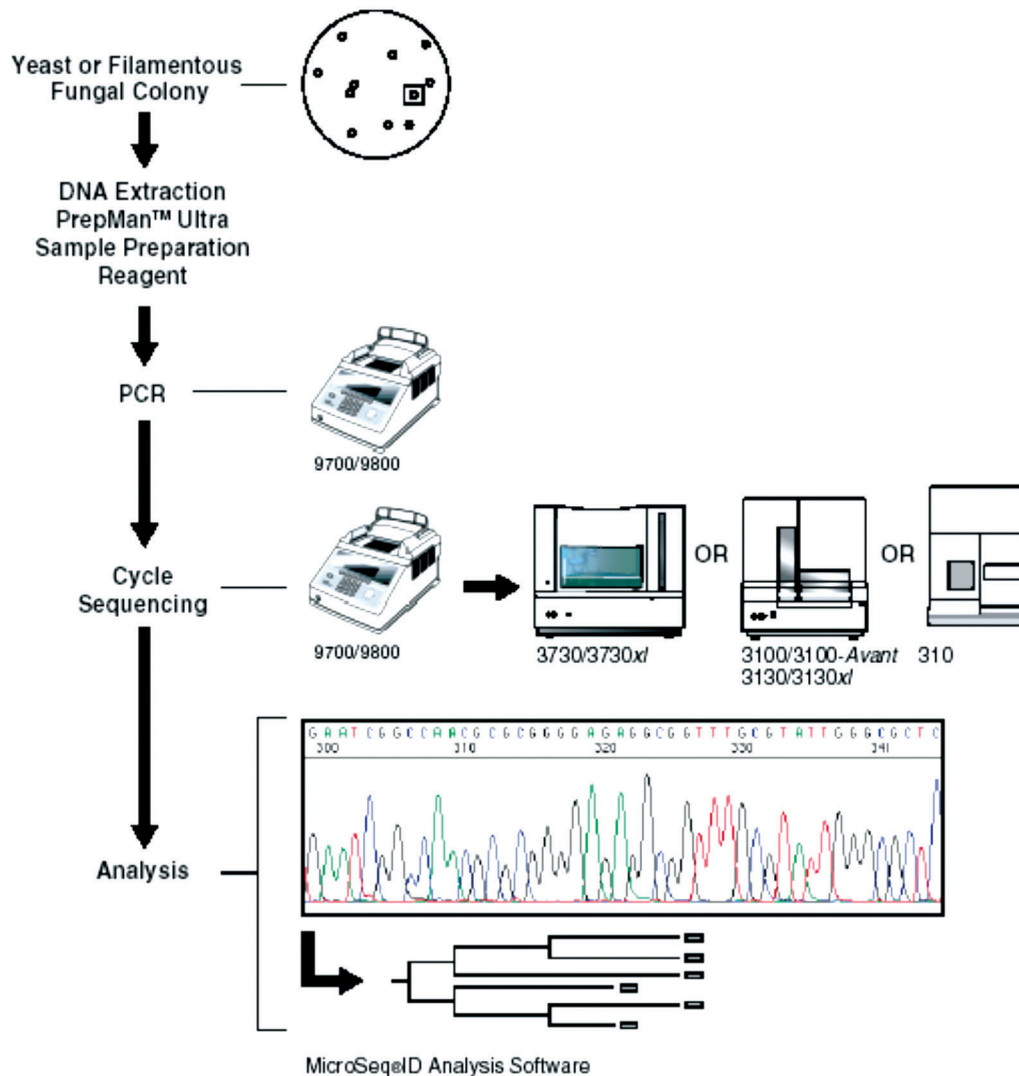


Figure 2. Fungal identification using Fungal Identification System 2 (FIDS 2).

dye labelled terminators in the reaction mix. The occurrence of the labelled product depended on the dye that was in contact with the base.

BENEFITS

FIDS 2 is a fast and reliable molecular fungal identification method using rapid sequencing analysis.

COST

The fee to identify a strain of fungi in 2014 is RM 2000 and is subject to change.

REFERENCES

SONNENBERG, R; NOLTE, A W; TAUTZ, D (2007). An evaluation of LSU rDNA D1-D2 sequences for their use in species identification. *Front. Zool.*, 4: 6.

SIPICZKI, M; PFLIEGLER, W P and HOLB, I J (2013). Metschnikowia species share a pool of diverse rRNA genes differing in regions that determine hairpin-loop structures and evolve by reticulation. *Plos One*: e67384.

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