Zootaxa 2427: 55–63 (2010)
www.mapress.com/zootaxa/

Article

ISSN 1175-5326 (print edition)
ZOOTAXA
ISSN 1175-5334 (online edition)

Linking operational clustered taxonomic units (OCTUs) from parallel ultra sequencing (PUS) to nematode species

DOROTA L PORAZINSKA[1,3], ROBIN M GIBLIN-DAVIS[1], WAY SUNG[2] & W KELLEY THOMAS[2]

[1]*Fort Lauderdale Research and Education Center, University of Florida, IFAS, 3205 College Avenue, Fort Lauderdale, FL 33314, USA. E-mail: dorotalp@ufl.edu, giblin@ufl.edu*

[2]*Hubbard Center for Genome Studies, University of New Hampshire, 35 Colovos Rd., Durham, NH 03824, USA. E-mail: way.sung@gmail.com, kelley.thomas@unh.edu*

[3]*Corresponding author. E-mail: dorotalp@ufl.edu*

Abstract

DNA-sequence-based approaches such as single nematode PCR and sequencing have been successfully used in nematode species diagnostics. Here, we use SSU rDNA sequences generated from an artificially-assembled community of nematodes of known identity and relative abundance to assess a relationship between nematode species and operationally clustered taxonomic units (OCTUs). OCTUs that formed at 95% similarity underestimated species richness by *ca* 30%, and all expected species were recovered only at 99% OCTU similarity. Although the number of OCTUs formed at 99% similarity was considerably higher than the actual number of nematode species, a pattern of the distribution of OCTUs within each species allowed an assignment of OCTUs to specific species. The pattern was highly predictable and helped to discern rather than obscure species recognition. The patterns and parameters that emerged from the control nematode dataset were then used to test our approach on environmental samples of nematodes obtained from a tropical rainforest in Costa Rica.

Key words:, biodiversity, DNA barcoding, high-throughput sequencing, metagenetic, nematode

Introduction

Current estimates of global nematode diversity are inferred predominantly from small-scale, disconnected studies conducted almost exclusively in temperate regions (Boag and Yeates 1998). Although predictions of species richness range from 100,000 to 1 million (Coomans 2000), only *ca* 27,000 species are known to science (Hugot *et al*. 2001) resulting in a well-acknowledged nematode identification gap (Blaxter 2003). As with other eukaryote microbiota (e.g. fungi, rotifers, and protozoa), high abundance, diversity, minute size, and obscure morphological characteristics present considerable difficulties for nematode species identification. Because microscopy-based nematode identification is extremely time consuming (Lawton *et al*. 1998), only a tiny fraction of randomly selected individuals is typically identified in any given sample. In practice, these taxonomic challenges are circumvented by identifying nematodes only to family or feeding-guild level. As a consequence, for most survey-scale studies no confident estimates of nematode species richness or species turnover are available or anticipated using traditional morphological assessment techniques only.

DNA-sequence-based approaches have been used to improve nematode species diagnostics over the last decade (Powers 2004). Several diagnostic DNA loci have been tested to distinguish nematode species (*i.e.*, nuclear ribosomal genes, the mitochondrial cytochrome oxidase 1 gene, and heat-shock protein genes) (e.g., Blaxter *et al*. 2005; Floyd *et al*. 2002; Powers *et al*. 1997; Skantar & Carta 2004, De Ley *et al*. 2005, Thomas *et al*. 1997, Ye *et al*. 2007). Traditional molecular approaches based on single-nematode PCR generate a consensus sequence for a chosen diagnostic locus that represents the sequence diversity of that locus within the individual. In the broad sense, this ignores heteroplasmy for mitochondrial sequences, heterozygosity for

Accepted by P. Mullin: 6 Mar. 2010; published: 15 Apr. 2010