

ESN Adana 2012 abstract submission

Preferred : Oral Presentation

Development of *Bursaphelenchus xylophilus*-specific microsatellite markers to assess the genetic diversity of populations from European forests.

Preferred session : Quarantine Nematology

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The pinewood nematode (PWN), *Bursaphelenchus xylophilus* (Steiner & Buhner, 1934), Nickle (Nematoda: Aphelenchoididae) is the causal agent of the pine wilt disease and is currently considered as one of the most important pests and pathogens in the world. Its introduction and spread in new forest ecosystems have considerable consequences both economically and environmentally. Therefore, it is of crucial importance to identify its invasion routes, to determine the origin of new outbreaks and to understand the invasion process of this species to prevent further dissemination of the disease in Europe. In order to address these questions using population genetic approaches, we have been developing a set of PWN-specific microsatellite markers, usable in routine conditions at the individual level, thanks to multiplex PCR coupled with a fast DNA extraction method. Microsatellites were isolated from a genomic library using a procedure combining DNA enrichment and high throughput pyrosequencing as recently described by Malausa et al. (2011). Primers were designed for 71 and 23 perfect and compound microsatellites, respectively, 26 of which were experimentally validated so far. Among them, 18 markers exhibited polymorphism after several rounds of amplification tests. Preliminary results on a set of 190 nematodes from 13 populations indicate a very low level of polymorphism in PWN populations from Portugal and Madeira Island, compared to populations from the native area in North America. The genotyping of a wide collection of samples from Europe, Asia and North America is currently underway in the laboratory. Assessing the genetic diversity of populations indeed constitutes the cornerstone to determine whether the European invasive PWN populations are the result of a single or several independent events of introduction.

Malausa T. et al., (2011) High-throughput microsatellite isolation through 454 GS-FLX Titanium pyrosequencing of enriched DNA libraries. *Molecular Ecology Resources* 11, 638-644.