

**P.S.I-5 Study of the genetic variability of Brazilian populations of *Cylindrocarpon* spp., causal agent of grapevine Black foot.**

A. RUSSI, R. NALIN, G. DEQUIGIOVANNI, R. GAVA, V. QUECINI, L.R. GARRIDO AND **P. RITSCHEL**\*

Molecular Biology Laboratory, Embrapa Grape and Wine. Zip Code 95700000, Bento Gonçalves, Brazil

\*Email: [patricia@cnpuv.embrapa.br](mailto:patricia@cnpuv.embrapa.br)

Two fungal species have been historically associated with grapevine Black foot, *C. destructans* and *C. macrodidymum*. Analyses of DNA sequences showed that *C. destructans* is represented by a complex of several species and, recently, grapevine isolates associated with black-foot have been reidentified as *C. liriodendri*. *C. destructans* was identified in Southern Brazil, but little is known about the fungus variability. The purpose of this work was to evaluate the genetic diversity of *Cylindrocarpon* in Southern Brazil. Twenty-four isolates were obtained from 12 localities and eight wine grapevine plants showing symptoms of black-foot. DNA was extracted and PCR amplified. The study was initiated with an investigative RAPD analysis to probe the variability of the fungus using 140 fragments produced by 32 arbitrary primers. Samples were then analyzed via ITS region amplification using primers ITS4 and ITS5, followed by digestion with seven restriction enzymes. NTSYS software package was used to estimate similarity between accessions as well as classification by the clustering algorithm UPGMA. Finally, selected ITS fragments were sequenced and compared to *C. destructans* and *C. liriodendri* sequences deposited at GenBank using ClustalX algorithm. Trees were obtained by TreeV32. RAPD dendrogram revealed a range of similarity (DICE coefficient) from 31% to 96%. Similarity based on ITS analysis varied from 16% to 100% and eleven patterns of enzyme digestion were observed. One representant of each pattern was sequenced. Sequences of Brazilian *Cylindrocarpon* clustered with *C. liriodendri* isolate CBS117526 collected on *V. vinifera* in Portugal.