Characterisation of different Chilean *Capsicum* spp. varieties and the antifungal P71 activity of their aqueous extracts

Sepúlveda M.¹, Cayún Y.², Gallardo V.¹, Barría E.², Nahuelcura J.², Costa J.³, Dias N.⁵, Ruíz A.², Cornejo P.⁶, Lima N.^{4,5}, Santos C.²

¹Doctorado en Ciencias de Recursos Naturales, Universidad de La Frontera, 4811-230 Temuco, Chile, ²Departamento de Ciencias Químicas y Recursos Naturales, Universidad de La Frontera, 4811-230 Temuco, Chile, ³Departamento de Biologia, Instituto de Ciências Biológicas-ICB, Universidade Federal do Amazonas, 69077-000 Manaus, AM, Brazil, ⁴CEB-Centre of Biological Engineering, Micoteca da Universidade do Minho (MUM), University of Minho, Campus de Gualtar, 4710-057 Braga, Portugal, ⁵LABBELS (Associate Laboratory, Braga/Guimarães), University of Minho, Campus de Gualtar, 4710-057 Braga, Portugal, ⁶Escuela de Agronomía, Facultad de Ciencias Agronómicas y de los Alimentos, Pontificia Universidad Católica de Valparaíso, Quillota, Chile

The increase in fungal resistance to synthetic antifungals used in agrifood production has brought the need to develop new technologies based on an eco-friendly approach. The main aim of this work was to evaluate the antifungal potential of Chilean *Capsicum* spp. extracts against plant pathogens and mycotoxigenic fungi found in agrifood production.

Five different varieties of Chilean *Capsicum* spp. were obtained from both farmers and local markets in the city of Temuco, Chile. A specialist Botanist at the Universidad de La Frontera (Chile) confirmed the identification of pepper species and varieties. Fresh samples were grounded with a blender and freeze-dried for 7 days in the dark. After that, dry powder samples were stored at -20 °C in the dark until use. Pepper pod aqueous extracts were obtained by blending the freeze-dried puree from *Capsicum* spp. with 300 mL distilled water. Samples were incubated at 90°C for 20 minutes in a water bath with intermittent cycles of manual stirring every 2 minutes. The determination of capsaicinoid content was performed on an HPLC-FD system and the total polyphenols content was performed on an HPLC-DAD system. The antioxidant activity was carried out in a microplate reader using the DPPH and CUPRAC method. Reference strains of *Alternaria, Aspergillus, Fusarium, Penicillium* and *Rhizopus* were subjected to susceptibility tests (disc and culture media diffusion methods and MIC assay) against different concentrations of each pepper pod extract. Pure capsaicin, dihydrocapsaicin, nordihydrocapsaicin and amphotericin B were used as standard in the susceptibility tests.

Significant differences in the concentration of capsaicinoids were found among the different varieties of the same *Capsicum* species. The pepper pod extracts affected the macro- and micro-morphological features of the analysed filamentous fungal strains. Fungal strains belonging to the genera *Alternaria*, *Aspergillus*, *Fusarium*, *Penicillium* and *Rhizopus* produced mycelium with thinning, fragile and easily-break structure. In addition, their conidiophores became fragile presenting easily-break structures. Regarding other fungal genera (data not shown), the main alteration was the absence of conidiophore formation in some strains. The morphological changes observed in the filamentous fungi strains suggest the fungistatic

potential of pepper pod extracts. Results suggest pepper pod extracts could not kill non-

target fungal biodiversity but could control the growth and reproduction of some fungal plant pathogens. Inhibition of mycotoxin production is now under evaluation. Additional work is being developed in the field to validate the in vitro results.

Acknowledgments: ANID (Agencia Nacional de Investigación y Desarrollo, Chile) through the ANID/FONDECYT/1221024 project. This work was partially funded by the Universidad de La Frontera (Chile). Authors thank the InES19-FRO19001 project, funded by the Ministerio de Educación (Chile) and executed by the Universidad de La Frontera. The authors thank Professor Ruben Carrillo (UFRO) for his support in confirming the pepper species and varieties identification.