Título: ANALYSIS AND GENETIC CHARACTERIZATION OF ISOLATES *Sclerotinia sclerotiorum* FOR PECTINASE PRODUCTION

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Abstract:

The *Sclerotinia sclerotiorum* is a ascomycete fungus that has a great economic importance given its functional capacity of infecting a wide range plants. The recent sequencing of its genome has confirmed the presence of a wide variety of genes encoding enzymes of biotechnological interest. The pectinases is considered one of great economic relevance, although there are others that have also been investigated. Microbial pectinolytic enzymes are widely used mainly in food industry, processing of fruit juices and wines in order to increase the efficiency of extraction, clarification and pulp viscosity redution. Previous studies in the genome of *S. sclerotiorum* found 45 genes that are involved in the degradation of plant cell wall. The aim of this study was to identify and characterize the genomic sequences of *S. sclerotiorum* encoding pectinases and evaluate the production of polygalacturonase (PG) and pectin lyase (PL) by different strains of this fungus. For this end, we can see that 72 isolates were evaluated for pectinase production in solid medium. All isolates analyzed produced degradation halo on the pectin solid medium. The most promising eights were analyzed of pectinases in liquid medium to quantify the polygalacturonase and pectin lyase enzymatic activities. In this study, we did not find PL producers, but three isolates (SSA 06B, SSA 01A and SSA11C) showed the highest enzymatic activities of PG when compared to control strains (*Penicillium griseoroseum* WT and a nitrate reductase mutant *Penicillium griseoroseum* 63). The analysis of regulatory regions of the sequences encoding pectinase revealed that these genes are under control of transcriptional elements like PacC (pH-responsive transcription factor), CreA (carbon catabolite repression), and Hap (CCAAT-binding complex). The analysis of the protein sequences from *S. sclerotiorum* indicated low identity with others fungal enzymes produced by representatives from the genus *Penicillium* and *Aspergillus*. The data also showed that these proteins can be vary widely in molecular weight and pI. The promising isolate will be used for isolation of polygalacturonase genes for genetic improvement and over production of the enzymes by the fungus *Penicillium griseoroseum* PG63, that is an efficient expression host.

Key Words: pectinases, genetic characterization, polygalacturonase, pectin lyase, *Sclerotinia sclerotiorum*

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