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**ABSTRACTS**



## PHENOLIC AND POLYSACCHARIDIC EXTRACTS OF *SUILLUS COLLINITUS*: CHEMICAL CHARACTERIZATION, GROWTH INHIBITORY ACTIVITY AND INDUCTION OF CELL CYCLE ARREST IN A BREAST CANCER CELL LINE

J. A. Vaz<sup>1,2,3,4</sup>, C. Tavares<sup>2</sup>, G. M. Almeida<sup>2</sup>, A. Martins<sup>1</sup>, M. H. Vasconcelos<sup>2,4</sup>,  
I. C.F.R. Ferreira<sup>1</sup>

<sup>1</sup> CIMO- Instituto Politécnico de Bragança Campus de Santa Apolónia, Apartado 1172, 5301-855 Bragança, Portugal.

<sup>2</sup> Cancer Drug Resistance Group, IPATIMUP - Institute of Molecular Pathology and Immunology of the University of Porto, Portugal.

<sup>3</sup> CEQUIMED-UP - Center of Medicinal Chemistry- University of Porto, Portugal.

<sup>4</sup> Department of Biological Sciences, Faculty of Pharmacy, University of Porto, Portugal

### INTRODUCTION

The Northeast of Portugal is one of the European regions with higher mushrooms biodiversity. Mushrooms are a powerful source of new compounds, including low-molecular-weight (LMW, e.g. phenolic compounds) and high-molecular-weight (HMW, e.g. polysaccharides) compounds, with potential antitumor and immunostimulating properties [1,2]. Furthermore, they may have potential as functional foods and sources of novel molecules. The aim of this work was to study the antitumor potential of the wild edible ectomycorrhizal mushroom, *Suillus collinitus*, collected from Northeast Portugal, and to characterize their LMW and HMW compounds.

### MATERIAL AND METHODS

Phenolic and polysaccharidic extracts were prepared and extract-induced cell growth inhibition was assessed with the sulforhodamine B assay in four human tumour cell lines (lung, breast, colon and gastric cancer). Both extracts were further characterized by chromatographic techniques: HPLC-DAD and HPLC-RI for analysis of phenolic and polysaccharidic extracts, respectively. Furthermore, the effect of the best extract on the cell cycle of the most susceptible human tumour cells (MCF-7) was evaluated by flow cytometry following PI staining.

### RESULTS AND DISCUSSION

The phenolic extract was more potent than the polysaccharidic extract. From the phenolic extracts, the methanolic extract was the most potent one, particularly in MCF-7 cells ( $GI_{50}$   $25.2 \pm 0.16$   $\mu\text{g/ml}$ ).

**Table 1.** Effect of extracts of *Suillus collinitus* on the growth of human tumour cell lines.

Extracts	NCI-H460 (lung cancer)	MCF-7 (breast cancer)	HCT-15 (colon cancer)	AGS (gastric cancer)
Phenolic (methanolic)	62.5 $\pm$ 6.3	25.2 $\pm$ 0.16	103.2 $\pm$ 9.9	79.2 $\pm$ 15.5
Phenolic (ethanolic)	407.4 $\pm$ 2.3	101.8 $\pm$ 8.9	139.4 $\pm$ 34.1	170.7 $\pm$ 35
Polysaccharidic (boiled water)	> 800	> 800	> 800	> 800

Results are expressed as  $GI_{50}$  (concentrations of extract in  $\mu\text{g/ml}$  that cause 50% of cell growth inhibition), and show means  $\pm$  SEM of 3-6 independent experiments performed in duplicate.

In order to further understand the growth inhibitory effect of the *Suillus collinitus* methanolic extract in the MCF-7 cells, the effects on cell cycle (following PI labelling) were evaluated by flow cytometry.

Results indicate that *Suillus collinitus* methanolic extract induced a cell cycle arrest in the G1 phase with a concomitant decrease in the percentage of cells in the S phase. Additionally, there was an increase in the percentage of cells in the sub-G1 region, indicating that this extract might induce apoptosis.

In order to identify the molecules responsible for the bioactive properties, the mushroom extracts were further chemically characterized and revealed:

- i) Phenolic extracts: protocatechuic acid ( $5.2 \pm 0.2$  mg/Kg dw), *p*-hydroxybenzoic acid ( $14.1 \pm 1.2$  mg/Kg) and cinnamic acid ( $1.3 \pm 0.2$  mg/Kg);
- ii) Polysaccharidic extracts after polysaccharides hydrolysis: arabinose ( $30.3 \pm 5.5$  g/Kg), mannitol ( $32.2 \pm 4.1$  g/Kg) and trehalose ( $11.0 \pm 0.2$  g/Kg), (Fig. 1).

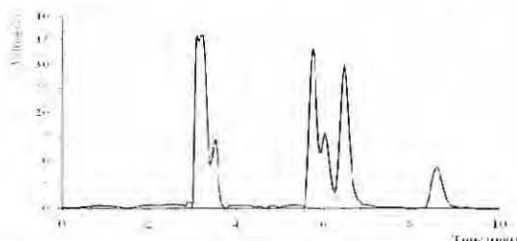


Figure 1. Sugars profile of *Suillus collinitus*. 1- Arabinose; 2- Fructose (IS); 3- Mannitol; 4- Trehalose

## CONCLUSIONS

The results indicate that *Suillus collinitus* is a promising source of bioactive LMW and HMW compounds. Particularly, its methanolic extract has an effect on the normal cell cycle distribution of the MCF-7 human breast tumor cells. It will be important to study the activity of the individual compounds identified in the phenolic extract and to conclude about the role of those molecules in the bioactivity of the whole extract.

## REFERENCES

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