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BIOACTIVITY OF *Agaricus bisporus* AND *Hericium erinaceus* MUSHROOMS BY-PRODUCTS

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ABSTRACT

Extracts obtained from *Hericium erinaceus* (HeE) and *Agaricus bisporus* (AbE) solid waste were investigated for their capacity to modulate the growth of the probiotic (*E. coli* Nissle 1917) and foodborne pathogen (*E. coli* O157:H7) *Escherichia* strains, and cytotoxic potential against the human hepatocellular carcinoma cell line (HepG2). In selected assays, the tested concentrations did not exceed 1 mg/mL. HeE extract stimulated growth, while AbE extract did not disturb the growth dynamics of both *E. coli* strains. HeE extract exhibited significant cytotoxicity towards HepG2 cells (up to 43.1%). The obtained results confirmed that not only medicinal and edible mushrooms, but also their by-products, could exhibit biological effects.

INTRODUCTION

Mushroom production generates a large amount of by-products such as waste and off-grade mushrooms that lack market value, that ranges between 5% and 20% of production volume [1]. By-products generated during mushroom production and processing are repurposed for energy production and bioremediation, and could find applications as animal feed, fertilizer, bio-based materials, and cosmeceuticals [2]. With this in mind, the discarded material from mushroom extraction could retain bioactives, the levels of which vary according to the specific extraction technique used. To date, components derived from specific medicinal mushrooms have become valuable ingredients of a functional food and dietary supplements [3]. Lion's mane mushroom, *Hericium erinaceus,* based supplements and products exhibit a diverse array of biological activities, including antimicrobial, immunoregulating, antioxidant, anti-inflammatory, hypolipidemic, and antiviral functions. In general, edible and medicinal mushrooms could serve as prebiotics, promoting the formation of the gut microbiota and so providing health benefits to the host [4]. *H. erinaceus* products

are available in various forms, including health wines, beverages, teas, extracts and solid food products [5]. Among the frequently grown varieties is also the button mushroom, *Agaricus bisporus* (J.E. Lange), which is widely cultivated in Europe and comprises about 32% of the world mushroom production [1]. The purpose of this study was to verify whether fungal by-products, much like the fungi they originate from, can be considered as carriers of biological activities. Therefore, this study investigates whether the extracts prepared from *A. bisporus* and *H. erinaceus* solid waste (AbE and HeE) can modulate growth of both probiotic and foodborne pathogen *Escherichia coli* strains and exert *in vitro* cytotoxicity towards hepatocarcinoma cells (HepG2).

EXPERIMENTAL

Mushrooms by-products were collected after commercial preparation of water extracts (Serbia). Here, AbE and HeE solid waste has been subjected to maceration in a 50% water/ethanol solution for 24 h with constant stirring (150 rpm). Then the liquid was separated from the plant material, filtered, and centrifuged (6200×g, 10 min). The solvent was evaporated using a rotary evaporator at 39°C under vacuum. The dried extracts were dissolved in ethanol to a final concentration of 100 mg/mL. Determination of growth curves was based on the methodology of Lehtinen *et al*. [6]. A cytotoxicity assay was carried out against HepG2 cells with AbE and HeE by employing an MTT assay [7]. GraphPad Prism 6.0 software was used for statistical data analysis.

RESULTS AND DISCUSSION

E. coli is the most dominant gram-negative bacterium of the normal intestinal microbiota. Among *E. coli* strains, probiotic strains, as well as pathogens and pathobionts, were recognized. Our study assessed the growth dynamics of probiotic *E. coli* Nissle 1917 and the foodborne pathogen *E. coli* O157:H7 with and without AbE and HeE extracts (Figure 1). HeE extract stimulated growth of both *E. coli* strains, while AbE extracts did not disturb the growth dynamics of *E. coli* strains. By contrast, *A. bisporus* was capable to stimulate the growth of G-positive probiotic bacteria. El-Maradny [4] demonstrated that polysaccharide extracts from *A. bisporus* significantly stimulated the growth of *L. paracasei*.
Contrary to our results, Ramos et al. [8] discussed that crude extracts obtained from *A. bisporus* exert anti-*E. coli* activity. It has also been discussed that *H. erinaceus* exert antibacterial effect against G-positive and G-negative bacteria including *E. coli* [9].

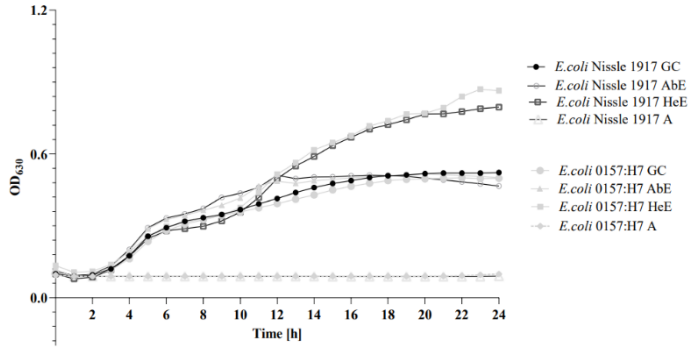


Figure 1. Growth dynamic of the *E. coli* strains during 24 h with AbE and HeE treatment. GC-growth control; A-antibiotic, vancomycin.

Previously, it has been confirmed that compounds isolated from *H. erinaceus,* such as the bioactive peptide Lys-Ser-Pro-Leu-Tyr (KSPLY, Mw 608.38 Da) lack cytotoxic potential. Moreover, it contributed to the proliferation of HepG2 cells [10]. In our study, HeE extracts significantly decreased the viability of HepG2 cells (up to 43.1%). Next, literature data indicated that *A. bisporus* exhibited cytotoxicity against HepG2 [11]. Within this study, we found no evidence of cytotoxicity in the by-products derived from *A. bisporus*.

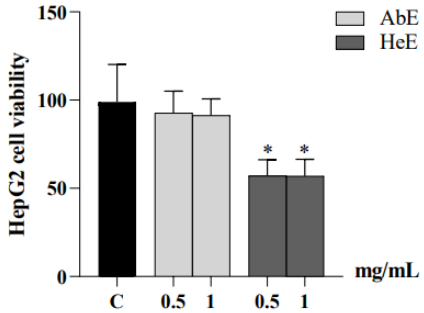


Figure 2. HepG2 cell viability. C-growth control. A significant difference in means between samples and control according to the Dunnett's test ($p<0.05$).

CONCLUSION

Edible mushrooms are recognized not only for their nutritional value but also for their diverse biological activities. Here, it was confirmed that extract from *Hericium erinaceus* solid waste can also exhibit prominent bioactivity.

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