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Design of functional foods with new ingredients from Pleurotus ostreatus grown on agri-food waste

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This PhD project aims at the development of applications of *Pleurotus ostreatus* biomass grown on a "sustainable" substrate in the production of added-value foods. In the first step, the project will lead to the definition of processing conditions to convert *P. ostreatus* grown on winemaking by-products into two food ingredients: the totally dried whole biomass and the isolated β-glucan-rich fraction encapsulating an oil-in-water emulsion of an oxidisable target. In the second step, model applications of these ingredients will be studied aimed at the development of vegetable purees enriched in vitamin D_2, β-glucans and an oxidisable bioactive compound.

Progettazione di alimenti funzionali con nuovi ingredienti ottenuti da *Pleurotus ostreatus* cresciuto su scarti agro-alimentari

Questo progetto di tesi di dottorato mira allo sviluppo di applicazioni della biomassa di *Pleurotus ostreatus* cresciuto su un substrato "sostenibile" nella produzione di alimenti a valore aggiunto. Nella prima fase, verranno definite le condizioni di trasformazione per la conversione di *P. ostreatus* coltivato sui sottoprodotti della vinificazione in due ingredienti alimentari: la biomassa totalmente essiccata e la frazione isolata di β-glucani, che verrà usata come incapsulante di un'emulsione olio-in-acqua contenente un target ossidabile. Nella seconda fase verranno studiate le applicazioni modello di questi ingredienti, allo scopo di mettere a punto delle puree vegetali arricchite in vitamina D_2, β-glucani e un composto bioattivo ossidabile.

1. State-of-the-Art

The food industry generates a high amount of waste causing a relevant impact on the decrease of natural resources and the environmental pollution. Consequently, there is an increasing need of waste management strategies: the basidiomycetes fungi belonging to the genus *Pleurotus* can make a valuable contribution, through the conversion of waste into added value foods. *Pleurotus* spp. can efficiently decompose lignocellulose due to their enzymatic complex system that includes phenol oxidases and peroxidases, yielding a fungal biomass that represents a source of protein with good level of essential aminoacid content, dietary fibre with unique structural features ((1/3),(1/6)-linked β-glucans and linear (1/3)-α-glucan) and low-molecular weight bioactive compounds, especially phenolics and ergosterol, which is the precursor of vitamin D_2. The natural level of vitamin D_2 in *Pleurotus* spp. is generally low; however, it could increase by light exposure of mushroom through ergosterol conversion (Lavelli *et al.*, 2018). Food uses of the fresh fruit body of *Pleurotus* are limited by its very short storage time i.e. 3-12 days from harvest at temperatures below 8 °C. Hence, the formulation of processed foods supplemented with ingredients derived from *Pleurotus* spp. is a promising area of study to enhance their nutritional and nutraceutical values.*Pleurotus* β-glucans have shown antimicrobial, anticancer, antioxidant, hypolipidemic, hypocholesterolemic, antihyperglycemic and immunomodulatory effects. Moreover, they have enormous potential in a wide variety of fields due to their gel-forming capacity. In the recent years, the potential use of cereal β-glucans as encapsulation carriers for delivery-controlled release of probiotics and nutraceuticals has begun to be explored (Lazaridou *et al.*, 2015; Salgado *et al.*, 2015). Conversely, the use of mushroom β-glucans is still unexplored. The stability of bioactive compounds is a critical parameter for their successful incorporation into various food systems. Specifically, health-promoting bioactive compounds such as vitamins, probiotics, polyphenols, PUFAs and phytosterols are sensitive to oxygen, light, heat, and water (Turchiuli *et al.*, 2017). Moreover, after been orally consumed, biological components are subjected to rapid intestinal and first-pass metabolism, causing the transformation of their chemical structure, and changes in their bioactivities. An additional issue arising during the implementation of bioactive compounds is to minimize possible undesirable odours, flavours and tastes (Lavelli *et al.*, 2017). Encapsulation is a powerful tool for overcoming the abovementioned issues and mushroom β-glucans could be a sustainable and healthy carrier polymer as an alternative to the most common carrier polymers, i.e. maltodextrins, arabic gum, whey proteins and cereal β-glucans.

2. PhD Thesis Objectives and Milestones

Within the overall objective mentioned above this PhD thesis project can be divided into the following phases reported in the Gantt diagram (Table 1):

A1) **Collection, processing and characterization of the biomass of *P. ostreatus*** (both commercial and cultivated on a grape waste substrate). Fungal biomass will be air-dried, milled (A1.1) and characterized for hygroscopicity and storage stability. The major bioactive compounds, namely: proteins, β-glucans, ergosterol, vitamin D_2 and phenolics will be evaluated by spectrophotometric and HPLC procedures. Antioxidant and antiglycation activities will be studied *in vitro* by different model reactions (A1.2).

A2) **Encapsulation of an oxidisable target using *Pleurotus* β-glucans as a carrier.** Extraction of β-glucans with a "food-grade" procedure will be optimized by measuring the yield, antioxidant and antiglycation activity (A2.1). The encapsulation technique will be carried out using a pilot scale spray dryer. Two series of spray drying conditions corresponding to slow and fast drying kinetics, respectively, will be tested in order to define the optimum drying conditions. The powder characterization will include: size distribution by laser light diffraction with a Mastersizer; particle morphology by Scanning Electron Microscopy (SEM). The oxidisable target content in the oil phase of powder will be measured with a HPLC system (A2.2). Powder oxidative stability will be evaluated in samples stored under accelerated thermo-oxidation conditions (A2.3).

A3) **Development of food applications.** The dehydrated fungal biomass and its β-glucan fraction will be used in various amounts as ingredients for three model functional foods. The first model food will be enriched in vitamin D_2 (A3.1), the second one in β-glucans (A3.2) and the third one will be added with the encapsulated oxidisable target (A3.3). The effect of processing on the stability of the bioactive compounds of the model food will be studied and kinetics model will be developed to minimize degradation phenomena (oxidation, browning). To select which formulations have the highest potential liking by consumers, the innovative food prototypes will be assessed by a consumers' preference study.

A4) **Writing and Editing** of the PhD thesis, scientific papers and oral and/or poster communications.

Table 1 *Gantt diagram for this PhD thesis project.*

Activity / Months	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25	26	27	28	29	30	31	32	33	34	35	36
A1) Processing of the fungal biomass																																				
1) Powder production																																				
2) Characterization and storage study																																				
A2) Development of encapsulated ingredient																																				
1) β-glucans extraction																																				
2) Powder development																																				
3) Characterization and storage study																																				
A3) Design of functional foods																																				
1) Vitamin D_2-enriched																																				
2) β-glucans-enriched																																				
3) Oxidisable target enriched																																				
A4) Writing and editing																																				

3. Selected References

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