

Fermentation of *Hericium erinaceus* Byproducts: A Microbial Stability Assessment

Mafalda Silva¹, Manuela Vida¹, Cláudia Correia¹, Ana Cristina Ramos^{1,2}, Fernando Reboredo², Elsa Gonçalves^{1,2}

¹ Unidade de Tecnologia e Inovação (UTI), Instituto Nacional de Investigação Agrária e Veterinária, I.P., Oeiras, 2780-157, Portugal

² GeoBioTec – Departamento de Ciências da Terra, Faculdade de Ciências e Tecnologia, Universidade NOVA de Lisboa, Caparica, 2829-516, Portugal



Instituto Nacional de
Investigação Agrária e
Veterinária, I.P.



GeoBioTec
GeoBioSciences, GeoTechnologies and
GeoEngineering

INTRODUCTION & AIM

Fermentation is a well-established technique used to preserve and enhance food by harnessing the metabolic activity of microorganisms. Through the production of various bioactive substances, this process can effectively suppress the growth of spoilage and pathogenic microbes.

A particularly efficient form is lactic acid fermentation, which relies on lactic acid bacteria (LAB) known for generating antimicrobial compounds, including bacteriocins. Despite its broad application in food systems, the fermentation of mushrooms, particularly for microbial control and shelf-life extension, remains under-investigated.

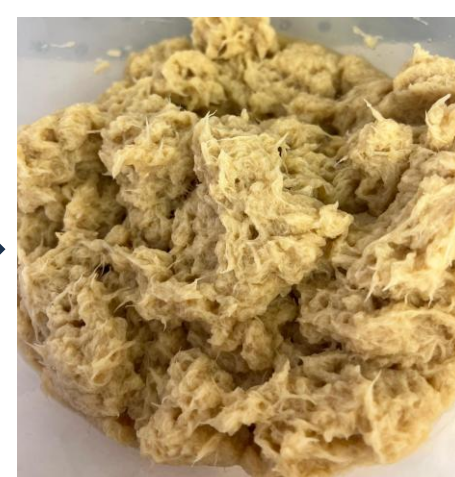
This study explores the biotechnological potential of *Hericium erinaceus* fermentation under three conditions: spontaneous fermentation, and fermentation inoculated with either *Lactobacillus plantarum* or *Lactobacillus casei*.

METHOD

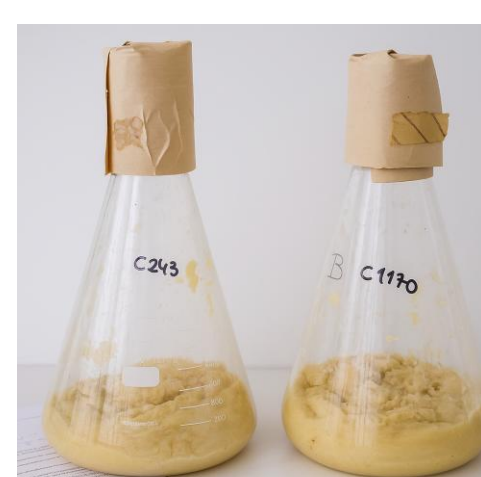
The fermentation substrate consisted of *Hericium erinaceus* fruiting bodies that were physically damaged or otherwise deemed unsuitable for commercial sale. These were classified as mushroom byproducts and represent a potential source of food waste. The procedure was composed by the steps below described:



Blanching in boiling water for 3 minutes



Homogenization with 2% NaCl



LAB inoculation
33 mL of 1.5×10^9 UFC/mL per
500 mL of homogenate



240 hour fermentation
at 21 °C

Microbiologic analysis: 10 g samples were aseptically collected in triplicate at 0 h, 24 h, 72 h, 144 h, and 240 h. Microbial determinations, expressed as log CFU/g, were made according to:

LAB - ISO 15214:1998 | *Pseudomonas* spp. - ISO 13720:2010 |

Yeasts and molds - ISO 21527-1.

Chemical determination: pH and titratable acidity, following the NP-1421 standard.

Statistical Analysis: Data were statistically analyzed using ANOVA and Tukey's HSD test ($\alpha = 0.05$) via Statistica™ v.8 software.

RESULTS & DISCUSSION

The results revealed that *L. plantarum* led to a rapid acidification of the substrate, with the pH dropping below 4.0 within the first 72 hours, which corresponded to a significant increase ($p < 0.05$) in TA (Figure 1).

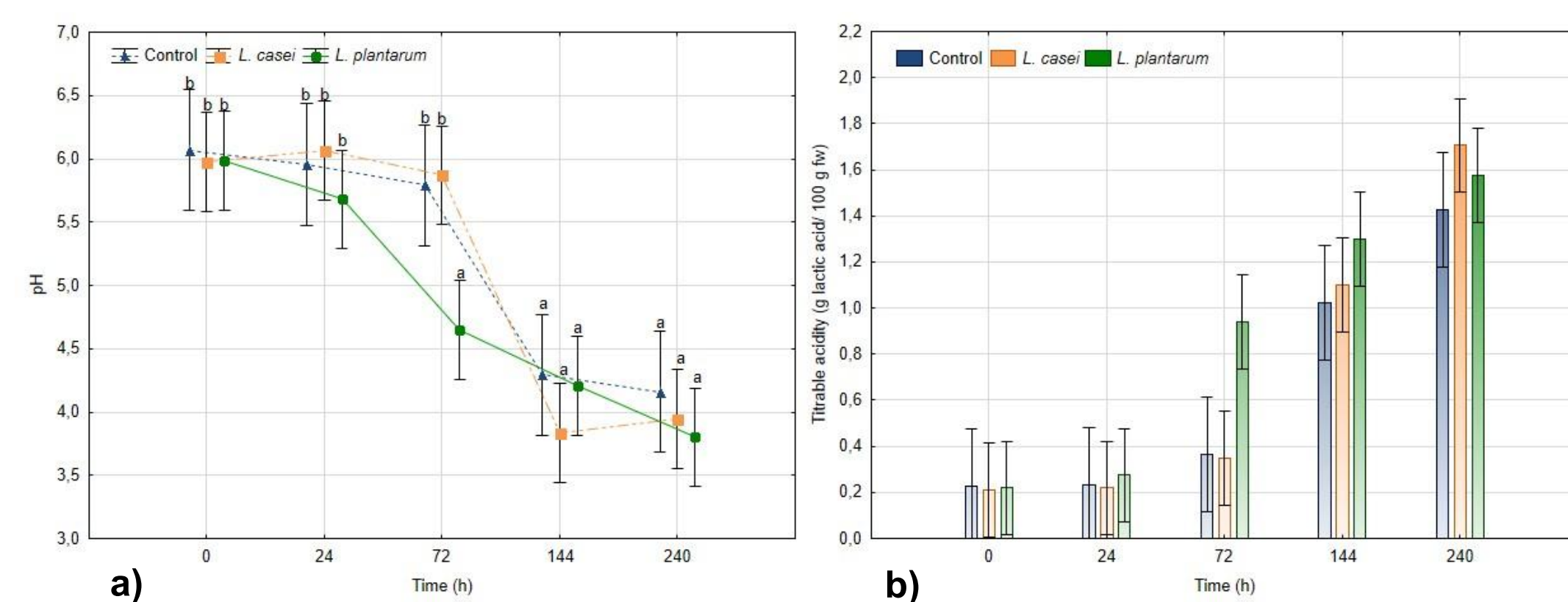


Figure 1. Chemical evolution during fermentation: a) pH and b) titratable acidity (TA).

The inoculated fermentations showed a clear antimicrobial advantage, with both *L. plantarum* and *L. casei* rapidly reducing *Pseudomonas* spp. and yeast/mold populations to below detectable levels after 144 hours (Figure 2a,b). This effect coincided with a strong acidification (Fig1a) and increase in titratable acidity (Fig1b), confirming that the metabolic activity of the inoculated LAB strains effectively suppressed spoilage and pathogenic microorganisms. Among the two inoculants, *L. plantarum* exhibited faster substrate acidification, whereas *L. casei* showed a more gradual but sustained inhibitory effect, consistent with its higher final acidity.

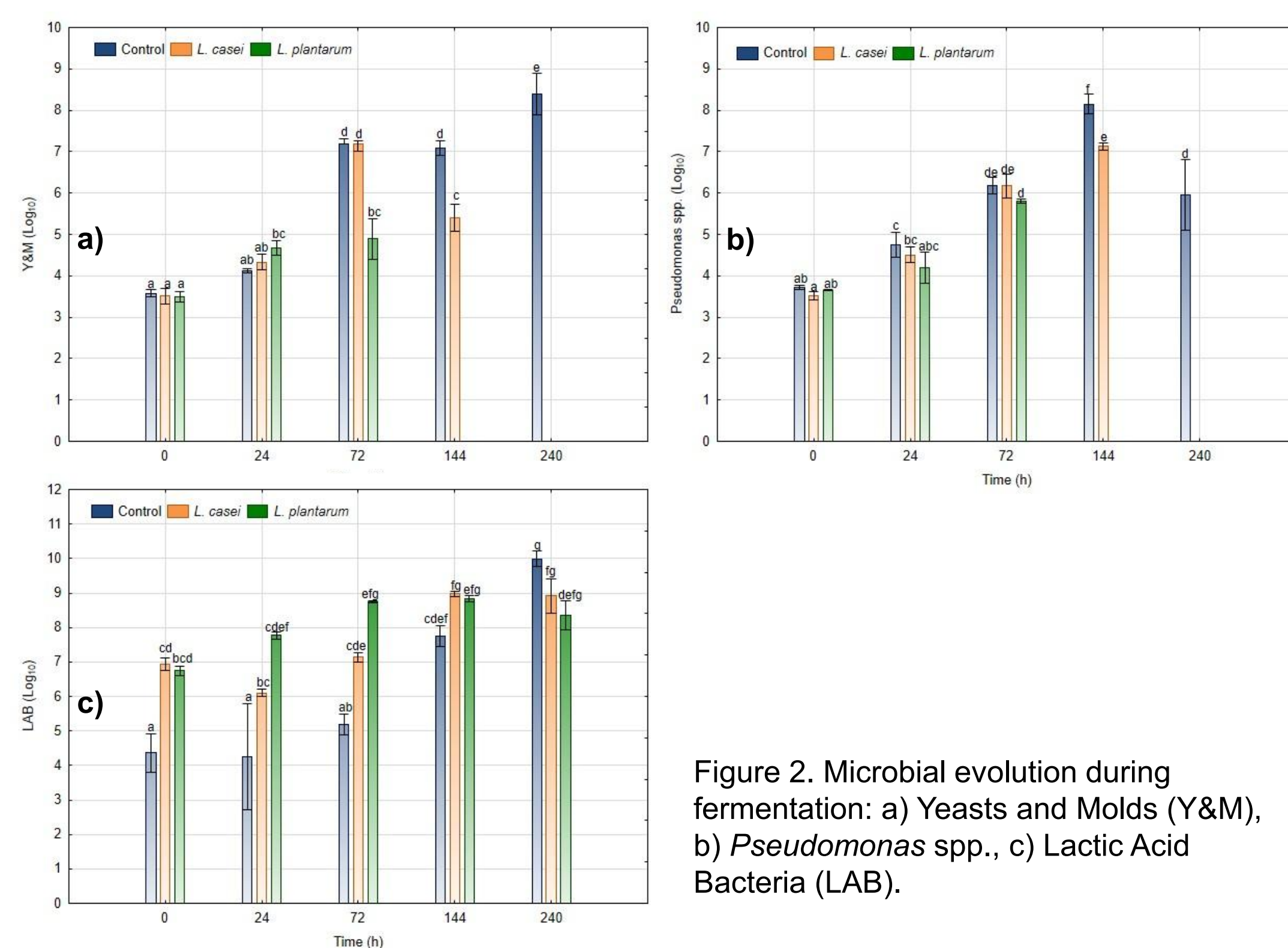


Figure 2. Microbial evolution during fermentation: a) Yeasts and Molds (Y&M), b) *Pseudomonas* spp., c) Lactic Acid Bacteria (LAB).

In the uninoculated control, a native LAB population developed naturally, likely favored by the saline conditions of the substrate. However, acidification progressed more slowly, and the reduction of *Pseudomonas* spp. and yeast/mold populations was limited. Although a moderate decline in spoilage organisms was observed after 144 hours, microbial stabilization was incomplete when compared with the inoculated fermentations, highlighting the lower antimicrobial efficiency of the spontaneous process.



CONCLUSION

MICROBIAL STABILIZATION



Inoculated fermentations with *L. plantarum* and *L. casei* achieved complete suppression of *Pseudomonas* spp. and yeasts/molds by 144 h. Demonstrated strong antimicrobial activity and rapid microbial control.

FERMENTATION DYNAMICS



Rapid acidification and high titratable acidity confirmed efficient LAB metabolism. *L. plantarum* → faster pH drop. *L. casei* → slower but sustained inhibitory effect.

CONTROL SAMPLES



Spontaneous fermentation showed limited microbial inhibition and slower acidification. Highlights the need for controlled inoculation to ensure safety and reproducibility.

SUSTAINABILITY IMPACT



Fermentation represents a viable strategy for valorizing *Hericium erinaceus* byproducts. Contributes to food waste reduction, extended shelf life, and microbial safety enhancement.