TISOCHRYSIS LUTEA BIOMASS AS A SUBSTRATE FOR LACTIC ACID FERMENTATION

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ABSTRACT

Microalgae contain high quality proteins, polyunsaturated fatty acids and bioactive molecules endowed with beneficial properties (Lafarga et al., 2021). They are, thus, considered interesting candidates for nutraceuticals and functional foods (Matos et al., 2017). The uses of microalgae in food production include the development of functional fermented foods. Lactic fermentation allows to add the beneficial effects of probiotic bacteria to the useful biomolecules lying in microalgal biomass, also taking advantage of the effects on organoleptic features produced by fermentation. The aims of this work were to evaluate the potential of *Tysochrysis lutea* F&M-M36 lyophilized biomass as a substrate for *Lactiplantibacillus plantarum* ATCC 8014 fermentation, as well as to investigate the effects of the indigestible fraction of the microalgal biomass on bacterial growth, and to verify whether the microalgal biomass could exert a protective role towards the probiotic bacterium during a simulated digestion process. Initially, three lactobacilli were characterized for growth and resistance to simulated digestion, then the best strain (*L. plantarum* ATCC 8014) was selected for the fermentation trial. This latter was set up with *T. lutea* F&M-M36 raw and digested biomasses suspended in two different matrixes. The biomasses obtained at the end of the fermentation trial were analyzed for gross biochemical composition, digestibility and radical scavenging activity and compared with the unfermented biomasses; moreover, survivability of *L. plantarum* ATCC 8014 to in vitro digestion was also evaluated. The higher growth and organic acid production was observed with microalgal raw biomass suspended in diluted organic medium. No protective effect on bacterial survivability was observed after fermentation. Radical scavenging activity of a lipophilic extract was higher in fermented than in raw microalgal biomass and was also higher than in the fermented post-digestion residue. Fermentation seems an interesting process to obtain functional foods from *T. lutea*, although further investigation is needed to optimize the matrix for fermentation so as to improve bacterial growth, and thus fully elucidate the role of fermentation for functionality improvement.

Keywords
Tisochrysis lutea, lactic fermentation, lactic bacteria survivability

References

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BIOGRAPHY

Natascia Biondi, PhD in Microbial Biotechnology, is a researcher at the Department of Agriculture, Food, Environment and Forestry (DAGRI) of the University of Florence. She has more than 20 years of experience in the microalgae field and has participated to several EC funded projects (FP4, FP7, H2020). She is secretary of the Italian Association for the Study and Application of Microalgae (AISAM). Main topics of research: microalgae cultivation and application in the food, nutraceutical and agricultural fields.

Tisochrysis lutea biomass as a substrate for lactic acid fermentation

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1. Introduction

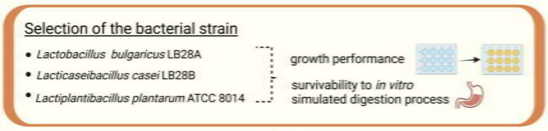
Microalgae contain high quality proteins, polyunsaturated fatty acids and bioactive molecules endowed with beneficial properties[1]. They are, thus, considered interesting candidates for nutraceuticals and functional foods[2]. The uses of microalgae in food production include the development of functional fermented foods. Lactic fermentation allows to add the beneficial effects of probiotic bacteria to the useful biomolecules lying in microalgal biomass, also taking advantage of the effects on organoleptic features produced by fermentation. *Tisochrysis lutea* is a marine haptophyte widely used in aquaculture and known to exert positive effects on lipid metabolism in rats[3] and to have anti-inflammatory activity[4], being thus an excellent candidate for functional food production.

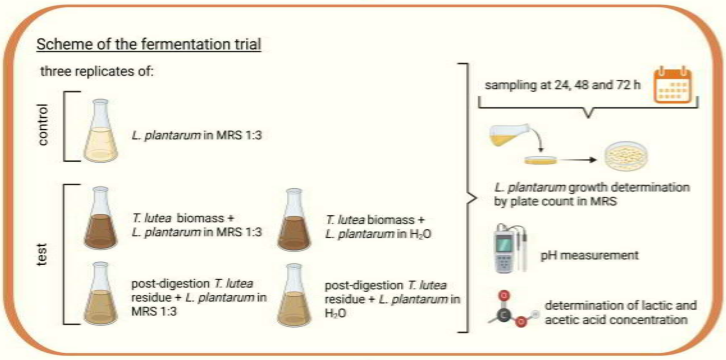
2. Aim

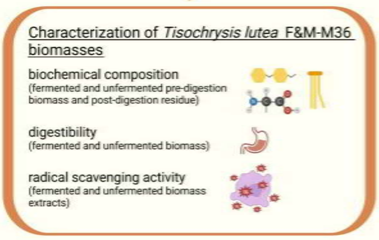
The aims of this work were to evaluate the potential of *Tysochrysis lutea* F&M-M36 lyophilized biomass as a substrate for *Lactiplantibacillus plantarum* ATCC 8014 fermentation, as well as to investigate the effects of the indigestible fraction of the microalgal biomass on bacterial growth, and to verify whether the microalgal biomass could exert a protective role towards the probiotic bacterium during a simulated digestion process.

3. Methods

Initially, three lactobacilli belonging to three different genera were characterized for growth and resistance to *in vitro* simulated digestion, then the best strain was selected for the fermentation trial. This latter was set up with *T. lutea* F&M-M36 raw and digested biomasses suspended in two different matrixes (H_2O and diluted organic medium, MRS 1:3). The raw biomasses obtained at the end of the fermentation trial were analyzed for gross biochemical composition, digestibility and radical scavenging activity and compared with the unfermented raw biomasses; moreover, bacterial survivability to *in vitro* digestion in the fermented product was also evaluated.







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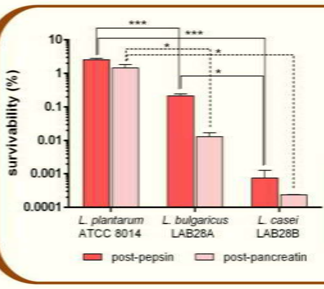
4. Results

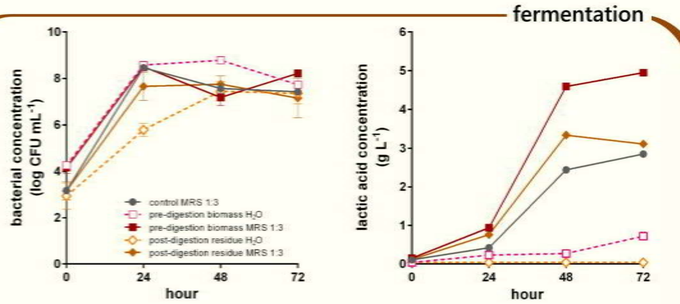
bacterial strain selection



	Growth rate (h^{-1})
L. plantarum ATCC 8014	0.33
L. bulgaricus LB28A	0.24
L. casei LB28B	0.17

Lactiplantibacillus plantarum showed the highest growth rate (top) and survivability to *in vitro* digestion (left) and was selected for the following fermentation trial.

fermentation



Higher growth (left) and organic acid production (right) was observed with microalgal raw biomass suspended in MRS 1:3.

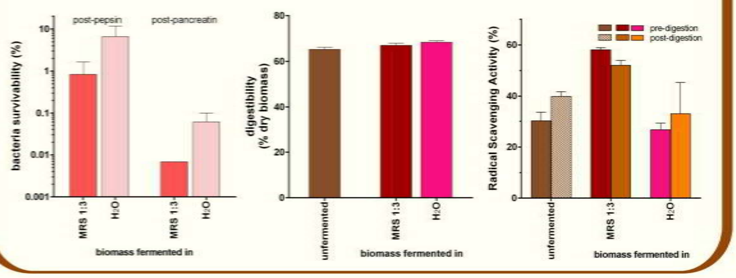
characterisation of biomasses

Biochemical composition

% dry weight	Protein	Carbohydrate	Lipid	Ash	TDF
Pre-digestion biomass					
unfermented	42.0 ± 2.5	12.6 ± 0.7[a]	29.3 ± 0.3	11.9	9.3
fermented in H_2O	48.6 ± 4.0	7.2 ± 0.5[b]	32.6 ± 5.7	9.1	nd
fermented in MRS 1:3	48.9 ± 1.1	7.9 ± 0.4[b]	33.4 ± 4.4	6.2	nd
Post-digestion residue					
unfermented	45.9 ± 4.1	10.2 ± 0.4[a]	30.6 ± 0.3	10.7	7.4± 1.3
fermented in H_2O	43.8 ± 1.0	8.8 ± 0.8	33.4 ± 3.4	5.8	nd
fermented in MRS 1:3	39.8 ± 1.6	8.3 ± 0.1[b]	32.9 ± 6.6	5.7	nd

nd, not determined

No effect on bacterial survivability, nor on biomass digestibility, was observed after fermentation. Radical scavenging activity of a $CHCl_3$:MeOH 1:2 extract was higher in microalgal biomass fermented in organic medium than in unfermented biomass and in post-digestion residue, either fermented or not.



5. Conclusions

- *Lactiplantibacillus plantarum* ATCC 8014 appeared to have the best probiotic characteristics among the tested strain
- fermentation did not improve functional characteristics, like probiotic strain viability, nor the digestibility of microalgal biomass, but improved microalgal biomass antioxidant activity
- prebiotic action seems to play only a minor role in *L. plantarum* growth enhancement determined by *Tisochrysis lutea* F&M-M36; further studies should be performed to clarify this point
- further investigation is needed to optimize the matrix for fermentation, which exerts a strong influence on fermentation outcomes, so as to improve bacterial growth and fully elucidate the role of fermentation for functionality

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