Valorization of defatted microalgae biomass and rapeseed as sources of nitrogen-rich concentrates for microalgae culture

Suarez-Alvarez S., Izaguirre J.K, Urreta I. and Castañón de la Torre, S.
Neiker-Tecnalia, Arkaute Agrifood Campus , P.O. Box 46, E-01080 Vitoria-Gasteiz, Spain
Contact: scdelatorre@neiker.eus

INTRODUCTION

Reuse of biomass-derived wastes is of paramount importance to the current concept of *circular economy*. After oil recovery from many oleaginous biomasses, a protein-rich by-product is obtained, which is potentially usefull as nutrient in microorganisms´s culture processes.

In this work, hydrolysates obtained from defatted rapeseed and *Schizochytrium* microalga were preliminarily tested as nitrogen source for heterotrophic growth of three microalgae species.

METHODOLOGY

Flours derived from defatted alga *Schizochytrium* and compressed rapeseed were subjected to enzymatic protein hydrolysis to obtain the algal hydrolysate (**AH)** and the rapeseed hydrolysate (**RH)**, respectively (composition in **Table 1**).

Each hydrolysate was added to the corresponding media to provide the same final concentration of total nitrogen (g/L) (**Table 2**). In the same way, yeast extract was used as control. Three media were tested for the growth of the microalgae *Chlorella vulgaris, Chlorella protothecoides and Schizochytrium limacinum.* Two growth experiments were conducted: A). in 100 ml erlenmeyers (orbital shaker, 140 rpm) and B). 5-L Fermentors (250 rpm, ≥25%pO_2, pH6), at 25ºC and under dark conditions.

Table 1. Hydrolysates chemical composition.

Composition	Hydrolysate	
	AH	RH
Total N (%)	0.99	0.89
Free Amino N (%)	5.50	5.44
Protein (%)	0.7	5.56
Carbon (%)	12.60	6.68
Phosphorus (%)	0.23	0.17
Potassium (%)	0.11	0.98

Table 2. Culture media composition.

Composition	100 ml Erlenmeyer	5 L Fermentor
Nitrogen (g/L)*		
Total N	0.2	0.3
Free amino N	1.11	1.8
Glucose (g/L)	10	30-40
Sea salts (g/L)	10	5

* N is provided by hydrolysates AH and RH or by Yeast Extract

RESULTS

A. Experiments in 100 ml Erlenmeyers

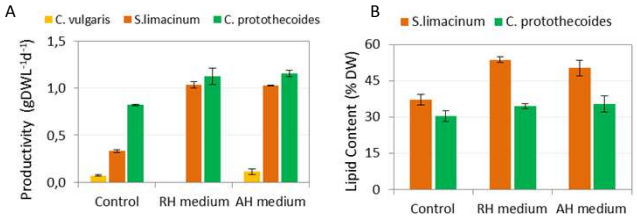

Figure 1. Biomass productivity and lipid content in a 4-days culture experiment. (A). Biomass productivity was enhanced by using hydrolysates compared with yeast extract (control). (B). Lipid content in biomass was also enhaced, specially for the DHA-producing microalga *S.limacinum.*

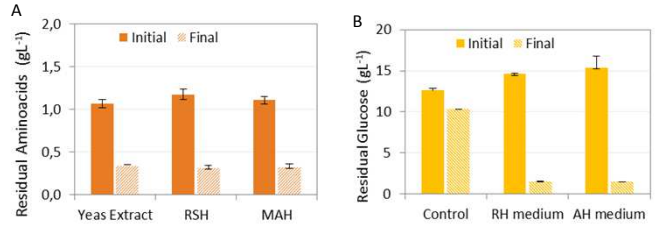

Figure 2. Residual nutrient concentration in the media at the beginning and at the end of the experiment with *Shizochytrium*. (A) More than 70% of supplied aminoacids were consumed in the three treatments. (B) An increased demand for glucose occurred in cultures supplemented with hydrolysates compared with yeast extract medium. This is consistent with the higher lipid content that showed the biomass obtained in cultures grown with hydrolysates (Fig. 1B). It is also noted that the hydrolysates provide extra glucose compared with yeast extract.

B. Experiments in 5-L Fermentors

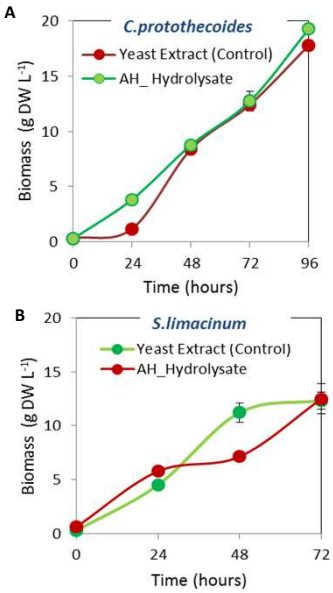

Figure 3. Growth of *C. protothecoides* and *S.limacinumi* in 5-L fermentors using algal hydrolysate and Yeast Extract as nitrogen sources.

Both compounds resulted equally suitable for microalgae cultivation at this scale. The biomass reached up to 18 g DW L^{-1} in a 4-day process (A), and 12 g DWL^{-1} in a 3-day process (B) resulting in a biomass productivity of 4.5 and 3.9 g $DWL^{-1}d^{-1}$ for *C. protothecoides* and *S. limacinun*, respectively.

C. Process Global vision and Benefits

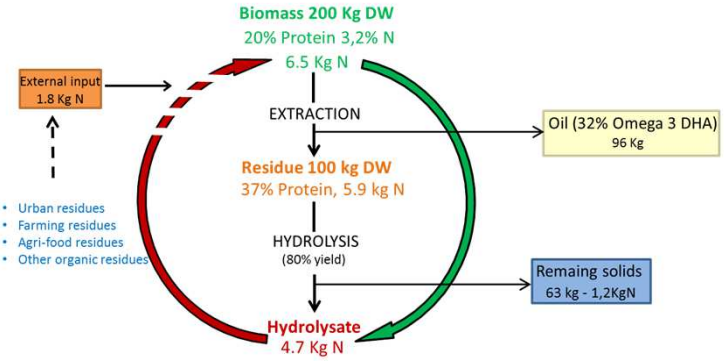

Figure 4. The scheme shows the recycling of the nitrogen contained in the defatted residue from *S.limacinum* into a new biomass through the use of a protein hydrolysate.

For *S.limacinum*, the proposed approach leads to a circular process where close to 80% of nitrogen contained in the residue can be turned into a high-value biomass (rich in Omega 3 nutritional oil). In this way, it can be attained up to 45% of reduction in the cost of the culture medium, and up to 32% of reduction in residues generation, thus providing enhanced environmental and economic features to the process. Other organic wastes could provide the supplementary nitrogen required for the culturing process, as it has been previously demonstrated (Pleissner et al.,2013)[1].

CONCLUSIONS

Hydrolysates obtained from deffated rapeseed and *Schizochytrium* biomasses resulted to be as suitable as yeast extract when used as nitrogen source for microalgae production.

This approach allows the revalorization of residues generated during the oil obtaining process, contributing to the bio-based circular economy. Specifically, for the DHA-producing microalga *Schizochytrium*, it is reached a circular flow when the nitrogen contained in residues is recirculated into a new valuable biomass through the culture process. This not only leads to the reduction of waste, but also to a decrease of the culture cost, being advantageous for the economy of the process.

1. Food waste as nutrient source in heterotrophic microalgae cultivation. Biores Techol 2013, 137:139-146.

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