## Bio-valorization of olive mill wastewaters and crude glycerol blends with the use of a *Yarrowia lipolytica* strain to produce citric acid, polyols and lipids

Dimitris Sarris, Zoe Sampani, Seraphim Papanikolaou Department of Food Science and Human Nutrition (Agricultural University of Athens, Greece

## Abstract

In the present study, crude glycerol, waste discharged from bio-diesel production, was used as substrate for screening eleven natural Yarrowia lipolytica strains when grown in nitrogen-limited submerged shake-flask experiments. In media with initial glycerol concentration of 40 g/L, all strains presented satisfactory microbial growth and almost complete glycerol uptake. The principal metabolic product was citric acid (Cit<sub>max</sub>~30.0 g/L, yield 0.30–0.80 g per g of glycerol consumed) simultaneous with the accumulation of storage lipid. Polyols mannitol, arabitol and erythritol were also synthesized as strain dependent compounds. Y. lipolytica strain ACA-YC 5031 produced sufficient amount of citric acid and presented the highest (amongst the screened strains) production of the polyol erythritol (~4.0 g/L, Y<sub>Ery/Glol</sub>~0.18 g/g) as also non negligible amounts of the polyols mannitol (~5.6 g/L, Y<sub>Man/Glol</sub>~0.25 g/g) and arabitol (~2.2 g/L, Y<sub>Ara/Glol</sub>~0.15 g/g). This strain was further grown on increased concentration of crude glycerol nitrogenlimited experiments of ~70 g/L (blank experiment) and on blends of crude glycerol and olive mill wastewaters (OMW). Specific volume of OMW was used with the rationale to partially substitute process tap water, thus giving initial phenolic compounds concentration of 1.0 g/L. The metabolism seemed to be shifted towards citric acid production at expense of erythritol production, due to the addition of OMW into the medium. The maximum production of biomass ( $X_{max}=10.2 \text{ g/L}$ ,  $Y_{X/Glol}=0.14 \text{ g/g}$ ) as also the accumulation of both citric acid (Cit<sub>max</sub>=37.1 g/L, Y<sub>Cit/Glol</sub>=0.51 g/g) and cellular lipids (L<sub>max</sub>=2.8 g/L,  $Y_{L/X}=0.25$  g per g of dry cell weight) was favored by OMW addition, compared to blank experiment. On the other hand erythritol production presented significantly lower values with OMW addition (Ery<sub>max</sub>=7.4 g/L,  $Y_{Erv/Glol}=0.11$  g/g) whereas mannitol and arabitol production showed no significant difference (Man<sub>max</sub>=11.6 g/L, Y<sub>Man/Glol</sub>=0.16 g/g; Ara<sub>max</sub>=2.6 g/L, Y<sub>Ara/Glol</sub>=0.04 g/g) compared to blank experiment. Finally, removal of medium color occured (up to  $\sim 20\%$ ).

Keywords—Citric acid, crude glycerol, olive mill wastewaters, polyols, Yarrowia lipolytica