SELECTION AND CHARACTERIZATION OF A STRAIN OF *Kluyveromyces marxianus* CAPABLE OF FERMENTING LACTOSE

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Introduction. It is currently recognized that only 2% of the yeasts species are capable of fermenting lactose (1). Strains of *Kluyveromyces marxianus* have been considered the most adequate for bioconvertion of lactose into biomass, ethanol or β.galactosidase (2). *K. marxianus* is a yeast with useful physiological features: it is thermotolerant, it usually has a high growth rate, it adapts to many different substrates and many strains have been already recognized as GRASS (Generally Recognized As Safe (3).

The aim of this work was to test several strains of yeast *K. marxianus* isolated from *Agave durangensis* must, in order to choose a strain capable to use lactose as carbon source.

Methods. Thirty seven isolates of *K. marxianus*, from the yeast collection of the Instituto Tecnológico de Durango (ITD), were grown in a medium with lactose as the sole carbon source. The strains selected as Lac^+ (lactose-fermenting) were tested on their ability to consume lactose and produce ethanol on LPY media (20 g/L lactose, 20 g/L peptone and, 10 g/L yeast extract) incubated during 24 h, at 28ºC and pH 4.8. The chosen yeast was subjected to a physiological study using an inoculum of $1x10^6$ cells/mL in LPY* media (20, 40 and 80 g/L lactose, 5 g/L peptone and, 5 g/L yeast extract) incubated at 28 and 37ºC and pH 4.8, under anaerobic conditions. Lactose and ethanol concentrations were assessed by HPLC.

Results. The *K. marxianus* strains ITD00089, ITD00090 and ITD00262 were capable of fermenting lactose but *K. marxianus* ITD00262 was selected for its ability to ferment lactose and produce ethanol as shown in Table 1.

Table 1 Strains of yeast *K. marxianus* selected as Lac^+.

K. marxianus	Lactose consumption (g/L)	Ethanol production (g/L)
ITD00089	4.84	3.127
ITD00090	5.126	2.469
ITD00262	9.066	4.254

The results of the physiological study are summarized in Table 2.

The residual lactose increases when the initial concentration of this is higher. Increasing the temperature doesn't have any effect in the residual sugars in both temperatures.

In other works using cheese whey as culture media have seen that additioning nitrogen ($(NH4)2SO4$, yeast extract and MgSO4) and shaking decreases the residual sugars further (4). Moreover the ethanol production was increased when the concentration of lactose and temperature was higher, however is not statistically significant.

Table 2 Lactose fermentation with the *strain* ITD00262 at 28 y 37ºC with 20, 40 y 80g/L of lactose.

Lactose 20g/L (72h)	**28ºC**	**37ºC**
Residual lactose (**g/L**)	0.53 ± 0.08^b	0.25 ± 0.01^a
Ethanol production (**g/L**)	9.14 ± 0.55^a	8.68 ± 1.15^a
Lactose 40g/L (96h)	**28ºC**	**37ºC**
Residual lactose (**g/L**)	13.17 ± 1.7^a	13.86 ± 0.057^a
Ethanol production (**g/L**)	12.35 ± 0.18^a	14.15 ± 1.42^a
Lactose 80g/L (96h)	**28ºC**	**37ºC**
Residual lactose (**g/L**)	50.63 ± 0.99^a	53.16 ± 5.93^a
Ethanol production (**g/L**)	12.93 ± 0.05^a	13.673 ± 1.95^a

Conclusions. Strains isolated from a source different from dairy environments were able to use lactose as carbon source. Fermentation requires over 96 hours consuming all sugars, but the selected strain can be a good candidate to produce ethanol from lactose.

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