



Maestría en Ingeniería en Diseño de Bioprocesos

Title

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## 1. Introduction

The interest for natural products with added value in the industry has been increased in the recent years. The lutein and the myristic acid are examples of this.

Lutein is a antioxidant potent which have high demand in human health and wide application in the food industry like additive. On the other hand, the products of hydrolysis of coconut oil like myristic acid have wide application in the industry cosmetic, food and pharmacological. These products are obtained by saponification that require after neutralized with mineral acid.

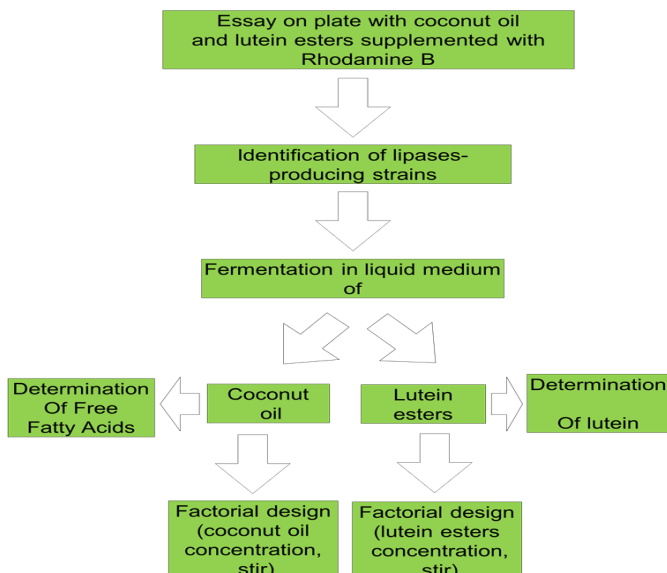
Hence, the interest in research the lipase-producing microorganisms with high catalytic activity due that these present advantage over chemical hydrolysis.

## 2. Aim

To obtain lutein and myristic acid from lutein esters of cempasúchil flowers and coconut oil, respectively by the application of microbial lipases.

- To identify the strains which produce lipases.
- To determine the lipolytic activity of the strains by qualitative essays on plate with Rhodamine B.
- To determine the ideal conditions to obtain lutein and myristic acid by the application of a factorial design (strain, substrate concentration and stir)
- To extract, purify and identify the lutein and myristic acid, as well the subproducts by qualitative, quantitative chemical analysis and spectroscopic analyse (RMN)

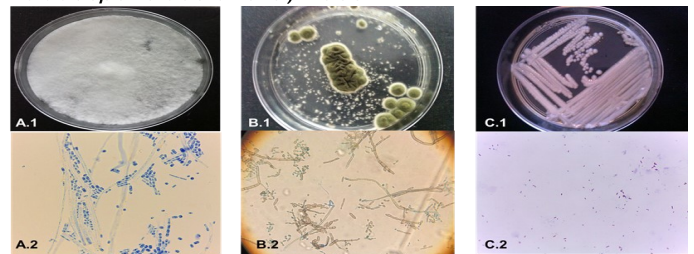
## 3. Method



## 4. Results

### ➤ Identification by morphology

**Figure 1.** Show the macroscopic and microscopic morphology of A) *Geotrichum candidum*, B) *Cladosporium cladosporioides* and C) LC001.



### ➤ Identification by phylogenetic analysis

*G. Candidum* and *C. cladosporioides* were confirmed by the comparison between genetic sequences in BLASTn of NCBI with 99.9% of identity.

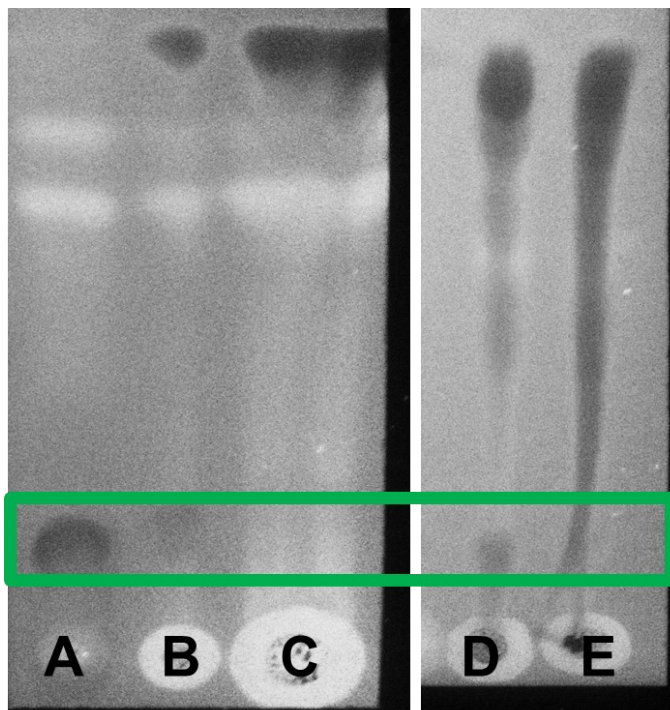
### ➤ Screening of lipase-producing strains by essay on plate supplemented with Rhodamine B

**Table 1.** Screening of lipases-producing strains by essay on plate supplemented with Rhodamine B using coconut oil and lutein esters like lipolytic substrates.

| Strain                   | Lutein esters | Coconut oil |
|--------------------------|---------------|-------------|
| LC001                    |               |             |
| <i>C.cladosporioides</i> |               |             |
| <i>G. candidum</i>       |               |             |

## ➤ TLC of hydrolysis products of lutein esters

**Figure 2.** A) lutein obtained by saponification, B) Hydrolysis products by LC001, C) Lutein esters, D) Hydrolysis products by *C. cladosporioides* and E) Hydrolysis products by *G. candidum*.



## ➤ Hydrolysis percent of coconut oil

**Table 2.** Hydrolysis percent of coconut oil using LC001, *C. cladosporioides* and *G. candidum*

| Strain                    | ml of NaOH | % of hydrolysis |
|---------------------------|------------|-----------------|
| LC001                     | 15.3       | 76.5            |
| <i>C. cladosporioides</i> | 10.6       | 53              |
| <i>G. candidum</i>        | 11.6       | 58              |

## 5. Conclusion

The identification of two strains were confirmed by phylogenetic analysis using a segment of 18S of rRNA.

The lipolytic activity quantified of the hydrolysis of coconut oil by fluorescence showed a similar activity in *G. candidum* and *C. cladosporioides* cultures, meanwhile LC001 had higher lipolytic activity..

However, in the case of the hydrolysis of lutein esters showed statistically significant differences between the activities of LC001, *G. candidum* and *C. cladosporioides*, being LC001 the strain with higher lipolytic activity.

The LC001 strain will be used to factorial design because showed higher lipolytic activity in both substrates in comparison to the other strains.

## Acknowledgements

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