



Cleaner and sustainable synthesis of high-quality monoglycerides by use of enzyme technologies: techno-economic and environmental study for monolaurin

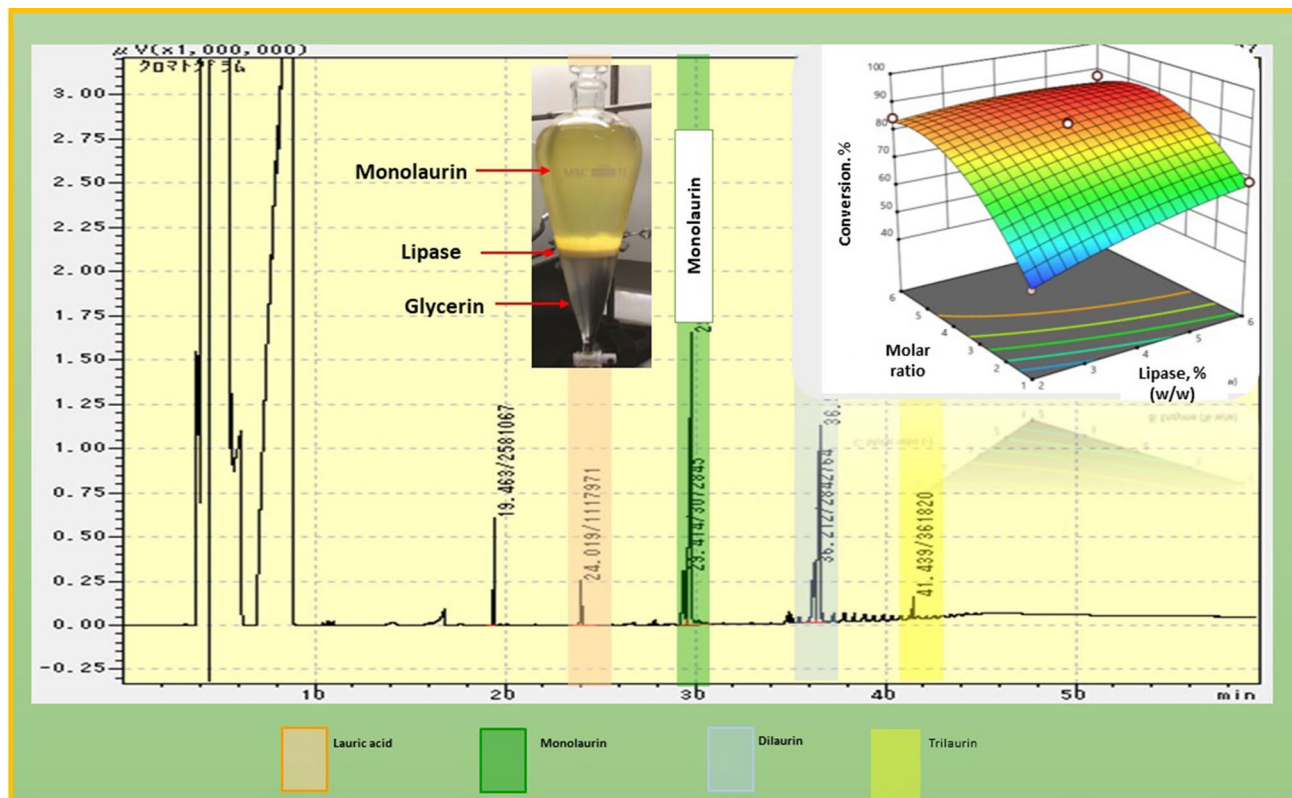
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Abstract

Currently, monoglycerides (MG) are produced using a complicated energy-intensive technology that contributes negatively toward greenhouse gas mitigation. This work suggests a cleaner and simpler one-step enzymatic production of α -monolaurin in an inert membrane reactor, where the reaction and enzyme separation are conducted simultaneously in one unit. *Candida antarctica* lipase (Lipozyme 435) was used to catalyze the esterification reaction between lauric acid and glycerin in a solvent-free system under mild temperatures. Response surface methodology was used to optimize the reaction conditions. The optimal conditions were a molecular sieve of 14.85% w/w, a temperature of 56.95 °C, an enzyme amount of 5.38% w/w, and a molar ratio of 4.75% w/w. The gas chromatography (GC) analysis showed that the α -monolaurin percentage was 49.5% when the enzymatic process (ENZ) was used. The conventional chemical (CHEM) and autocatalytic (AUT) esterification methods were also performed to study their proportional MG yields. The GC results showed the MG percentages of 43.9 and 41.7% for CHEM and AUT, respectively. Economic analysis was also conducted for the suggested enzymatic technique, and the findings were compared with those of the CHEM and AUT technologies. Using a plant capacity of 4950 t/year and 11% interest for the proposed ENZ process, the total capital investment of α -monolaurin production was preferably four times less than that of the CHEM process and three times less than that of the AUT method, presenting investment possibilities. However, the ENZ process showed the least profitability (net profit per day) among the three processes. Nevertheless, the return on investment and net present value for the ENZ process were preferably higher than those of CHEM and AUT because of its interestingly lower inside battery limit plant cost and less energy consumption. The AUT/CHEM processes generated a total carbon dioxide (CO₂) exhaust of 678.7 t CO₂ eq./year. In contrast, the ENZ process exhausted a total CO₂ of only 50 t CO₂ eq./year. The present integrated techno-economic and environmental study of α -monolaurin production emphasizes the green and cost benefits of the proposed ENZ technology.

Graphical Abstract



Keywords α -Monolaurin · Sustainable development goals (SDGs) · Climate change · Esterification · Lipozyme 435 · Cleaner production · Economic evaluation · Environment · Energy consumption

Abbreviations

FFA	Free fatty acids
AUT	Autocatalytic process
ENZ	Enzymatic process
CHEM	Chemical process
MG	Monoglycerides
ISBL	Inside battery limit
CCOP	Cash cost of production
ROI	Return on investment
OSBL	Outside battery limit
NPV	Net present value

Introduction

In the last decade, monoglycerides and their derivatives shared 75% of the global emulsifiers market (Mustafa et al. 2022). They find many applications as emulsifiers such as in food products, pharmaceutical formulations, cosmetics, and animal feed additives (Abdelmoez and Mustafa 2014). α -Monolaurin is the ester of lauric acid and glycerol.

Because of its great antimicrobial effect, α -monolaurin has recently gained much attention as an immune stimulant (Abd El Fadeel et al. 2022). It can be manufactured through the esterification process that takes place between glycerol and lauric acid (Rarokar et al. 2017). This reaction can be performed using biocatalysts (enzymatic technology [ENZ]), without any catalyst (autocatalytic esterification [AUT]), or using chemical catalysts (chemical technology [CHEM]) (Abdelmoez and Mustafa 2014). The key obstacle in the synthesis of monoglycerides is the separation of contaminants due to free glycerol and various concentrations of diglycerides and triglycerides ordinarily present with monoglycerides (Lozano et al. 2019). Nevertheless, to meet actual demands, greatly condensed perfect monoglycerides are ordinarily needed. Therefore, the application of short-path distillation to obtain a product with a high order of monoglycerides is necessary (Abdelmoez et al. 2013; Ashby et al. 2023).

CHEM-catalyzed processes basically use acid catalysts. Strong mineral acids such as sulfuric and hydrochloric acids are generally used to catalyze such reactions. In

addition, Lewis acids such as zinc and tin salts, organotinates, aluminum halides, and boron trifluoride are also utilized (Jegannathan et al. 2011). Likewise, heterogeneous catalysts, including cation-exchange resins, tin catalysts, and zeolites, are also used. In comparison, autocatalytic processes are performed without using catalysts as their elevated temperatures support the conversion of fatty acids (Ibrahim and Mustafa 2022). High temperatures of 190–220 °C are utilized in both technologies (Abd Maurad et al. 2018).

However, aside from the high energy consumption of both the above methods, their high temperatures lead to random reactions and darken the final product's color (Chen et al. 2022b; Mustafa and Niikura 2022). Furthermore, these methods produce high amounts of diglycerides and triglycerides rather than monoglycerides (He et al. 2022). Such approaches can result in low product yields and nonspecific ways, which require further processes such as distillation. However, conventional distillation is not possible because of the low vapor pressure of glycerides (Chen et al. 2022a; Fregolente et al. 2010).

Currently, industrial short-path distillation is required for monoglyceride synthesis to acquire a monoglyceride yield of more than 90% (Chen et al. 2022c; Xu 2019). Such technology requires additional molecular distillation, intensive energy, and a great vacuum, resulting in high capital costs (Hosney and Mustafa 2020). Moreover, both distillation and reaction processes yield many effluents and are energy intensive, causing ecological disadvantages (Hosney et al. 2020a). It should be highlighted that despite its great energy demand, the chemical-catalyzed reaction is the current commercial α -monolaurin production approach. This is mainly because it is considered economically feasible owing to the catalysts' low cost (Hosney et al. 2020b).

Meanwhile, the production of α -monolaurin as a feed-additive utilizing enzyme points to cleaner production and appears more attractive. The lipases' specificity can synthesize tailored outputs with improved quality (Ching-Velasquez et al. 2020). Being a one-reaction process without the need for the expensive distillation process, this method might persuade many investors and startups to invest in such clean technology (Mustafa 2021).

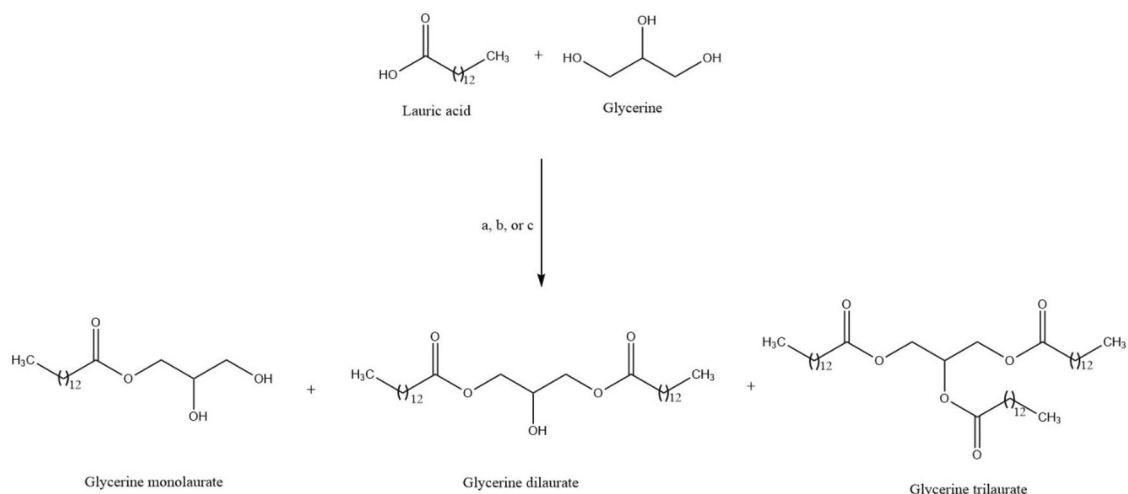
In the context of ester production using membrane technology, various process configurations have been developed in the literature, including the following: (1) an esterification reactor followed by a heterogeneous catalyst membrane separating module in different units and (2) both the reactor and the membrane module represent one unit, where both reaction and catalyst removal are performed in only one unit. The latter approach has been receiving much attention because of its reduced capital cost (Leite et al. 2022; Nawaz et al. 2022). Such an approach can be classified into two types: first, the membrane is used only for separation and has

no catalyst in its structure (inert membrane reactor [IMR]); second, the membrane contains an active catalyzing material in its structure. In our study, both reaction and lipase separation have been conducted simultaneously in one unit using IMR, suggesting less capital investment.

It was found that, the α -monolaurin yield was high enough and suitable for use as an animal feed additive after mixing it with a cellulose carrier. Compared with the conventional α -monolaurin production process, distillation must be applied after esterification to produce suitable animal feed-additive products. These merits imply a lipase-catalyzed process that is much more economically feasible than those of AUT- and CHEM-based plants from an investment perspective. For instance, the land area required for the investment in the ENZ process and the number of equipment pieces are much less. Furthermore, the higher product yields and lower energy demands indicate the viability of this process to compete with the AUT/CHEM methods. In this respect, the return on investment (ROI), capital cost, and manufacturing calculation are crucial indicators that can give a trustworthy view of investment viability, and such estimations determine whether a suggested approach is feasible for execution (Jambulingam et al. 2020; Lee et al. 2020).

Environmental impact has also been increasingly considered to be a primary element in maximizing existing processes or designing new ones (Munir et al. 2022). Moreover, governments have placed numerous legislation and regulations to control carbon exhaust emissions mainly for greenhouse gas mitigation and alignment with the United Nations' sustainable development goals (SDGs) (Buturca et al. 2013; Rekhletskaia et al. 2022). Apart from the alignment with the SDGs, the calculations of carbon footprint can also offer information for top management and decision-makers to evaluate processes' energy consumption before an investment decision is established. There have been many published papers on monoglycerides synthesis. However, only a few of them considered the economic validity of this process. Therefore, an integrated techno-economic investigation is needed as it can help plant decision-makers and manufacturers expect and decide on forthcoming investment possibilities. Furthermore, it can assist the research community with a broad overview of suggested strategies and define challenges and chances.

The main goal of this assessment is to ultimately decide whether or not to proceed with a particular manufacturing technique as an investment. Additionally, ROI and net present value (NPV), two well-known metrics for economic analysis, were assessed. These two variables were included because their values can indicate if a method is feasible and profitable. The study has been also projected for over 15 years, including the years leading up to and just after the year significant for sustainable development (2030). The SDGs place a strong focus on the idea that by 2030,



Scheme 1 Esterification of lauric acid and glycerine by three different technologies: enzymatic, autocatalytic and chemical. **a** Conditions (enzymatic process): molar ratio of 6:1 glycerine to lauric acid, temperature of 60 °C, reaction time of 6 h, Lipozyme 435 amount of 6% w/w, molecular sieves of 10% w/w, and agitation speed of 150 rpm. **b** Conditions (Autocatalytic process): molar ratio of 1:1 glycerine to

lauric acid, temperature of 190 °C, mechanical agitation speed of 80 rpm, and reaction time of 6 h. **c** Conditions (Chemical process): molar ratio of 1:1 glycerine to lauric acid, temperature of 190 °C, reaction time of 6 h, TIB160 of 0.06% w/w, and mechanical agitation speed of 80 rpm

everyone should have access to dependable, affordable, and cutting-edge energy services (Ershov et al. 2022a, b; Hák et al. 2016). Thus, an integrated techno-economic and environmental analysis for the synthesis of α -monolaurin using ENZ catalysis was conducted in this study. The results were compared with those of conventional production routes (AUT and CHEM). The manufacturing and capital costs, as well as the ROI for all production methods, were estimated. Finally, sensitivity analysis was carried out to identify the controlling parameter affecting the process feasibility. As far as is known, the proposed study is the first comprehensive comparison of the ENZ, CHEM, and AUT manufacturing methods for monolaurin synthesis from technical, economic, and environmental standpoints.

Methodology

Materials

Lauric acid 99% and glycerol with purity of 99.7% were provided by Oleo Misr for Oleochemicals Company (Sadat City-Egypt). Furthermore, immobilized lipase, Lipozyme 435, (triacylglycerol hydrolase, EC 3.1.1.3), *Candida antarctica* lipase, 10,000 PLU/g, supported on a macroporous acrylic resin was granted by Novozymes A/S (Denmark-Copenhagen). Moreover, beads 8–12 mesh, 4 Å, molecular sieves, as well as GC standards (Lauric acid, 1 lauroyl rac glycerol, and glyceryl tridodecanoate) were provided from

Sigma-Aldrich (St. Louis, MO, USA). TIB Kat 160, and TIB Tinex S were gifted by TIB Chemicals AG (Mannheim, Germany). All other chemicals were of analytical grade and utilized as supplied.

Production of α -monolaurin

TIB 160 was used as a heterogeneous catalyst in the CHEM process, Lipozyme 435 was utilized as the biocatalyst in the ENZ method, and no catalyst was utilized in the AUT process. The suitable reaction time for the three strategies was studied and was found to be 6 h. NaOH was used as a titrant for the calculation of the concentration of ester formation due to fatty acid depletion. After the reaction time elapsed, 200- μ l aliquots were withdrawn for analysis. Ethanol/acetone blends of 10 ml (50/50, %, v/v) were mixed with the 200- μ l aliquots. Thereafter, the conversion content was calculated based on the unreacted lauric acid using Eq. (1):

$$\text{Conversion to ester (\%)} = \frac{N - N^0}{N} \times 100, \quad (1)$$

where N is the amount of NaOH added along with the lipase. Meanwhile, N^0 represents the amount of NaOH used in the samples without lipase addition.

It must also be noted that the conversions obtained using the titration method were in excellent agreement with those obtained using gas chromatography (GC). All analyses were performed in triplicate in the current study, and the average conversion reflected the mean of the three values. The proposed synthesis processes are represented in Scheme 1.

Enzymatic production of α -monolaurin

Lauric acid and glycerol were reacted in a 100-ml stoppered batch reactor in the presence of lipase. The reaction conditions were optimized using the Box–Behnken design. Lipozyme 435 was used in various quantities from 2 to 6% (w/w). In addition, the temperature effect was investigated from 50 to 60 °C. Likewise, the molar ratio of glycerin to lauric acid was studied from 8:1 to 1:1. Molecular sieves were supplied to remove the formed moisture within the advance of the reaction, which ranged between 5 and 15% (w/w).

Scale-up of monolaurin by the ENZ method Lauric acid and glycerol were introduced to a 1000-ml four-necked batch reactor. Reaction heating was performed using a hot water bath at a controlled temperature. Agitation was conducted at a low magnetic stirring speed-assisted nitrogen purging to avoid enzyme denaturation. Molecular sieves were used to remove the generated moisture within the enzymatic reaction's advance, and Lipozyme 435 was used as a biocatalyst. An inert membrane of nonwoven polypropylene fabric was placed at the outlet of the reaction medium to separate enzymes and molecular sieves during reactor discharge (after reaction completion). After emptying all the reactor content, the new reactants were fed using the same outlet connection but in the opposite of the existing direction to suspend the accumulated enzyme and molecular sieves on the membrane. A permeate was introduced to a separating funnel to separate phases. The heavy glycerin phase was removed from the bottom after allowing adequate settling time so it could be used in future esterification processes. The glycerin laurate phase separated to the top as it was the lighter phase. This product was further blended with a carrier and then supplied to the feed processing facilities to be mixed with the main animal feed ingredients.

Synthesis of α -monolaurin using the autocatalytic method

A 2-L stirred-tank reactor (four-necked round flask) was used to conduct the esterification reaction between glycerin

and lauric acid. The reaction temperature was set to 190 °C using a heating mantle. A molar ratio of 1:1 and a reaction time of 6 h were applied. Additionally, the speed of mechanical stirring was adjusted to 80 rpm. Moreover, nitrogen blanket purging was applied to continuously remove water vapor. A condenser was placed at the top of the reactor to condense the water vapor. After the reaction time, the reactor's content was sent to a decanter to separate the excess glycerin in the bottom phase. Both titration and GC analysis were performed periodically (every 1 h) to examine the conversion and monoglyceride formation.

Chemical synthesis of α -monolaurin

The chemically catalyzed esterification reaction was conducted similarly to the method explained in the AUT process except for the addition of the heterogenous catalyst TIB Kat 160 with an amount of 0.06% (w/w). The reaction time was set to 6 h. After the completion of the reaction time and to remove the catalyst, Tinex S with a concentration of 0.1% (w/w) was added to the reaction mixture to mix with TIB160 and form solids that can be separated through filtration. As per the catalyst manufacturer, TIB Tinex S is particularly suitable for removing inorganic tin catalysts from organic esterification products. In addition, the bleaching ability of TIB Tinex S allows for the decoloration of the product. After 10 min of stirring time, filtration was performed to remove catalyst' residuals. Glycerin laurate was separated from glycerin through simple settling, as performed for the AUT method.

Product characterization by GC analysis

The gas chromatograph, GC (SHIMADZU GC-2025), has been used for the analysis of the reaction products equipped with automated injector (AOC-20i). The chromatographic separation was achieved with a non-polar column, Column, DB-1HT (non-polar, ID 0.25 mm, 30 m, and thickness of film is 0.1 μ m). The carrier gas used was helium at a flow rate of 7.9 mL/min. The oven was programmed at 50 °C, held for 15 min, then a temperature ramp at 10 °C/min to a final temperature of 395 °C, being then held for 15 min. The injector and detector temperatures were set at 400 and 380 °C, respectively. The samples' derivatization was carried out using the silylation protocol (Smith et al. 1968). Samples of mono-, di-, and triglycerides were dissolved in (20 mg/mL) of pyridine and allowed portions of 1000 μ L to react with 500 μ L N,O-bis(trimethylsilyl)trifluoroacetamide for 30 min at 80 °C. A split ratio of 1:10 was used to inject 1 μ L of the obtained solution onto the GC column.

Table 1 The level of independent variables used in the Box–Behnken design model

Independent variables	Factor levels		
	– 1	0	1
A: Temperature (°C)	50	55	60
B: Enzyme amount (%w/w)	2	4	6
C: Molar ratio (-)	1	3,5	6
D: Molecular sieve (%w/w)	5	10	15

Design of experiment

The experimental series are generated by Box–Behnken design being a response surface method. The Box–Behnken design is used to create higher-order response surfaces using fewer runs than three-level factorials (Rao and Kumar 2012) and can optimize the effect of three or more independent variables. For the present study, temperature, amount of enzyme, glycerol-to-lauric acid molar ratio and molecular sieve were chosen as four independent variables whereas the conversion selected as response.

The result of a four factor-three levels Box–Behnken design with five replicates at the center point was conducted twenty-nine trials in total. The level of variables chosen for generating the design matrix is shown in Table 1. The statistical and correlation analysis of the response of the model was conducted by analysis of variance (ANOVA). The effect of four variables was examined for the response, and response surfaces were used for optimization of the process. Experimental data were fitted to a second-order quadratic model written as Eq. 2

$$Y_k = \beta_{k0} + \sum_{i=1}^3 \beta_{ki}x_i + \sum_{i=1}^3 \beta_{kii}x_i^2 + \sum_{i<j=2}^3 \beta_{kij}x_ix_j \quad (2)$$

where Y_k is the response function, x_i and x_j are the independent variables; β_{k0} , β_{ki} , β_{kii} , and β_{kij} are the constant regression coefficients for intercept, linear, quadratic and interaction model terms, respectively. A confidence level of 95% was employed to provide the convenience of regression model. The statistical software package (Design expert 13.0-Trial Version; State Ease, USA) was used for experimental design, ANOVA, convenient equation model, 3D surface plot, and optimization.

Economic evaluation and model analysis

The principal target of the economic contributions from all authors was to determine the cost of operation and execution as well as the income sources. Likewise, similar studies determined investment feasibilities by studying cash flows. In this paper, the authors targeted to propose information-aided decision-making to the oleochemical community for selecting the type of investment at the decision stage. A comparative techno-economic investigation would offer detailed economic-related details on the three proposed processes (ENZ vs. CHEM vs. AUT).

Total capital investment

The total capital investment is the sum of the required working capital, starting expense, and fixed capital investment (Sinnott and Towler 2019). The fixed capital investment cost

is a major indicator concerning a factory's total cost, installation, construction, and design. Therefore, accurate estimations of such costs are crucial in avoiding wrong budgeting and operations (Thoppil and Zein 2021). By adding the expenses of inside battery limits (ISBLs), outside battery limits (OSBLs), engineering, and contingencies, the current cost may be calculated.

Thus, the calculated ISBL can significantly affect the overall process design financing. Therefore, inaccurate calculations should be avoided. Furthermore, the objective of ISBL must be properly determined with utmost care. This involves valves, piping, equipment, instruments, and other connected additions required to begin synthesis. The ISBL cost can be calculated using many techniques based on the synthesis capacity and kind of industry. The standard calculation methods include those of Gore, Stallworthy, Bridgewater, Taylor, Fromme, Klumpar, and Brown (Tsagkari et al. 2016). Bridgewater's method was used in this study for the ISBL calculations. In addition, the authors of this research received a quote from a Malaysian engineering company for a chemically catalyzed esterification plant for the same studied capacity. The two ISBL costs were relatively close to each other, which emphasizes that the present calculations are free of deviations and errors.

Currently, no engineering companies supply mono-glycerides using ENZ technology. Consequently, the ENZ plant ISBL cost was calculated directly using Bridgewater's method. In this method, three parameters should be estimated with high accuracy to obtain precise outputs. This involves plant capacity, reactor conversion, and the number of main units. The capacity of the plants was set to 4950 t/year, identical to the capacity of the quoted CHEM factory. Additionally, experimental lab data were used to calculate the conversion. The cost of ISBL was then determined using Bridgewater's technique (Eq. (3)).

$$C = 280,000N \left(\frac{Q}{s} \right)^{0.3} \quad (3)$$

N stands for the number of main units, C for the ISBL capital cost in pounds, Q for the plant capacity in tons per year, and S for the reactor conversion.

Meanwhile, the cost of off-site alterations and improvements needed for the plant's operation was included in the OSBL cost. Infrastructure improvements such as linking water and gas pipelines underground are part of these improvements. In this study, the OSBL cost was estimated as a percentage of ISBL, which might vary from 10 to 100% (Mustafa et al. 2022). An OSBL percentage of only 30% was considered in this study. This percentage selection was primarily because the first author of this work was aware of the execution site and readiness of the current site where the plant will be installed. An Egyptian oleochemical company

had a future expansion area dedicated to the execution of this plant. Such an area was previously supplied with all utilities, including water and natural gas, during the construction of the plant.

The engineering cost involves the expenses of an engineering package, including equipment lists, pipes and instrumentation drawings, constructive drawings, and an operation manual. In the current context, no additional costs were added for all methods (ENZ, CHEM, and AUT). It was considered that the research team of the company already possessed the complete engineering package for the required investment.

The contingency cost indicates the cost of uncertain expenses, and its minimum value is 10% of the direct capital cost. This cost is usually granted to considerably conventional technologies (CHEM/AUT). However, a 15% value was decided for the ENZ technology because the production of oleochemicals using enzymes is a technology still under research and may thus retain uncertainties.

Another economic item is the operating capital cost, which includes the cost of commissioning and plant operation. An operating capital parentage of 15% of the direct capital cost was considered in this study. Lastly, the expense of the startup was estimated as 10% of OSBL + ISBL (Thop-pil and Zein 2021).

Operating expenses

The fixed production costs and the variable production costs are the two basic factors that control operational expenses. Environmental fees, labor costs, land rent, property insurance, taxes, maintenance, and administrative overhead are all included in the fixed production costs (Sinnott and Towler 2019). Such costs are compulsory whether a study works at an optimal status or not. Moreover, the variable production costs are usually associated with the production rate and output. The current cost includes costs for utilities (such as cooling water, electricity, and steam for heating), raw materials (such as glycerin, lauric acid, chemical catalyst, and lipase), and any other consumables, packing, products, shipping costs, and disposal costs for waste streams such as used molecular sieves, lipases, and heterogeneous catalysts. Thus, the optimal use of resources, including the prolonged use of enzymes, reduced raw materials losses, and energy saving, can guarantee a decrease in the variable cost. Up-to-date prices for all raw materials were directly quoted by Novozymes A/S for lipase, TIB chemicals AG for TIB166 catalyst, and Chinese and Malaysian companies for lauric acid and glycerin.

The mass and energy balance calculations were performed under the following assumptions:

- The plant capacity was 15 t/day of α -monolaurin, which was equal to 4950 t/year.
- The CHEM and AUT processes worked in a continuous operation and consisted of a deglycerination unit, a short-path distillation unit, and an esterification unit. The deglycerination unit sought to regenerate excess glycerol to be fed back to the principal reaction. The CHEM process differed from AUT in that it had a catalyst removal filtration system. Therefore, the CHEM process comprised four units, whereas the AUT process consisted of only three units.
- Short-path distillation was used in CHEM and AUT to separate monolaurin, and the resulting dilaurin and trilaurin were then returned to the esterification process, where they were mixed with glycerin and lauric acid.
- In the CHEM process, the catalyst TIB 166 was removed through filtration and was not used again (as per the catalyst manufacturer's information).
- Because of the delay in the off-loading of the batch after esterification, the ENZ technique operated in batch mode. By adding an esterification reactor, the recommended ENZ process can also be used in continuous mode. That is, when one reactor is in operation, the other is emptying its content for phase separation.
- The ENZ method-based plant consisted of a one-step unit, which was esterification. Although the process conversion did not exceed 91%, the remaining free lauric acid did not show a defect for the final product. This was because the target of the process was to use the product as a feed additive for animal feed applications. This particular application accepts the presence of free lauric acid at any percentage. However, it should be noted that the efficacy of monolaurin as an immune stimulant is much higher than that of lauric acid.
- Lipase usage was regarded as 1 kg of Lipozyme 435 for every 2 t of α -monolaurin synthesis. The current usage was based on a literature review on ester synthesis using Lipozyme 435 (Xu et al. 2012).
- Energy consumption was considered the same in the AUT and CHEM processes.
- Product losses due to downstream processes (deglycerination, short-path distillation, and catalyst removal through filtration) were 5%, 10%, and 15% w/w for ENZ, AUT, and CHEM, respectively. The losses in the ENZ process were determined from lab trials, whereas the losses in the AUT and CHEM were provided by experts from Oleo Misr for Oleochemicals Company.

Economic feasibility parameters

The revenues associated with the project is the income obtained from the sale of products for both the desired

product and any byproducts produced. In the present case, no credits for the byproducts generated by all processes were determined. In ENZ process, the free lauric acid was not separated from the final product by neutralization. This is mainly because that the targeted product application is animal feed additive. This application does not put constraints on the free fatty acid content, putting into account that the price of the product may be affected if the percentage of lauric acid exceeded 10%.

Gross margin is another parameter that participates to process's economic feasibility. The gross margin be calculated by subtracting the revenues from the sales of product from the raw materials cost. Therefore, the gross margin value plays a crucial role in offering high indication about the revenues retained from sales that do not relate to production costs. In the suggested research, the gross margin is remarkably impacted by the raw materials cost as the latter participate to more than 90% of the cash cost of production (CCOP).

To obtain finer details about a research's economic feasibility, profit should be calculated. Here the profit value depends significantly on the CCOP. CCOP involves the sum of the entire variable synthesis cost and the entire fixed synthesis cost. After then, the profit is calculated by deducting the CCOP from the -monolaurin revenues. The estimated profit is still a gross profit. The corporate tax is taken from the gross profit to estimate the net profit—the corporate tax rate changes based on the country and the year the research is carried out. In Egypt for instance, in 2022, the corporation tax rate will be 22.5%. Equation (4) can be used to compute the amount of tax paid.

$$\text{The amount of tax paid} = \text{the tax rate} \times \text{the taxable income} \quad (4)$$

The taxable income can be determined by subtracting the tax allowance from the gross profit. The depreciation cost is a typical example of a tax allowance.

The declining balance depreciation strategy can calculate the depreciation charges where the cash flow would be controlled. In the current study, a depreciation rate of 10% was anticipated over the research recovery period of five years. In addition, the current investigation took into account that the recovery period for all procedures using the CHEM, AUT, and ENZ methodologies is 15 years. Such a recovery period is crucial and demonstrates to investors how economically feasible the procedure is (Sinnott and Towler 2019). An 11% discount rate was taken into consideration in the proposed economic inquiry.

Another economic indicator is the net present value (NPV), which calculates the difference between the current values of cash inflows and outflows. In order to account for the time value of money, this value annualizes the present value using the interest rate. Equation (5) is employed to determine the NPV (Thoppil and Zein 2021):

$$\text{NPV} = \sum_{n=1}^{n=t} \frac{\text{CF}_n}{(1+i)^n} \quad (5)$$

If CF_n is the cash flow in years, i is the discount rate, and t is the project duration in years.

The ratio of investment to net income is known as the return on investment, or ROI. A high ROI concentration shows that an investment's returns outperform its expenses. ROI is used to assess the efficacy of various investments as a performance indicator of investment efficiency. The following Eq. (6) is used to calculate ROI (Sinnott and Towler 2019):

$$\text{ROI} = \frac{\text{Cumulative net profit}}{\text{Plant life} \times \text{initial investment}} \times 100 \quad (6)$$

Environmental analysis

As climate change is currently a worldwide concern, the environmental effects (carbon footprint) of the suggested method were addressed. The carbon footprint is defined as the greenhouse gas amounts generated by an organization per unit of product. This terminology is usually examined with respect to the carbon dioxide (CO_2) mass equivalent. Greenhouse gases are generated from energy use in industrial operations.

The energy demands attributed to α -monolaurin synthesis through CHEM, AUT, and ENZ were calculated on the basis of energy balance requirements. The energy usage in the ENZ method is principally due to the esterification step. Conversely, the energy connected with the CHEM and AUT methods is mainly consumed in the short-path distillation step, esterification, and glycerin recovery. The CHEM and AUT methods, in contrast to ENZ, require a heavy-duty steam boiler to produce both high- and low-pressure steam. The esterification reaction is heated to 190 °C using this steam. Furthermore, the vacuum ejector system's pressure is produced using this steam (to serve the deglycerination and short-path distillation units).

In this study, two CO_2 sources were considered: one from the usage of electricity and another from the production of steam for heating. Equation (7) was used to determine how much CO_2 was created as a result of power use:

$$QE_{\text{CO}_2} = \text{Electricity consumption} \times EF_{\text{CO}_2} \quad (7)$$

QE_{CO_2} is the total CO_2 emissions attributable to the process' use of electricity, and EF_{CO_2} is the emission factor determined by the energy provider. Following Mata et al. (2018), EF_{CO_2} was calculated in this study to be 438.64 g CO_2 eq/kWh. Equation (8) was used to determine the emissions due to steam generation. It was assumed

Table 2 Experimental results of enzymatic method

No	Factor levels	A: Temperature [°C]	B: Enzyme amount [% w/w]	C: Molar ratio [-]	D: Mol. sieve [% w/w]	Conversion [%]
1	--00	50	2	3.5	10	76.16
2	+ -00	60	2	3.5	10	81.30
3	- +00	50	6	3.5	10	85.91
4	+ +00	60	6	3.5	10	85.96
5	00--	55	4	1	5	64.28
6	00+-	55	4	6	5	88.05
7	00-+	55	4	1	15	65.71
8	00++	55	4	6	15	87.16
9	-00-	50	4	3.5	5	85.91
10	+00-	60	4	3.5	5	83.80
11	-00+	50	4	3.5	15	85.59
12	+00+	60	4	3.5	15	84.88
13	0--0	55	2	1	10	50.00
14	0+-0	55	6	1	10	68.57
15	0-+0	55	2	6	10	84.44
16	0++0	55	6	6	10	87.54
17	-0-0	50	4	1	10	60.50
18	+0-0	60	4	1	10	50.32
19	-0+0	50	4	6	10	73.13
20	+0+0	60	4	6	10	89.18
21	0-0-	55	2	3.5	5	83.98
22	0+0-	55	6	3.5	5	84.47
23	0-0+	55	2	3.5	15	68.00
24	0+0+	55	6	3.5	15	88.00
25	0000	55	4	3.5	10	84.51
26	0000	55	4	3.5	10	85.55
27	0000	55	4	3.5	10	85.91
28	0000	55	4	3.5	10	85.55
29	0000	55	4	3.5	10	83.09

Table 3 The ANOVA results of fitted model

	SS	df	MS	F-value	p-value	R-squared (R^2)	Adjusted R^2	Predicted R^2
Model	3424.79	12	285.40	27.27	<0.0001	0.9534	0.9184	0.7792
Residual	167.45	16	10.47					
Lack of Fit	162.16	12	13.51	10.22	0.0189			
Pure Error	5.29	4	1.32					
Cor Total	3592.25	28						

Adeq. Precision: 19.5628

that the steam generator (boiler) relied on natural gas for its operation.

$$F_{\text{energy}} = 0.0471 \text{ GJ/kg} \quad (\text{B o o k } 2011);$$

$$F_{\text{emissions}} = 0.05582 \text{ tCO}_2/\text{GJ} \quad (\text{Revised } 1996)$$

$$\text{TE}_{\text{steam}} = \text{fuel amount} \times F_{\text{energy}} \times F_{\text{emission}} \quad (8)$$

F_{energy} is the energy required to produce one mass of steam, and F_{emission} is the amount of CO_2 emitted for every unit of energy.

The emissions associated with raw materials employed in producing monolaurin are calculated. These included the emissions from the production of lauric acid, glycerin, and catalysts. Palm kernel oil was considered the source of lauric acid and glycerin. Lauric acid was industrially produced using two process steps of fat splitting followed by fractional distillation. At the same time, pure glycerin was produced after passing three process steps: sweet water purification, evaporation and distillation.

Table 4 The results of each fitted model terms

	SS	df	F-value	p-value
A-Temperature	5.66	1	0.5406	0.4728
B-Enzyme amount	266.68	1	25.48	0.0001
C-Molar ratio	1878.00	1	179.44	<0.0001
D-Mol. Sieve	10.36	1	0.9899	0.3346
AB	6.48	1	0.6189	0.4430
AC	172.00	1	16.43	0.0009
BC	59.83	1	5.72	0.0294
BD	95.16	1	9.09	0.0082
A ²	32.82	1	3.14	0.0956
B ²	26.94	1	2.57	0.1282
C ²	853.27	1	81.53	<0.0001
D ²	8.57	1	0.8188	0.3789

The energy and power consumption while producing the aforementioned raw materials were sourced from Oleo Mir for Oleochemicals Company, Egypt. The data given were converted into a mass of carbon dioxide emissions. Due to enzyme production, the emission factor was considered 25 kg CO₂/Kg lipase (Kim et al. 2009). Transportation of raw materials was also considered. The distance from Malaysia to Egypt was considered for palm kernel oil. While the distances from Denmark to Egypt were considered for Lipozyme 435. The transportation emission factor was considered 0.133 kg CO₂ eq/ t. km (Mata et al. 2018).

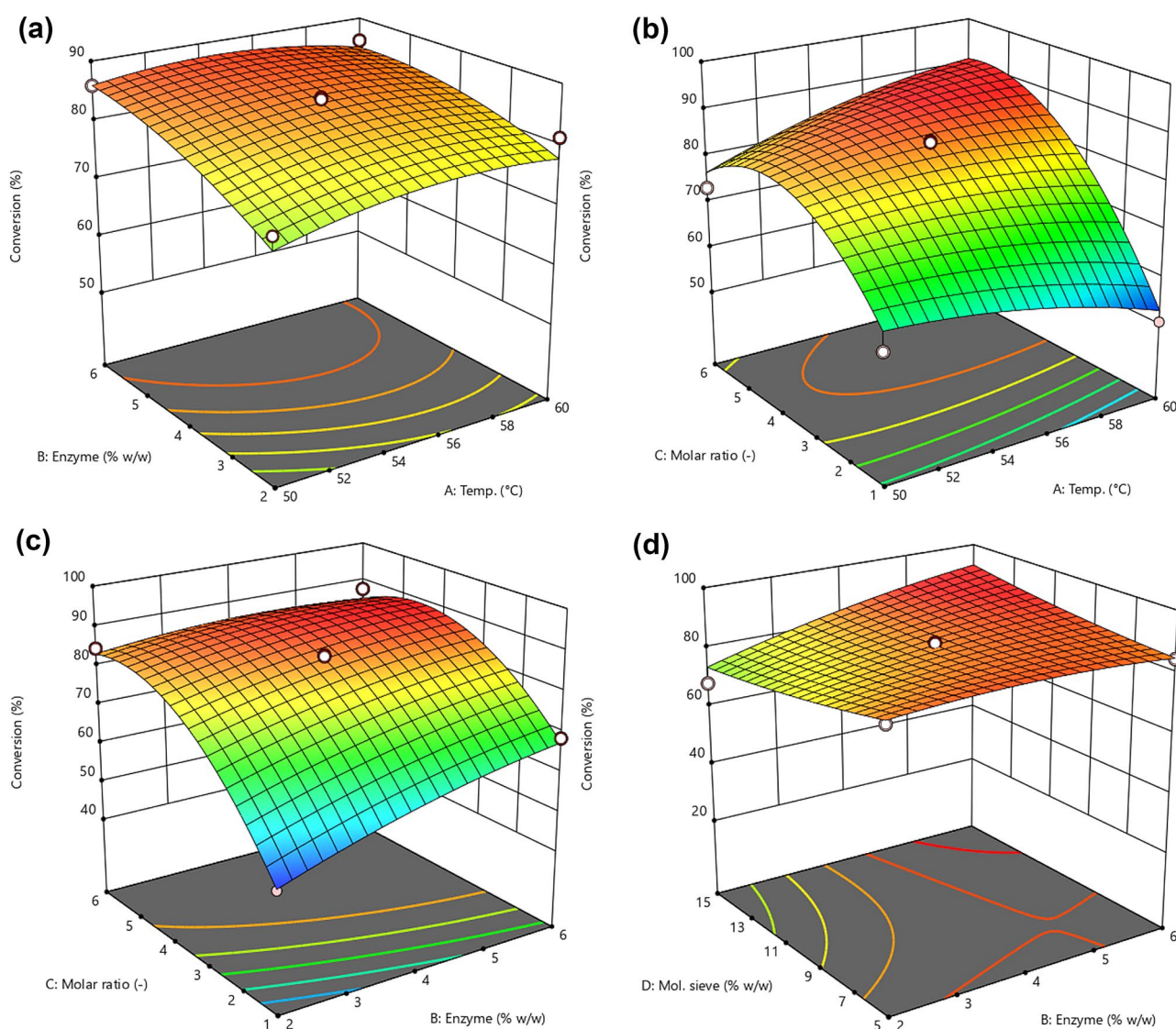


Fig. 1 Three-dimensional surface plots showing the interaction of two variables at the zero-factor level of other variables; the combined effects of the temperature and enzyme (a), temperature and molar ratio (b), enzyme and molar ratio (c), and the enzyme and molecular sieve (d)

Results and discussion

Validations of model equations

For the investigation of monolaurin synthesis by enzymatic method, the conversion of process was chosen as response. Table 2 shows the experimental design matrix created by using the Box–Behnken model and the conversion results.

A quadratic model equation of the conversion is generated by applying ANOVA. For better definition of this model equation, the *F*-value, *p*-value and lack of fit test of each model term can be evaluated (Bakari et al. 2020). At this case, the improved model equation formed by the actual factor values can be used directly to predict the experimental conversion results. After elimination of some insignificant terms, the model equation was developed in this study as follows.

$$\begin{aligned} \text{Conversion} = & -164.25 + 8.71 * A + 11.26 * B - 7.91 \\ & * C - 3.06 * D - 0.13 * AB + 0.52 \\ & * AC - 0.77 * BC + 0.49 * BD - 0.09 \\ & * A^2 - 0.51 * B^2 - 1.84 * C^2 + 0.05 * D^2 \end{aligned}$$

where *A* is the temperature (°C); *B* is the enzyme amount (%w/w); *C* is the molar ratio (-) and *D* is molecular sieve (%w/w). In this equation, *AB*, *AC*, *BC* and *BD* represent the interaction terms of the factors, and *A*², *B*², *C*² and *D*² denotes the quadratic equation terms of the factors. The sign of regression coefficients within this equation also indicates how the response will be affected by the change in the factor (Anupam et al. 2016). Depending on linear model term coefficients, the molar ratio and molecular sieve negatively affect the reaction conversion, while the temperature and enzyme amount affect it positively. The quadratic coefficient of the molar ratio also illustrates negative sign, but those of the temperature, enzyme amount, and molecular sieve exhibit opposite signs to the related linear term coefficients. Considering the interaction coefficients, the positive effects display in the interaction of the temperature and molar ratio, and the enzyme amount and molecular sieve, whereas the other remaining interactions affect negatively. It is stand out that the linear coefficient of the molar ratio has an opposite effect in the 4-factor model comparing with the 3-factor equation created by Mustafa et al. (2016) at the different level. It is thought that this opposite effect could be resulted by increasing the molar ratio from 4 to 6, as well as by adding the molecular sieve as a new factor.

As a result of ANOVA, a model with low *p*-value (<0.05) and high *F*-value are accepted to be significant (Kutlu and Kocar 2020). In the current study, the adequate *p*-value and *F*-value indicated correspondingly the significant model equation (Table 3). Other analyses that determine how compatible the model is are the *p*-value of the lack of fit

Table 5 Selectivity differences of type of technology toward monoglycerides formation

Technology	CHEM	AUT	ENZ
<i>α</i> -Monolaurin	43.9	41.7	49.5
1,3 Dilaurin	32.6	29.8	41
Trilaurin	5.4	4.5	1.6

test and *R*-squared results. The result of *R*-squared (95.32%) significantly shows the compatibility of model equation. The predicted *R*-squared of 0.7792 is in reasonable agreement with the adjusted *R*-squared of 0.9184. Adequate precision of 19.563 also indicates that this model can be used to navigate the design space. On the other hand, it is also seen that the model can be further improved according to the *p*-value of lack of fit. The *p*-value, which was expected to be insignificant, turned out to be significant in this model, indicating that there are abundant residuals in some experimental runs. With some repeated experiments, this issue was improved. It is thought that the model equation obtained in this study can be improved more, especially by changing in intervals for temperature and molecular sieve. This approach is also understood from the *F*-values of the factor. Frišták et al. (2015) indicated that a model term having high *F*-value also possesses the highest impact on the response. In this model, the molar ratio has the highest effect on the reaction conversion, followed by the amount of enzyme (Table 4).

Some insignificant model terms are still present (*AB*, *A*², *B*² and *D*²) as shown in Table 4. Not all taken away from the equation because of their low effects on the *R*-squared of model, the terms with the highest effect neglected. Depending on *p*-values calculated, the most effective insignificant model terms were found as *AD* and *CD*, therefore these terms were eliminated from the models.

Parametric interaction assessment

The possible combined effects on the response in case of variation in the factors can be easily assessed by the three-dimensional surface plots. It is understood that the interaction between the two factors can be seen obviously in the model in case elliptical contour plots (Muralidhar et al. 2001).

The interaction of the two factors is shown in Fig. 1, and the factors other than the interacting factors in the plots were set at the zero-factor level. Among the four interaction terms used in the model equation, only the enzyme amount and molecular sieve interaction (*BD*) did not form perfect elliptical contours. This issue is thought to be caused by the linear effect of the molecular sieve. The *BD* interaction produced a saddle surface plot, resulting in multiple maximum or minimum stationary points. The conversion of

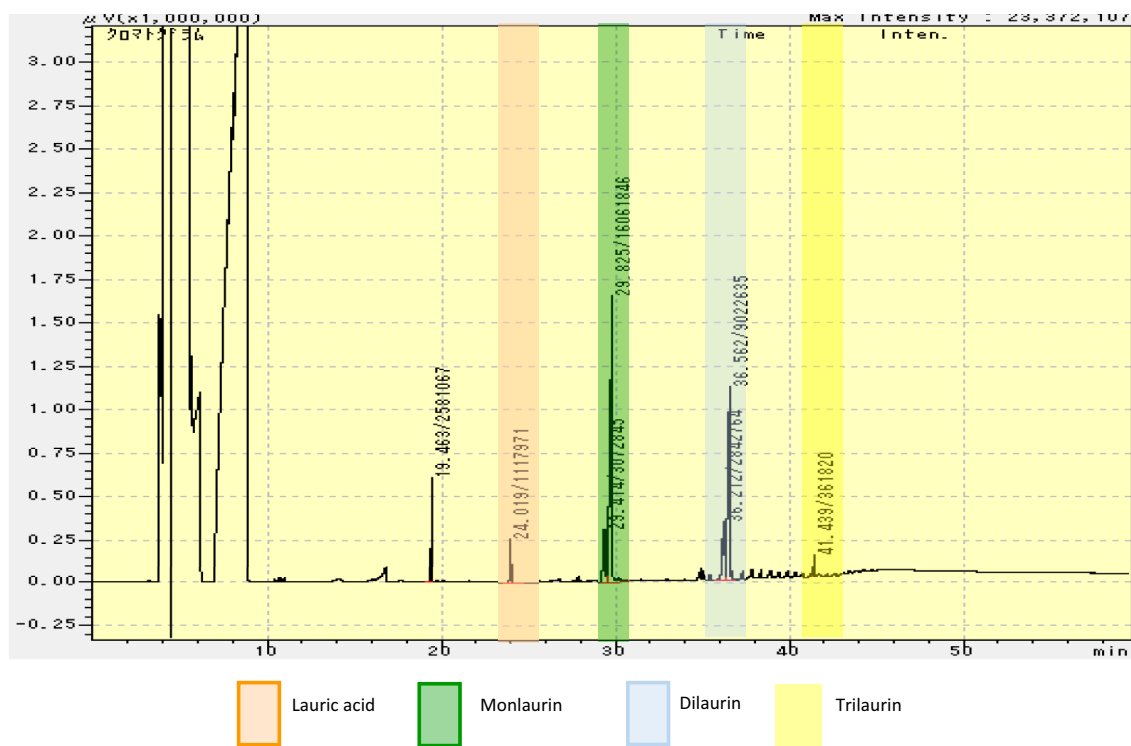


Fig. 2 GC chromatogram for Monolaurin synthesis under optimized conditions (ENZ): temperature of 56.95 °C, molecular sieve of 14.85% w/w, enzyme amount of 5.38% w/w, as well as molar ratio of 4.75% w/w

Table 6 Summary of total capital investment

Cost parameter	Cost of CHEM plant (USD)	Cost of AUT plant (USD)	Cost of ENZ plant (USD)
ISBL	3,394,918	2,561,988	870,692
OSBL	1,018,476	768,596	261,208
Direct capital investment cost (ISBL + OSBL)	4,413,394	3,330,584	1,131,899
Cost of engineering	–	–	–
Cost of contingency	441,339	333,058	169,785
Fixed capital cost	4,854,733	3,663,642	1,301,684
Startup expenses	441,339	333,058	113,190
Working capital	662,009	499,588	169,785
Total capital investment	7,149,698	5,395,546	1,901,591

50% was obtained at the maximum temperature and minimum molar ratio. On the other hand, two maximum points about 90% of the conversion were found by increase in the molar ratio at high temperature, medium enzyme amount and molecular sieve.

The perfect elliptical surfaces are obtained in other interaction plots, therefore only one optimum conversion is occurred. In the AB interaction plot, the maximum

conversion is determined with high enzyme amount and low-medium temperature in case of the other factors at zero-level. As a result of increasing molar ratio and decreasing molecular sieves value, this effect is reversed, i.e., the maximum conversion can be achieved at low enzyme amount and high temperature values.

According to the AC interaction plot, the changes in the constant factors did not affect this result, while the maximum conversion was observed at high temperature and molar ratio. It was determined that the maximum conversion was affected by the changes in the constant parameters in the BC interaction. With the decrease in constant factors, a conversion of 80% can be achieved at the level of medium enzyme amount and molar ratio. While the response (> 90%) increased with the increase of temperature and molecular sieve, the high enzyme amount and molar ratio were supported to that result.

When these assessments are considered, the conversion is affected by the chosen factors in different ways. For this reason, finding the maximum conversion using contour or surface plots can be misleading. In order to reach the maximum conversion, the response was optimized depending on the fitted model equation via software program. It was aimed to keep the variable factors within the limit range as well as to maximize the conversion, and the importance level of the variables was selected as same. Eventually, hundred

Table 7 Summary of total cost of production

Reactants	Cost (\$)	CHEM route		AUT route		ENZ route	
		Amount (t)	Price(\$)	Amount (t)	Price(\$)	Amount (t)	Price(\$)
<i>Raw materials</i>							
Glycerin ^a (31 of plant capacity)	2.75/kg	5.3475	14,706	5.208	14,322	4.883	13,427
Lauric acid (69% of plant capacity)	1.493/kg	11.9025	17,770	11.592	17,307	10.868	16,225
TIB166 (0.06% w/w) (Price of Tinex S is included)	405/kg	10.35 kg	4,192	–	–	–	–
Lipozyme 435 (1 kg/2tons of Monolaurin)	–	–	–	–	–	1085/kg	7.5 kg
Washing water	0.00227/kg	–	15	–	15	–	15
<i>Utilities</i>							
Steam	0.0227/MJ ^a	22,700 MJ	515	22,700 MJ ^b	515	1713 MJ	38
Electric power	0.136/kWh ^a	1800 kWh	225	1800 kWh ^b	225	125 kWh	17
Variable production cost/day			37,423		32,384		37,837
Packing	1%		374		324		378
Repair and maintenance	1%		374		324		378
Waste stream disposal	1%		374		324		378
Total production cost/day		\$/day	38,545		33,355		38,972
Total production cost/ton		\$/ton	2,570		2,224		2,598
Total production cost/year		\$/year	12,720,012		11,007,273		12,860,814
Gross Profit*/d		\$/d	34,805		39,995		34,378
Gross Profit*/y		\$/y	11,485,488		13,198,227		11,344,686
Profit, %		%	90.3		112		88
*Market price of α -Monolaurin is 4890 \$/t							

^aReference: (Mustafa et al. 2022)

^bReference: Oleo Misr for Oleochemicals Company–Egypt

alternative conditions where the conversions change in the range of 89–91% were found. The optimum conversion having the highest desirability was determined at the temperature of 56.95 °C, %5.38 enzyme amount, 4.75 molar ratio and 14.85 molecular sieve.

Effect of reaction route selectivity on monoglycerides formation

Table 5 shows the selectivity differences for the type of technology used for monoglycerides formation. When glycerin esters are synthesized, high percentages of monoglycerides and diglycerides are usually appreciated, especially monoglycerides owing to their high antimicrobial effect (Satyawali et al. 2021). As can be seen in Fig. 2, the highest α -monolaurin of 49.5% and 1,3 dilaurin of 41% were obtained when the ENZ process was applied. This was because of the higher selectivity of lipase toward monoglyceride formation. Meanwhile, the CHEM process yielded 43.9% of α -monolaurin and 32.6% of 1,3 dilaurin. The AUT

technology produced 41.3% of α -monolaurin and 29.8% of 1,3 dilaurin. This indicates that TIB160 also promoted the monoglyceride and diglyceride formation compared with the case without any catalyst (AUT). It could be also noted that trilaurin was favorably less in the case of the ENZ process than those in AUT and CHEM, where the values obtained were 1.6, 4.5, and 5.4%, respectively. Similar observations were found by Satyawali et al. (2021) who reported a yield of 35–50 wt% when monolaurin was produced under different reactor configurations (batch and fixed-bed reactors) in a solvent-free system.

Economic evaluation

When a type of investment must be selected (CHEM/AUT/ENZ), total capital cost calculations play an important role for decision-makers. In this context, the ISBL cost was initially calculated as it examines the feasibility of a process. Bridgewater's method was used in this study to calculate the ISBL cost. Table 6 shows that the ISBL costs were

\$ 4,413,394, \$ 3,330,584, and \$ 1,131,899 for CHEM, AUT, and ENZ, respectively. It must be mentioned that the plant cost of the CHEM technology was the highest because of the presence of more downstream processes, namely, short-path distillation and heterogeneous catalyst removal. Meanwhile, the ENZ method represented the lowest investment cost among all methods because of its simplicity (one-step reaction process). Table 6 shows that the investment needed for the ENZ process is four times less than that of the CHEM process and three times lower than that of the AUT process. Such a large variation in the investment cost demonstrates a promising opportunity for the implementation of large-scale ENZ plants for the production of oleochemicals. It should be mentioned that the authors of this paper received a direct esterification plant offer from a common Malaysian engineering vendor last year. The vendor quoted a price of \$ 4 million for the same capacity chosen here (4950 t/year). Such a quotation is highly close to the ISBL cost calculated in this research using Bridgewater’s method. This validates the calculations and price comparisons presented in this research. In our previous work, Mustafa et al. (2022) reported that the ISBL cost of the CHEM process for glycerin monostearate production is higher than that of the ENZ process by two and half times considering the same capacity presented in this paper. The higher price difference in this research (that for CHEM was four times higher than that for ENZ) is due to two reasons. The first is the simplicity of the proposed ENZ method conducted in a solvent-free system. This is unlike that in Mustafa et al. (2022), where α -monostearin production was conducted in an organic medium, which required solvent removal process steps that entailed additional ISBL costs. The second reason is that in this work, the catalyst is heterogeneous, which required a filtration unit. In comparison, in our previous work (Mustafa et al. 2022), the catalyst presented was homogenous (NaOH), and it needed no additional unit operation to separate it as it remained in the short-path distillation residues.

Concerning the manufacturing cost, the ENZ method is attractive because of its reduced energy usage. Table 7 shows that the energy consumption due to steam generation is only 1713 MJ/d for the ENZ process, as opposed to 22,700 MJ/d for the CHEM and AUT methods. The main reason for this great difference is the presence of short-path distillation in the CHEM and AUT processes, which consumes greater amounts of energy. In addition, the high reaction temperature of 190 °C contributes to the higher energy consumption. Such favorable less energy usage proves the cleaner nature of the suggested ENZ method over the CHEM and AUT processes in compliance with SDG No. 12, which states the principle of responsible usage and synthesis. The current SDGs draw attention to “doing more and better with less,” which is about upgrading resources and energy performance (Sachs, 2012). The SDGs are also concerned with

Table 8 Summary of NPV for α -monolaurin production using the CHEM technology

Project year	Investment	Capacity	Charge of depreciation	Investment	Expenses of operation	Gross profit	Expenses of depreciation	Tax income	Tax paid	Cash flow	Discounted cash flow	Pretax cash flow
1	0	0	0	0	\$0.00	\$0.00	\$0.00	\$0.00	\$0.00	\$0.00	\$0.00	\$0.00
2	100	0	0	-\$7,149,697.96	\$0.00	\$0.00	\$0.00	\$0.00	\$0.00	-\$7,149,697.96	-\$6,441,169.34	-\$7,149,697.96
3	0	100	0.2	0	-\$12,720,012.27	\$11,485,487.73	\$2,297,097.55	\$9,188,390.18	\$2,067,387.79	\$22,138,112.21	\$17,967,788.50	\$20,070,724.42
4	0	100	0.32	0	-\$12,720,012.27	\$11,485,487.73	\$3,675,356.07	\$7,810,131.66	\$1,757,279.62	\$22,448,220.38	\$16,187,196.85	\$20,690,940.75
5	0	100	0.192	0	-\$12,720,012.27	\$11,485,487.73	\$2,205,213.64	\$9,280,274.09	\$2,088,061.67	\$22,117,438.33	\$14,583,060.22	\$20,029,376.66
6	0	100	0.1152	0	-\$12,720,012.27	\$11,485,487.73	\$1,323,128.19	\$10,162,359.54	\$2,286,530.90	\$21,918,969.10	\$13,137,892.09	\$19,632,438.21
7	0	100	0.1152	0	-\$12,720,012.27	\$11,485,487.73	\$1,323,128.19	\$10,162,359.54	\$2,286,530.90	\$21,918,969.10	\$11,835,938.82	\$19,632,438.21
8	0	100	0.0576	0	-\$12,720,012.27	\$11,485,487.73	\$661,564.09	\$10,823,923.64	\$2,435,382.82	\$21,770,117.18	\$10,663,007.95	\$19,334,734.36
9	0	100	0	0	-\$12,720,012.27	\$11,485,487.73	\$0.00	\$11,485,487.73	\$2,584,234.74	\$21,621,265.26	\$9,606,313.47	\$19,037,030.52
10	0	100	0	0	-\$12,720,012.27	\$11,485,487.73	\$0.00	\$11,485,487.73	\$2,584,234.74	\$21,621,265.26	\$8,654,336.46	\$19,037,030.52
11	0	100	0	0	-\$12,720,012.27	\$11,485,487.73	\$0.00	\$11,485,487.73	\$2,584,234.74	\$21,621,265.26	\$7,796,699.51	\$19,037,030.52
12	0	100	0	0	-\$12,720,012.27	\$11,485,487.73	\$0.00	\$11,485,487.73	\$2,584,234.74	\$21,621,265.26	\$7,024,053.61	\$19,037,030.52
13	0	100	0	0	-\$12,720,012.27	\$11,485,487.73	\$0.00	\$11,485,487.73	\$2,584,234.74	\$21,621,265.26	\$6,327,976.23	\$19,037,030.52
14	0	100	0	0	-\$12,720,012.27	\$11,485,487.73	\$0.00	\$11,485,487.73	\$2,584,234.74	\$21,621,265.26	\$5,700,879.48	\$19,037,030.52
15	0	100	0	0	-\$12,720,012.27	\$11,485,487.73	\$0.00	\$11,485,487.73	\$2,584,234.74	\$21,621,265.26	\$5,135,927.46	\$19,037,030.52
					Cumulative profit	\$149,311,340	ROI = 139%				NPV	\$128,179,901

Table 9 Summary of NPV for α -monolaurin production using the AUT technology

Project year	Investment	Capacity	Charge of depreciation	Investment	Expenses of operation	Gross Profit	Expenses of depreciation	Tax income	Tax paid	Cash flow	Discounted cash flow	Pretax cash flow
1	0	0	0	0	\$0.00	\$0.00	\$0.00	\$0.00	\$0.00	\$0.00	\$0.00	\$0.00
2	100	0	0	-\$5,395,546.15	\$0.00	\$0.00	\$0.00	\$0.00	\$0.00	-\$5,395,546.15	-\$4,860,852.39	-\$5,395,546.15
3	0	100	0.2	0	-\$11,007,272.65	\$13,198,227.35	\$2,639,645.47	\$10,558,581.88	\$2,375,680.92	\$21,829,819.08	\$17,717,570.88	\$19,454,138.16
4	0	100	0.32	0	-\$11,007,272.65	\$13,198,227.35	\$4,223,432.75	\$8,974,794.60	\$2,019,328.78	\$22,186,171.22	\$15,961,775.57	\$20,166,842.43
5	0	100	0.192	0	-\$11,007,272.65	\$13,198,227.35	\$2,534,059.65	\$10,664,167.70	\$2,399,437.73	\$21,806,062.27	\$14,379,977.99	\$19,406,624.54
6	0	100	0.1152	0	-\$11,007,272.65	\$13,198,227.35	\$1,520,435.79	\$11,677,791.56	\$2,627,503.10	\$21,577,996.90	\$12,954,935.12	\$18,950,493.80
7	0	100	0.1152	0	-\$11,007,272.65	\$13,198,227.35	\$1,520,435.79	\$11,677,791.56	\$2,627,503.10	\$21,577,996.90	\$11,671,112.72	\$18,950,493.80
8	0	100	0.0576	0	-\$11,007,272.65	\$13,198,227.35	\$760,217.90	\$12,438,009.45	\$2,798,552.13	\$21,406,947.87	\$10,514,515.97	\$18,608,395.75
9	0	100	0	0	-\$11,007,272.65	\$13,198,227.35	\$0.00	\$13,198,227.35	\$2,969,601.15	\$21,235,898.85	\$9,472,536.91	\$18,266,297.69
10	0	100	0	0	-\$11,007,272.65	\$13,198,227.35	\$0.00	\$13,198,227.35	\$2,969,601.15	\$21,235,898.85	\$8,533,817.03	\$18,266,297.69
11	0	100	0	0	-\$11,007,272.65	\$13,198,227.35	\$0.00	\$13,198,227.35	\$2,969,601.15	\$21,235,898.85	\$7,688,123.45	\$18,266,297.69
12	0	100	0	0	-\$11,007,272.65	\$13,198,227.35	\$0.00	\$13,198,227.35	\$2,969,601.15	\$21,235,898.85	\$6,926,237.35	\$18,266,297.69
13	0	100	0	0	-\$11,007,272.65	\$13,198,227.35	\$0.00	\$13,198,227.35	\$2,969,601.15	\$21,235,898.85	\$6,239,853.46	\$18,266,297.69
14	0	100	0	0	-\$11,007,272.65	\$13,198,227.35	\$0.00	\$13,198,227.35	\$2,969,601.15	\$21,235,898.85	\$5,621,489.61	\$18,266,297.69
15	0	100	0	0	-\$11,007,272.65	\$13,198,227.35	\$0.00	\$13,198,227.35	\$2,969,601.15	\$21,235,898.85	\$5,064,405.05	\$18,266,297.69
					Cumulative profit	\$171,576,955	ROI = 212%				NPV	\$127,885,499

Table 10 Summary of NPV for α -monolaurin production using the ENZ technology

Project Year	Investment	Capacity	Charge of depreciation	Investment	Expenses of operation	Gross profit	Expenses of depreciation	Tax income	Tax paid	Cash flow	Discounted cash flow	Pretax cash flow
1	0	0	0	0	\$0.00	\$0.00	\$0.00	\$0.00	\$0.00	\$0.00	\$0.00	\$0.00
2	100	0	0	-\$1,901,590.68	\$0.00	\$0.00	\$0.00	\$0.00	\$0.00	-\$1,901,590.68	-\$1,713,144.76	-\$1,901,590.68
3	0	100	0.2	0	-\$12,860,814.14	\$11,344,685.86	\$2,268,937.17	\$9,075,748.68	\$2,042,043.45	\$22,163,456.55	\$17,988,358.53	\$20,121,413.09
4	0	100	0.32	0	-\$12,860,814.14	\$11,344,685.86	\$3,630,299.47	\$7,714,386.38	\$1,735,736.94	\$22,469,763.06	\$16,205,728.41	\$20,734,026.13
5	0	100	0.192	0	-\$12,860,814.14	\$11,344,685.86	\$2,178,179.68	\$9,166,506.17	\$2,062,463.89	\$22,143,036.11	\$14,599,755.32	\$20,080,572.22
6	0	100	0.1152	0	-\$12,860,814.14	\$11,344,685.86	\$1,306,907.81	\$10,037,778.04	\$2,258,500.06	\$21,946,999.94	\$13,152,932.72	\$19,688,499.88
7	0	100	0.1152	0	-\$12,860,814.14	\$11,344,685.86	\$1,306,907.81	\$10,037,778.04	\$2,258,500.06	\$21,946,999.94	\$11,849,488.94	\$19,688,499.88
8	0	100	0.0576	0	-\$12,860,814.14	\$11,344,685.86	\$653,453.91	\$10,691,231.95	\$2,405,527.19	\$21,799,972.81	\$10,675,215.26	\$19,394,445.62
9	0	100	0	0	-\$12,860,814.14	\$11,344,685.86	\$0.00	\$11,344,685.86	\$2,552,554.32	\$21,652,945.68	\$9,617,311.04	\$19,100,391.37
10	0	100	0	0	-\$12,860,814.14	\$11,344,685.86	\$0.00	\$11,344,685.86	\$2,552,554.32	\$21,652,945.68	\$8,664,244.18	\$19,100,391.37
11	0	100	0	0	-\$12,860,814.14	\$11,344,685.86	\$0.00	\$11,344,685.86	\$2,552,554.32	\$21,652,945.68	\$7,805,625.39	\$19,100,391.37
12	0	100	0	0	-\$12,860,814.14	\$11,344,685.86	\$0.00	\$11,344,685.86	\$2,552,554.32	\$21,652,945.68	\$7,032,094.95	\$19,100,391.37
13	0	100	0	0	-\$12,860,814.14	\$11,344,685.86	\$0.00	\$11,344,685.86	\$2,552,554.32	\$21,652,945.68	\$6,335,220.67	\$19,100,391.37
14	0	100	0	0	-\$12,860,814.14	\$11,344,685.86	\$0.00	\$11,344,685.86	\$2,552,554.32	\$21,652,945.68	\$5,707,406.01	\$19,100,391.37
15	0	100	0	0	-\$12,860,814.14	\$11,344,685.86	\$0.00	\$11,344,685.86	\$2,552,554.32	\$21,652,945.68	\$5,141,807.22	\$19,100,391.37
					Cumulative profit	\$147,480,916.12	ROI = 517%				NPV	\$133,062,043.89



Fig. 3 Contribution of the individual raw materials used for monolaurin production. **a** Sensitivity of lauric acid purchasing price on ROI. **b** Sensitivity of Glycerin price variation on ROI. **c** ROI dependency on

monolaurin market price. **d** Sensitivity of monolaurin quantity produced per one kg of Lipozyme 435 on ROI

mitigating climate change and improving carbon neutrality. In this regard, the suggested ENZ method has less energy and fewer losses usage and therefore matches the main aim of SDG No. 12.

Besides the ISBL calculations, the feasibility of a process application depends on the value of the total production cost. The calculated total production costs in this study were 2570, 2224, and 2598 \$/t for the CHEM, AUT, and

ENZ methods, respectively, as shown in Table 7. Such results reveal that the production cost of the ENZ process is higher than that of the CHEM and AUT processes by 10% and 6.5%, respectively. Similar results were reported by Jegannathan et al. (2011), who found that the production cost of biodiesel using ENZ technology is higher than that of the alkali-catalyzed process by 2.1 times. In this study, the catalyst contributed 21% (Lipozyme 435) and 11% (TIB160) of the total variable production cost of the ENZ and CHEM processes, respectively. Meanwhile, the energy accounted for 1.7% of the variable production cost in the CHEM/AUT methods compared with only 0.11% for the ENZ process.

Table 7 also shows that the most profitable method is AUT, with a profit percentage of about 112%, followed by the CHEM and ENZ processes, with profits of 90.3% and 88%, respectively. This suggests that all proposed processes are highly profitable and economically viable. Nevertheless, in the authors' opinion, no competition between the CHEM/AUT and ENZ methods must be created considering the synthesis cost as long as profit is there. The authors believe that the production cost of the ENZ method is usually greater than that of traditional methods and that the contrary must not be predicted because it is a matter of comparing energy costs with the costs of immobilized enzymes. The cost of energy requirements for the CHEM/AUT processes is less than the cost of the immobilized enzymes in the ENZ method, although there is an interesting energy saving due to the use of enzymes. Nevertheless, consumers these days have become more educated on and lean over purchasing green products. Consequently, they began to pay extra for products that had appealing phrases like "green" and "free of."

NPV is an intriguing technique that tackles the difference between the present value of cash inflows and outflows over a specific period (in this study, 15 years). The NPV must be positive to have an economically feasible technique. The NPV conclusions for CHEM, AUT, and ENZ are, respectively, shown in Tables 8, 9, and 10. The first year of a project is often regarded as the design stage and the beginning of the cash flow. Then, Year 2 is thought to include the project's building and installation stages, where the total fixed capital cost is specified. When the facility reaches its maximum production in Year 3, depreciation costs start to be subtracted from the gross profit.

Tables 8, 9, and 10 show that all methods (CHEM, AUT, and ENZ) have positive NPV and ROI, which emphasize the economic feasibility of all three production methods. It should be noted that the ENZ method had the highest NPV and ROI even though it had the highest manufacturing costs. The less ISBL cost contributed to such positive value after the 15-year recovery period. In comparison, our previous work (Mustafa et al. 2022) reported a negative NPV and ROI for the ENZ process using the same plant

Table 11 Annual carbon footprint of α -monolaurin production using enzymatic and autocatalytic approaches^a

CO ₂ emissions (t CO ₂ eq./year)	Auto-catalytic process	Enzymatic process
Electricity	260.5	18
Steam	418	32
Subtotal	678.5	50
<i>Materials</i>		
Lauric acid	147	138.3
Glycerin	92.8	87
Lipozyme 435	–	61.875
Transportation of enzyme ^b	–	1.2
Transportation of palm kernel oil ^c	4380	3608
Total/year	5300	3950

^aThe detailed calculations of the environmental study are presented in the supplementary materials

^bDistance between Denmark and Egypt is 3688 km

^cDistance between Malaysia and Egypt is 7925 km

capacity for α -monostearin (commodity oleochemicals) production. In that research, we recommended the use of ENZ technology to produce specialty oleochemicals instead (highly priced with high-profit margins) to cover the cost of expensive enzymes. We herein propose the production of α -monolaurin, which is highly priced compared with the α -monostearin presented in our previous research (Mustafa et al. 2022). Although both monolaurin and monostearin are produced using the same equipment/technology, the current selling price of α -monolaurin is 4890 \$/t, whereas the market price of α -monostearin is only 2100 \$/t. In this work, we again emphasize the validity of green enzymatic production for specialty oleochemical production (high prices) instead of basic oleochemicals. Additional research efforts should be done to further reduce manufacturing costs in producing basic oleochemicals such as fatty acids, glycerin, and commodity esters using enzymes. This mainly can be achieved with the development of new reactor configurations, process optimization, enzyme reusability, cheap immobilization techniques and carriers, and customer awareness of the benefits of using ENZ methods in greenhouse gas mitigation.

Sensitivity analysis

The sensitivity analysis offers a comprehensive economic understanding of cost variation in the α -monolaurin production process. The economic factor, namely ROI, was the focus of this investigation. The feedstock price, including lauric acid and glycerin, played a significant role in the cost of producing α -monolaurin, as shown in Fig. 3a. Also, the market price of α -monolaurin potentially affected the process profitability. The positive ROI indicates a profitable

production process. The sensitivity analysis of lauric acid and glycerin purchase costs on ROI is shown in Figs. 3b and c, respectively. It should be mentioned that when studying the effect of lauric acid price variation on ROI, the glycerin cost was fixed at 2750 \$/t, while when studying the impact of glycerin price variation, the lauric acid price was set at 1493 \$/t. The price of Lipozyme 435 was fixed in all cases at 1082 \$/kg.

Figure 3b shows that the ROI percent decreased dramatically from 566 to 297% by varying the price of lauric acid from 1200 \$/t to 2800\$/t, respectively. The ROI for glycerin decreased from 604 to 490% when ranging the glycerin price from 1600 \$/t to 3100\$/t, respectively. It can be noted that the ROI percent value is more affected by varying the price of lauric acid than the price of glycerin, mainly because the higher mass stoichiometric portion of lauric acid (about 70%) compared to glycerin (30%) while feeding the esterification plant. Likewise, the sensitivity analysis of α -monolaurin price on the ROI was investigated. As shown in Fig. 3d, not surprisingly, the ROI increased as the market price of the final product increased at fixed lauric acid and glycerin prices.

Nevertheless, it is a common practice that the α -monolaurin price is directly affected by the price of lauric acid and glycerin. Therefore, practically the ROI may not significantly be affected by the α -monolaurin price variation, as the relation between the prices of α -monolaurin, lauric acid, and glycerin are directly proportional.

Finally, regarding Lipozyme 435, it was crucial to study the sensitivity of enzyme consumption on ROI. In this sense, the quantity of α -monolaurin produced was varied against consuming 1 kg of Lipozyme 435. Figure 3e shows the sensitivity of the amount of product per one unit mass of Lipozyme 435. The tested range of α -monolaurin production quantity was from 0.25 to 4 t. The study was performed considering the following market prices of 2750 \$/t, 1493 \$/t, 4890\$/t, and 1082 \$/kg for glycerin, lauric acid, α -monolaurin, and Lipozyme 435, respectively. Figure 3e shows a negative ROI value at 0.25 t of product lipase-1; the negative ROI illustrates that the project is not profitable. The production quantity of 0.5 t of product lipase-1 was the minimum quantity that gave a positive ROI.

Environmental assessment

Greenhouse gas mitigation is a subject of great concern currently. In this study, we looked at the proposed ENZ process' carbon footprint and compared the findings to those of the CHEM and AUT processes, as shown in Table 11. It should be noted that the current study did not consider the emissions caused by raw materials (life cycle emissions), stainless steel, or steel used in plant building. Therefore, only emissions resulting from the use of heat energy and

electricity for α -monolaurin synthesis were taken into account. Table 11 shows that the AUT/CHEM processes generated a total CO₂ exhaust amount of 260.5 and 418 eq./year due to electricity and steam generation, respectively. In comparison, the ENZ process exhausted a total amount of only 18 and 32 t CO₂ eq./year due to electricity and steam generation, respectively. The current results demonstrate that the CHEM/AUT routes produced 14 times more emissions of CO₂ than that of the ENZ method (excluding the emissions associated with raw materials production and transportation), emphasizing the greener synthesis route of the suggested technique.

Conclusion

This work reported a cleaner and cost-feasible production route for α -monolaurin using the developed ENZ method. For comparison purposes, three different routes (CHEM, AUT, and ENZ) were studied. Techno-economic and environmental investigations were carefully established to understand in-depth the feasibility of the studied processes. The ENZ esterification reaction between glycerin and lauric acid was conducted in a solvent-free system and yielded about 50% of α -monolaurin, compared with 43.9 and 41.7% for the CHEM and AUT processes, respectively. The capital costs were estimated to be \$7,149,698, \$5,395,546, and \$1,901,591 for the CHEM, AUT, and ENZ methods, respectively, based on the same plant capacity of 4950 t/year. The remarkable investment returns of the ENZ technology create a promising opportunity to encourage manufacturers to invest. The ROI and NPV estimations for the ENZ method, after a recovery period of 15 years, were favorably higher than those of both the CHEM and AUT processes, which proves the economic viability of the proposed technology. The AUT/CHEM processes generated a total CO₂ exhaust of 678.7 t CO₂ eq./year. In contrast, the ENZ process exhausted a total CO₂ of only 50 t CO₂ eq./year. The current results demonstrate that the CHEM/AUT routes produced 14 times more CO₂ emissions than that of the ENZ method, emphasizing the greener synthesis route of the suggested technique.

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Author contributions A.M. conceived of the presented idea and wrote the manuscript. S.F. and OK developed the theory and performed RSM study. F.N. and A.I. verified the analytical methods. M.M. developed the GC analysis.. T.A. and A.B. developed the mass and energy balance calculations. O.S. supervised the findings of this work. C.B. and L.B. performed the economic assessment. M.T. developed the sensitivity analysis. M.M. developed the environmental analysis. R.M. managed, revised and edited all the manuscript. All authors discussed the results and contributed to the final manuscript.

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Data Availability Enquiries about data availability should be directed to the authors.

Declarations

Conflict of interest The authors declare no competing interests.

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