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Activity of hydrolytic lipase enzyme synthesised by *Aspergillus terreus* isolated from petroleum-contaminated soil

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Abstract

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Lipase enzyme-mediated processes have biological significance and tremendous potential in various industrial and technological areas due to the enzymatic properties and broad specificity for a wide spectrum of substrates. This research involved the production of hydrolytic lipase enzyme by *Aspergillus terreus* extracted from petroleum-contaminated soil and optimising the environmental parameters that may affect lipase production. Hydrolytic lipase enzyme activity of the isolated *Aspergillus terreus* from petroleum-contaminated soil was undertaken in submerged fermentation. Dry cell weight, lipase enzyme pellets, and lipase enzyme concentration by measuring the percentage transmission by spectrophotometry were measured to determine the effects of various abiotic factors (pH, temperature, carbon and nitrogen sources, lipid substances and vegetable oils) affecting cell growth and lipase enzyme production by *Aspergillus terreus* on modified lipase production media. The results showed a precipitate on Tween 20 and a bright pink-fluorescent halo on the rhodamine B agar plate. After temperature adjustment to 35 °C, statistical analysis showed that *Aspergillus terreus* produced a dry cell weight of 3.14 g/L and a lipase enzyme pellet of 0.33 g with a spectrophotometric transmission of 0.03%. The sucrose substrate displayed a dry cell weight of 3.11 g/L and a lipase enzyme pellet of 1.15 g. Spectrophotometric analysis showed that sucrose, glucose and peptone had a similarity in transmission at $p = 0.321$. The addition of Neem oil improved enzyme yield as a dry cell weight of 3.02 g/L and a lipase enzyme pellet of 0.95 g was obtained. Statistically, sunflower and tributyrin similarly affected enzyme production by showing a spectrophotometric transmission at $p = 0.024$. This study has indicated that *Aspergillus terreus* could be employed as a reliable source of lipase enzymes for industrial processes due to its stability across different abiotic factors.

Keywords: abiotic factors, dry cell weight, hydrolytic lipase enzyme, lipase enzyme pellet, percentage transmission.

INTRODUCTION

The use of enzyme-mediated processes can be traced back to ancient civilisations (Chandra et al., 2020). Over 4000 enzymes are known and about 200 are exploited for commercial and industrial uses (Arantes et al., 2020). The sales market of enzymes has grown spectacularly due to an improved understanding of synthetic organic chemistry applications and biochemical and catalytic activity during the fermentation process (Xu et al., 2021; Sharma et al., 2022). Besides catalysing alcoholysis, acidolysis,

esterification and aminolysis, lipase is also involved in hydrolysis and interesterification reactions (Chandra et al., 2020; Muigano et al., 2023). In addition to their biological significance, lipases have tremendous potential in food and leather technology, biomedical sciences, agrochemical, paper manufacture, nutrition, cosmetics, pharmaceutical, dairy and chemical industries (Arantes et al., 2020; Sharma et al., 2022). Other promising fields for applying lipases include detergent formulations, synthesis of biosurfactants, biodegradation of plastics and the resolution of racemic mixtures to produce optically active compounds. Thus, the development of lipase-based technologies for synthesising novel compounds is rapidly expanding the uses of these enzymes (Zarinviarsagh et al., 2017; Ali et al., 2023).

Multi-faceted microbial lipases (glycerol ester hydrolases) have an unsurpassed role in the swiftly growing modern biotechnology due to their indispensable bioconversion properties of lipids (triacylglycerols) from one organism to another or within the same organisms (Pandey et al., 1999). Lipase possesses various properties, including stability in organic solvents that confer broad specificity for a wide spectrum of substrates (Arpigny & Jaeger, 1999; Albayati et al., 2020). Unlike esterases, lipase enzymes possess the unique feature of acting at an interface between the aqueous and non-aqueous organic phases (Xu et al., 2021). Lipases act tremendously in mild conditions; their catalytic activity arises from the interfacial activation that generally depends on the aggregation state of the substrate and the surface area exposed to the lipase enzyme (Urbanek et al., 2020).

Lipase is used in various organic solvents. It shows selectivity for a specific type of reaction, as described by Zarinviarsagh et al. (2017), on the biochemical activity of lipase enzymes in novel media. Research shows biotransformation and the biotechnological applications of polyunsaturated fatty acids, which involve the unique nature of lipase-catalysed processes (Pandey et al., 1999).

Massive production of an enzyme is limited by its production cost and specificity in terms of action properties, even though there is a renewed interest in developing lipase enzymes due to its various applications in the modern world (Urbanek et al., 2020). The catalytic properties of hydrolytic lipase enzyme have diversified its commercial use in the formulation of laundry detergents since they are thermostable and remain active in the alkaline environment, which is dominant in the washing process. Approximately 1000 tons of lipases are added to 13 billion tons of detergents produced yearly (Arantes et al., 2020).

Numerous species of bacteria, yeasts and moulds produce lipases with different enzymological properties and specificities. Still, moulds are known to be more potent lipase producers on solid substrate and submerged fermentations (Aachapa et al., 2021). This research intends to produce hydrolytic lipase enzymes from a soil fungus and find optimal conditions to improve the mass cultivation of lipase enzyme production.

Most commercially available lipases are synthesised by fungi and yeasts, including *Rhizopus delemar*, *Humicola lanuginosa*, *Penicillium chrysogenum*, *Fusarium heterosporum*, *Rhizopus chinensis* and *Candida rugosa* (Ramos-Sánchez et al., 2015; Kumar et al., 2023). A search for reliable sources of lipase enzyme by solid-state fermentation continues due to its cheapness, simplicity, lower levels of catabolite repression, better product recovery and high-quality production (Sharma et al., 2022).

The Filamentous *Aspergillus terreus* fungi has been found suitable for solid-state fermentation because of its hyphal mode of fungal growth and its tolerance to environmental parameters like high pH, temperature and osmotic pressure thus, making it a good source of Lovastatin, and its semisynthetic derivative simvastatin (Abu-Tahon et al., 2020; Al-Sa'ady & Aziz, 2020). Due to these characteristics of *Aspergillus terreus* and given the potential applications of lipase enzyme, the present study aimed at the production of hydrolytic lipase enzyme by *Aspergillus terreus* extracted from petroleum-contaminated soil and optimising the environmental parameters that may affect lipase production.

MATERIALS AND METHODS

Soil sample and culture media

The soil sample was collected from a petrol bunk located at Automax garage, Kinondoni municipality, Dar es Salaam, Tanzania (6°42′19.08″ S; 39°6′45.72″ E) and transported to the Laboratory of Microbiology at Kampala International University in Tanzania (KIUT), Dar es Salaam, Tanzania. Cultur-

ing media, including potato dextrose agar, Tween 20 medium and yeast extract peptone medium, were bought from *Lab Equip Suppliers Limited* at Kariako, Dar es Salaam, Tanzania.

Isolation, identification and cultivation of *Aspergillus terreus*

The petroleum-contaminated soil was serially diluted at 10^{-1}, 10^{-2}, 10^{-3} and 10^{-4}, as adapted by Booth (1971). The lowest dilution of 10^{-4} was subjected to culturing on potato dextrose agar and incubated at 30 °C for 24 hours. From the various fungal colonies on the potato dextrose agar plates, the *Aspergillus terreus* strain was identified, isolated and sub-cultured in a potato dextrose agar slant and maintained at 30 °C in the laboratory for further experimentation.

Determination of the hydrolytic lipase enzyme activity produced by *Aspergillus terreus*

The lipase activity was performed on plates with Tween 20 (10.0 g of peptone, 5.0 g of NaCl, 0.1 g of $CaCl_2 2H_2O$, 20.0 g of agar, 1000 mL of water and the pH was adjusted to 6.0) and on rhodamine B (15.0 g of olive oil, 0.01 g of rhodamine B, 20.0 g of agar dissolved in 1000 mL of water). The plates were inoculated with *Aspergillus terreus* at the centre using a pinpoint inoculum. Lipolytic activity on the Tween 20 agar plate was determined after 24 hours of incubation at 28 °C, as the rhodamine B agar plate was checked after seven days of incubation at 28 °C. The rhodamine B agar plate was irradiated with UV light (350 nm) to visualise the colour changes (Pérez et al., 2021).

Hydrolytic lipase enzyme production and recovery in submerged fermentation

A colony of *Aspergillus terreus* fungus was picked from a plate, introduced into 10 mL Potato Dextrose Agar broth and allowed to grow for 24 h to create the starter culture. For lipase enzyme accumulation, 2 mL of the starter culture was drawn and introduced in 100 mL of the modified lipase production media in 250 mL Elerlynmer flasks and allowed to grow in a rotary shaker incubator at 150 rpm at 30 °C for five days. The modified lipase production media used in this study was prepared using the formula adopted by Fatima et al. (2021), where 10.0 g peptone, 1.0 g yeast extract, 1 g of filter sterilised glucose to 1 L of 0.1 M phosphate buffer and pH was adjusted to 7.0. After five days of incubation, the culture was filtered through a filter paper to remove the mycelium. The cells were harvested by centrifugation at 8600× g for 15 min. The pellet was air-dried overnight to obtain a constant dry weight, which was taken and designated a dry cell weight.

The dry cell pellet was suspended in a solution containing 10 mL of 10% (*v/v*) sodium hypochlorite and 10 mL chloroform and was incubated at 37 °C for 2 h to digest the cellular components. After incubation, the suspension was centrifuged at 8600× g for 15 min, and the bottom phase was recovered. The dry cell pellet was suspended in a solution containing 10 mL of 10% (v/v) sodium hypochlorite and 10 mL chloroform and was incubated at 37 °C for 2 h to digest the cellular components. After incubation, the suspension was centrifuged at 8600× g for 15 min, and the bottom phase was recovered and air-dried overnight, its weight taken and designated as the lipase enzyme pellet. The weight difference between the dry cell weight and the lipase enzyme pellets was defined as the residual biomass. The percentage of lipase enzyme pellet accumulation was computed as the proportion of lipase enzyme in a sample to the dry cell weight. The lipase enzyme pellet was refrigerated at -20 °C and used as the enzyme source to study the enzyme activity.

Quantification of cell growth

Cell growth was monitored by measuring the turbidity of forty-eight-hour-old *Aspergillus terreus* culture on a UV spectrophotometer at an optical density of 600 nm. Cell growth also involved monitoring different parameters such as carbon, nitrogen, pH, temperature, and effects of lipid and vegetable oils to determine the optimal conditions for biomass yield.

Effect of pH and temperature

The isolated culture of *Aspergillus terreus* was inoculated in 100 mL of lipase production medium before incubation at five different temperatures of 4 °C, 15 °C, 25 °C, 35 °C and 45 °C and the effect of pH was studied by adjusting the pH of the lipase pro-

duction medium to different pH values as 5, 6, 7, 8 and 9. The experiments were conducted in triplicate.

Effect of carbon and nitrogen sources

The *Aspergillus terreus* culture was inoculated in a 100 mL lipase production medium, and 1 g of glucose, sucrose, maltose, lactose and starch were added as carbon sources. In other different inoculations, 1 g of yeast extract, peptone, malt extract, ammonium sulphate, ammonium nitrate and ammonium chloride were added as nitrogen sources. The experiments were conducted in triplicate.

Effect of lipid substances and vegetable oils

The *Aspergillus terreus* culture was inoculated in the 100 mL lipase production medium and added 1 g of Tween 20, Tween 40, Tween 60, Tween 80 and tributyrin as lipase production mediums. The *Aspergillus terreus* culture was inoculated in 100 mL lipase production medium, and 1 mL of sunflower, castor, mustard, groundnut, palm, neem and gingelly (sesame) oils, respectively, were added as vegetable oil sources.

Concentration (percentage transmission) of lipase enzyme

The lipase activity was assayed quantitatively using the method applied by Winkler & Stuckmann (1979). This included the mixing of the collected lipase enzyme pellet of the test substance with 2 mL of 4-nitrophenyl palmitate (10 mL isopropanol containing 30 mg 4-nitrophenyl palmitate (Sigma) mixed with 90 mL of 0.05 M sodium phosphate buffer containing 207 mg sodium deoxycholate and 100 mg gum Arabic at a pH of 8.0). The mixture was pre-warmed for 15 min at 37 °C before being subjected to spectrophotometry at an absorbance of 410 nm against a blank to determine the percentage transmission (%T).

Data analysis

Computed results on dry cell weight, lipase enzyme pellet and percentage transmission (%T) collected after adjustment of different abiotic parameters (pH, temperature, carbon, nitrogen, lipid and vegetable oils) were subjected to SPSS software (Version 29) for analyses. The analysis involved the determination of the descriptive statistics of the mean-variance and subjecting the data to a generalised linear model by utilising the least significant difference test at $p \leq 0.05$. In the analysis model, the test substrate's volume was the independent variable, as dry cell weight, lipase enzyme pellet, and percentage transmission (%T) were the dependent variables.

RESULTS

Isolation of *Aspergillus terreus* and detection of hydrolytic lipase enzyme activity

In the 10^{-4} serially diluted samples of the petroleum-contaminated soil, a total of 32 colonies of wild fungal strains were isolated. Among the colonies, eight species of fungal species were isolated that belonged to two genera, these were collected from the Zygomycotina (*Rhizopus stolonifer*), Deuteromycotina (*Aspergillus fumigatus*, *Aspergillus japonicus*, *Aspergillus nidulans*, *Aspergillus niger*, *Aspergillus terreus*, *Penicillium citrinum*, *Penicillium oxalicum*) (Fig. 1).

Aspergillus terreus inoculated on Tween 20 agar plates exhibited lipolytic activity, whereby a precipitate was visualised around the inoculated area. Lipolytic activity was also found to take place on the rhodamine B agar after visualisation under UV light, whereby a bright pink fluorescent halo was visualised (Fig. 2).

Production of hydrolytic lipase enzyme by the isolated *Aspergillus terreus* by submerged fermentation

A dry cell weight of 2.04 g/L was obtained from the 2 mL of *Aspergillus terreus* grown in the 100 mL of modified lipase production media. A 0.18 g lipase enzyme pellet was obtained after processing and purification of the dry cell weight of 2.04 g; this was equivalent to 8.82% of the dry cell weight.

Effects of abiotic variables on hydrolytic lipase production by *Aspergillus terreus*

Temperature and pH

Temperature and pH changes affected lipase enzyme production by *Aspergillus terreus* on modi-

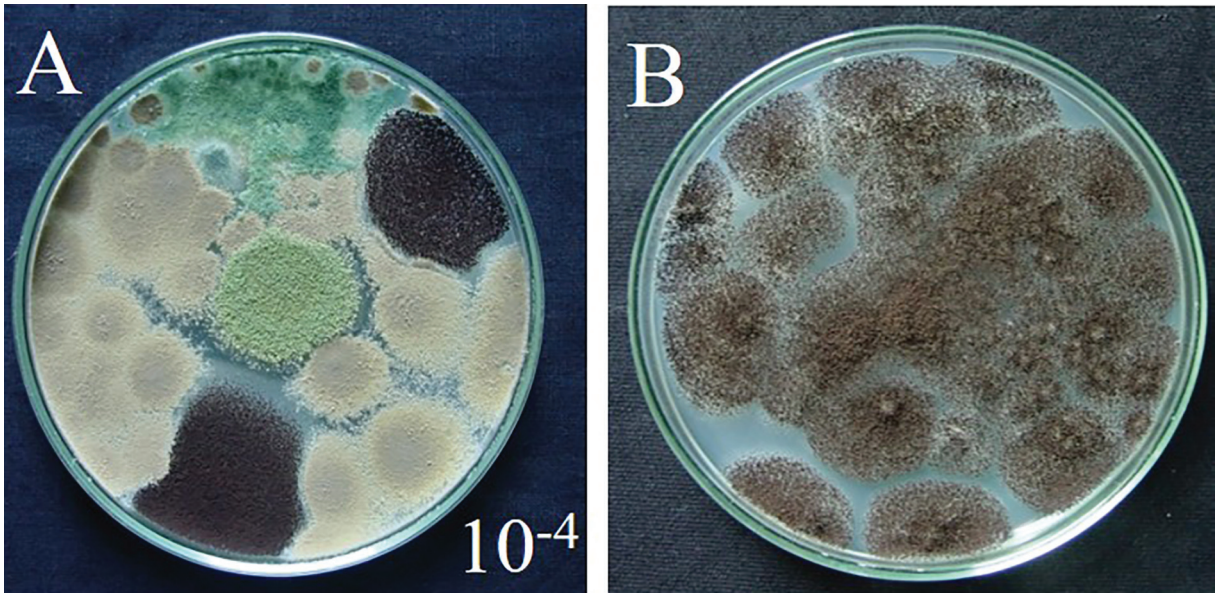

Fig. 1. Potato dextrose agar plate of the 10^{-4} dilution of the petroleum-contaminated soil (A) and pure culture of *Aspergillus terreus* (B)

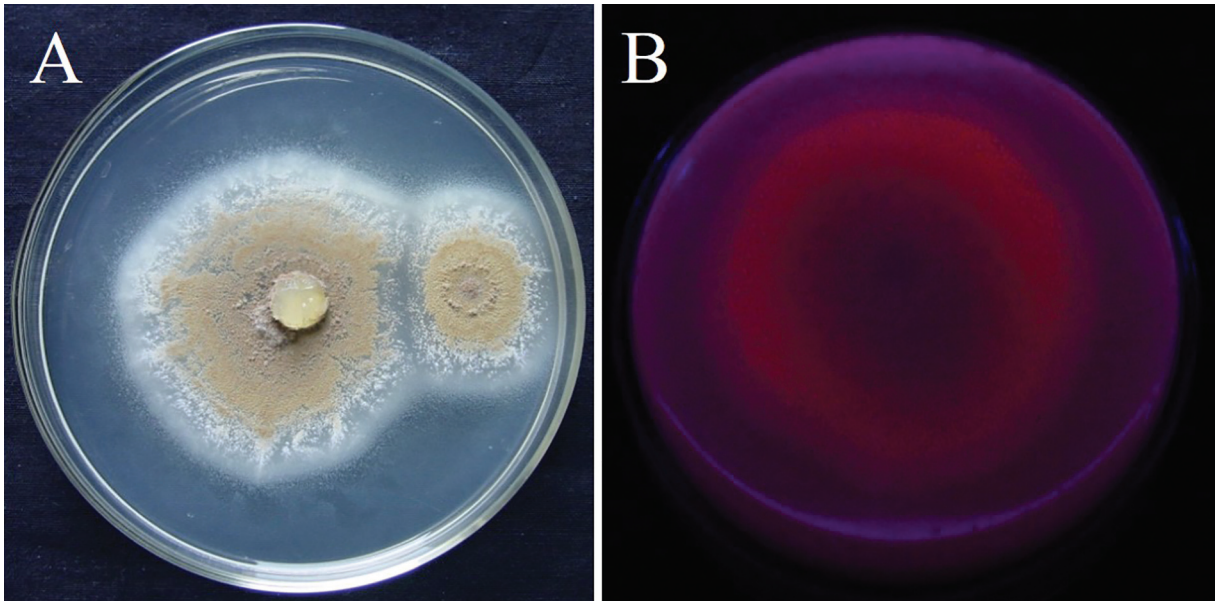

Fig. 2. *Aspergillus terreus* causes a precipitate zone on the Tween 20 agar plate (A) and a fluorescent halo on the rhodamine-B agar plate (B)

fied lipase production media. A 3.14 g/L of dry cell weight, a lipase enzyme pellet of 0.33 g with a transmission of 0.03% was obtained at a temperature of 35 °C. At pH 7, a dry cell weight yield of 3.03 g/L and a lipase enzyme pellet of 0.29 g were obtained. The lowest performances were obtained at a temperature of 4 °C and pH 5 with transmission of 0.1 % at $p = 0.001$.

Carbon and nitrogen sources

Aspergillus terreus on modified lipase production media after the addition of sucrose sugar displayed a quantity of 3.11 g/L of dry cell weight that yielded 1.15 g of lipase enzyme pellet followed by glucose with a dry cell weight of 3.03 g/L and a lipase enzyme pellet of 1.07 g. Peptone added to the modified lipase production media enhanced a lipase enzyme pellet of 1.12 g

and 0.66 g for peptone and malt extract, respectively. Spectrophotometric analysis showed a significantly similar transmission of 0.05 at $p = 0.321$ for sucrose, glucose and peptone, as starch, malt and yeast extract attained transmission of 0.06%, significance in %T exhibited similarity of lipase concentration in the solutions. Maltose and ammonium nitrate nitrogen sources were similar in the produced dry cell weight of 2.09 g/L and a percentage transmission of 0.08; $p = 0.105$ (Table 1).

Lipid substances and vegetable oils

Aspergillus terreus on modified lipase production media yielded 3.02 g/L of dry cell weight after adding Neem oil, whereby 0.95 g of lipase enzyme pellet was obtained. The addition of Tween 60 modified lipase production media yielded 2.92 g/L of dry cell weight and 0.84 g of lipase enzyme pellet. A spectrophotometric transmission of 0.04 and 0.05 was obtained on modified lipase production media after the addition of neem oil and tributyrin (Fig. 3). Statistically, castor oil and mustard oil showed a similarity of 0.6 g lipase enzyme pellet production at $p = 0.102$ as sunflower and Tween 80 had 0.57 g at $p = 0.081$. Spectrophotometric analysis showed a significant transmission of 0.05 % at $p = 0.024$ for sunflower and tributyrin, 0.06 at $p = 0.011$ for Tween 40, castor, mustard and sesame oils, while groundnut oil and Tween 20 exhibited a transmission of 0.07% at $p = 0.025$ (Fig. 4).

DISCUSSION

The result acquired in the present experimentation indicates the capability of *Aspergillus terreus* to produce lipase enzymes. Various techniques have been used for lipase detection, including those with Tween 80 and tributyrin as substrate in solid media (Pérez et al., 2021). Based on results obtained in this experiment, *Aspergillus terreus* was found to produce lipase enzyme as visualised on Tween 20 and rhodamine B agar plates. The formation of a precipitate layer around the inoculated area on the Tween 20 agar plate was a significant indication of lipase activity. Even though Tweens' are not suitable as substrates because they are heterogeneous, they are hydrolysed by lipases, resulting in competition with the intended substrate (Corbellini et al., 2007). Rhodamine B agar contains a chromogenic substrate (rhodamine B) with an action mechanism involving the union with fatty acids and diglycerides. This action can be detected by changing colour to bright pink on the rhodamine B agar plate when viewed under fluorescent light, thus, semi-quantifying lipase activity (Lechuga et al., 2016; Urbanek et al., 2020).

Lipolysis can be determined through many different methods, such as titrimetry, colourimetry, spectrophotometry or fluorimetry, radioactive fatty acids, gas-liquid chromatography and immunological techniques. Visual assessment of colourimetric and/or fluorimetric assays can be used in experiments where quantitation is unnecessary (Jensen, 1983; Kilcawley, 2021). This experiment utilised the spectrophotometry technique to determine the lipase activity by determining its percentage transmission, whereby the lower the transmission depicted, the higher the concentration of the lipase enzyme in the measured

Table 1. Effects of various carbon and nitrogen sources on the production of hydrolytic lipase enzyme by *Aspergillus terreus*. Means with similar letters (a, b, c, d and e) are not significantly different at $p \leq 0.05$ applying the test of least significant difference

Substances		Dry cell weight (g/L)	Lipase enzyme pellets (g)	Concentration of lipase enzyme (%T)
Carbon source	Lactose	2.34 ± 0.12	0.22 ± 1.09	0.07 ± 0.22^{c}
	Maltose	2.09 ± 1.03^{e}	0.04 ± 0.09	0.08 ± 1.32^{d}
	Sucrose	3.11 ± 0.99	1.15 ± 1.33	0.05 ± 041^{a}
	Glucose	3.03 ± 2.01	1.07 ± 0.87	0.05 ± 1.16^{a}
	Starch	2.62 ± 0.44	0.15 ± 1.02	0.06 ± 2.05^{b}
Nitrogen source	Ammonium chloride	2.18 ± 1.14	0.13 ± 1.25	0.07 ± 0.38^{c}
	Ammonium nitrate	2.09± 1.11^{e}	0.07 ± 1.18	0.08 ± 1.03^{d}
	Ammonium sulphate	2.03 ± 0.32	0.02 ± 0.52	0.10 ± 0.02
	Malt extract	3.05 ± 2.01	0.66 ± 0.44	0.06 ± 1.15^{b}
	Peptone	3.08 ± 0.45	1.12 ± 0.65	0.05 ± 1.09^{a}
	Yeast extract	2.58 ± 1.24	0.38 ± 0.12	0.06 ± 2.13^{b}

substance. In this experimentation, a dry cell weight of 2.04 g/L, 0.18 g of lipase enzyme pellet and 0.081 percentage transmission (%T) of lipase enzyme were obtained from the activity of *Aspergillus terreus* on 100 mL of modified lipase production media. These results were relatively similar to *Pseudomonas aeuruginosa* grown on modified lipase production media where lipase enzyme with a dry cell weight of 2.2 g/L

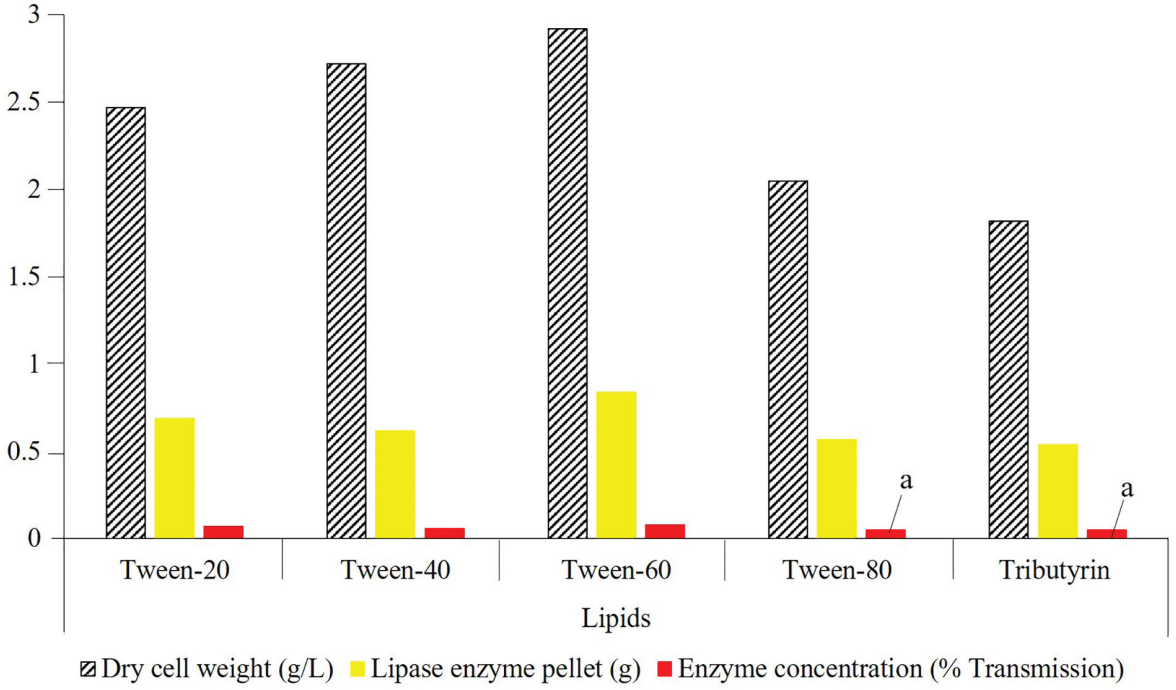

Fig. 3. Effects of lipids on the production of lipase enzyme by *Aspergillus terreus,* similar letters indicate similar expression at $p \leq 0.05$ using the least significant difference test

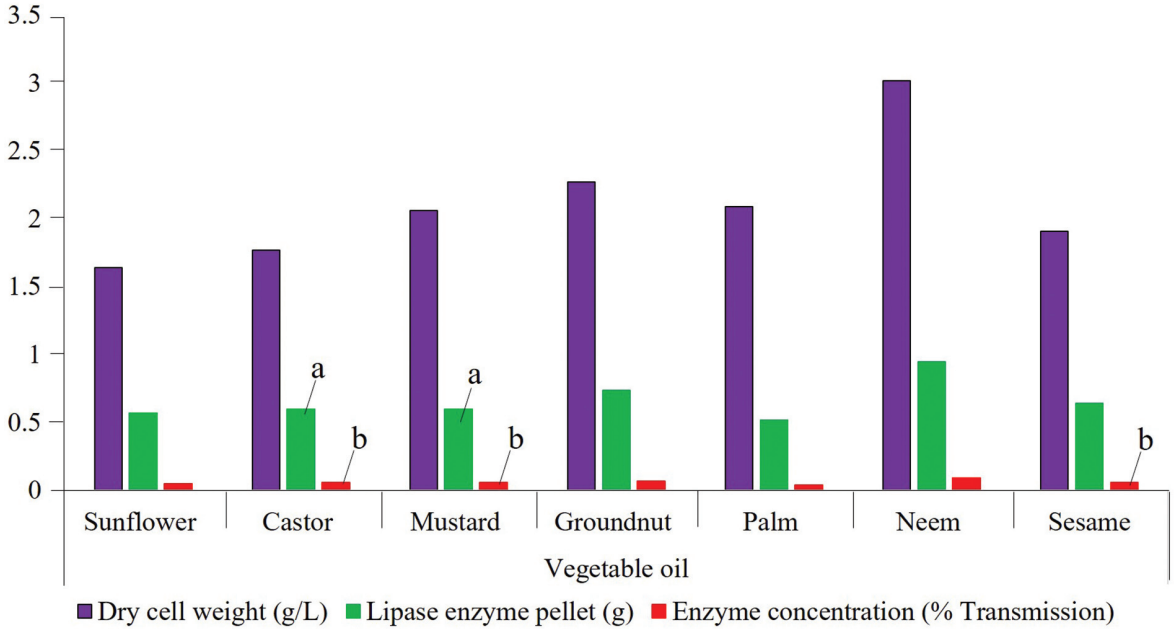

Fig. 4. Effects of vegetable oils on the production of lipase enzyme by *Aspergillus terreus*, similar letters indicate similar expression at $p \leq 0.05$ using the least significant difference test

and lipase enzyme pellet of 0.12 g was obtained (Ito et al., 2001; Mauti, 2022). Obtained results in this research are comparatively different from a report by Ramos-Sánchez et al. (2015), where *Aspergillus niger*, *Aspergillus ochraceus*, *Rhizopus stolonifer* and *Aspergillus terreus* on rhodamine B agar media generated a dry cell weight of 1.81, 1.71, 1.53 and 1.28 g/L with a lipase enzyme pellet of 0.11, 0.08, 0.12 and 0.09 g, respectively.

It is widely accepted that abiotic factors such as pH, temperature, moisture, carbon and nitrogen sources and growth medium can strongly influence lipase production and enzymatic activities (Rehman et al., 2019). Lipase enzymes can metabolise well in various pH and temperature conditions (Mauti et al., 2016; Bibi et al., 2022). The results of the current study showed that a moderate temperature of 35 °C and pH 7 effectively produced the lipase enzyme by *Aspergillus terreus* on modified lipase production media. Research by Patel et al. (2020) has shown that maximum lipase production on lipase production media by *Pseudomonas aeuriginosa* grown on modified lipase production media is at pH 6 and 7. A different study by Bouras et al. (2017) has displayed that pH 5 and a temperature of 38 °C is appropriate for lipase enzyme synthesis by *Aspergillus niger* and *Rhizopus stolonifera* on rhodamine B agar media.

In this study, it was noted that at a temperature reduction to 4 °C and pH 5, there was a reduction in the production of the lipase enzyme by *Aspergillus terreus*. This asserts studies by Mehta et al. (2018), where a temperature of 5 °C shows a spectrophotometric transmission of 0.11, indicating a reduced amylase enzyme production by *Aspergillus fumigatus*. Rehman et al. (2019) have stated that temperature above 48 °C and pH 10 denature lipase enzyme produced by *Aspergillus niger*. Sucrose and glucose displayed a recommendable dry cell weight and lipase enzyme pellet and a low spectrophotometric transmission of 0.05%. Similar results on carbon sources were obtained by lipase produced by *Bacillus pumilus* on modified nutrient broth media. The addition of glucose and sucrose substrates on the modified nutrient broth media has displayed a spectrophotometric transmission of 0.06 and 0.09%, respectively (Kowanga et al., 2016; Aachapa et al., 2021).

Lipolytic activity of *Aspergillus terreus* on peptone and yeast extracts, when added to the nutrient broth, shows recommendable results with a lipase enzyme pellet of 1.36 and 1.01 g and a spectrophotometric transmission of 0.04 and 0.07%, respectively (Shabbir & Mukhtar, 2018). Similar results were obtained in this study, where the peptone displayed a significant dry cell weight and lipase enzyme pellet, thus being a potential substrate for lipase enzyme production. The results in this study were contrary to the activity of *Aspergillus terreus* and *Aspergillus eucalypticola* strains on rhodamine B agar when ammonium nitrate substrate was added to attain a spectrophotometric transmission of 0.04 and 0.02%, respectively. Adding ammonium nitrate intensified extracellular lipase production compared to peptone and yeast extracts (Gulati et al., 1999; Kowanga et al., 2015; Shreya & Sharma, 2023).

Neem oil and Tween 60 substrates displayed recommendable results on dry cell weight and lipase enzyme pellet. This was contrary to the effects recorded by Moyses et al. (2021) on lipase enzyme extracted from *Penicillium restrictum*, whereby castor oil substrate had a dry cell weight and lipase enzyme pellet 2.01 g/L and 0.81 g, respectively. This research attained a percentage transmission of 0.06 on the activity of *Aspergillus terreus* on castor and mustard oil, which was much lower when compared to results displayed by Barros et al. (2023). In this research, spectrophotometry transmission indicated a reduction in the lipase enzyme concentration in Tween 80, 60, 40 and 20 compared to the activity on the tributyrin substrate. Similar research by Shabbir & Mukhtar (2018) has attained a spectrophotometric transmission of 0.21% and has stated that the Tween 80 substrate increases cell permeability, thus reducing the synthesis of cell-bound lipase by *Aspergillus terreus*. Thus, this brief study indicated that *Aspergillus terreus* could be employed as a source of lipase enzymes for use in specific industrial processes since it showed higher stability across different abiotic factors. However, further studies are required to analyse the optimisation of the lipase enzyme production by the filamentous *Aspergillus terreus* fungus.

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