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PHYSICOCHEMICAL PROPRIETIES OF LIPASE FROM NEWLY *STREPTOMYCES* SP. OLIVE POMACE ISOLATE

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ABSTRACT

Screening of new producing strains is required to meet the constant industrial demand for useful enzymes. In this context, an actinobacteria was isolated from the olive pomace, tested for extracellular lipase production, and identified by partial 16S rDNA sequencing. Then, the physicochemical characteristics of the enzyme are determined. Strain is a member of *Streptomyces* genus. Lipase was partially purified at 62.5 times, with a yield of 14.83% and a specific activity of 337.5 U/mg. Optimum activity was achieved at pH 7.0 and 60°C; it was completely preserved at pH 6.0 to 9.0, and more than 70% at 40 to 70°C, after 1 hour. The metal ions: K^+, Na^+, Ca^{2+}, Co^{2+}, Cu^{2+}, Fe^{2+}, Mg^{2+}, Mn^{2+}, and Zn^{2+} had no significant impact. Lipase activity was stable with ethylene diamine tetra acetic acid (EDTA), sodium dodecyl sulfate (SDS), H_2O_2, Triton x100, and Tween 80. Moreover, it was enhanced in butanol, chloroform, ethyl acetate, isopropanol, isobutyl acetate, hexane, petrol ether, and toluene. This enzyme can be suitable for various industrial applications.

Keywords: olive pomace, *Streptomyces* sp., lipase characterization, stability, stimulation

INTRODUCTION

Lipase enzyme catalyzes the hydrolysis of long-chain triacylglycerides, to yield diglycerides, monoglycerides, fatty acid, and glycerol **(Chandra *et al.*, 2018; Enespa and Kumar, 2020; Yao *et al.*, 2021)**. The reaction proceeds at the interface, between immiscible phases **(Chandra *et al.*, 2020; Ali *et al.*, 2023)**. In addition, lipases have a large scale of substrate specificity and stability. These factors have attracted much attention for a wide range of industrial applications **(Yao *et al.*, 2021; Saadoun *et al.*, 2023)**. Lipases are, therefore, the third most commonly used enzymes after proteases and amylases **(Mohamed *et al.*, 2021; Yao *et al.*, 2021)**. Despite their diverse origins (plant, animal, and microorganism), microbial lipases have a higher catalytic activity with a diverse specificity, making them widely used in biotechnological and organic chemistry applications **(Sharma and Kanwar, 2014; Javed *et al.*, 2018; Yao *et al.*, 2021)**.
Streptomyces species are recognized as the most important producers of bioactive molecules, including a variety of hydrolytic enzymes **(Sevillano *et al.*, 2016; Saadoun *et al.*, 2023)**. These bacteria play an effective role in the biological control of a wide range of substrates in different ecosystems, giving them great potential in several industries **(Rodríguez-Fonseca *et al.*, 2021)**. Lipolytic enzymes from *Streptomyces* have been reported to be applicable in the food, biodiesel, detergent, pharmaceutical, cosmetic, leather, textile, and paper industries **(Kumar *et al.*, 2016)**.
The catalytic reaction is critically influenced by the conditions of the reaction environment, notably pH and temperature, metallic ions, inhibitors, surfactants, and organic solvents. Enzymes from different strains have different properties; it is, hence, mandatory to define optimal activity conditions and critical requirements for each industrial process **(Yao *et al.*, 2021)**. Moreover, the ongoing industrial demand for new products requires the search for new enzyme molecules with interesting properties; the most efficient strategy is screening and exploiting new producing strains from different natural environments.
The present study describes the physicochemical characterization of the extracellular lipase produced by a new *Streptomyces* strain isolated from olive mill waste collected from a region in eastern algeria.

MATERIAL AND METHODS

Strain isolation, lipase test, and identification

An actinobacteria strain OP1 was isolated on ISP_2 medium from an olive pomace sample collected in December 2020 from an oil mill located in the Mila region of eastern Algeria. It was screened for lipase production on Rhodamine-B agar and was found to be potent with high lipolytic activity. The strain was identified by 16S rDNA sequence analysis; DNA extraction was performed by **Wilson (1987)** method; PCR was achieved with universal primers S-D-Bact-0008-c-S-20 and S-D-Bact-1495-a-A-20 **(Khessairi *et al.*, 2014)**; sequence analysis was performed by Big Day terminator cycle sequencing kit V3.1, using capillary electrophoresis ABI Prism 3130TM DNA sequencer (Applied Biosysthems, Hitachi, Japan); and the sequence similarities were analyzed with BLAST (*https://blast.ncbi.nlm.nih.gov/*). The phylogenetic tree was constructed using MEGA version 11 (*https://www.megasoftware.net/*).

Enzyme production and assay

The strain was inoculated into a previously optimized production medium containing (%): olive oil 1, peptone 1.32, $(NH_4)_2SO_4$ 1.41, pH 7.0, and was incubated at 35°C for 5 days on a rotating agitator at 150 rpm. The fermented broth was broken by centrifugation at 5000 rpm for 30 min at 4°C, was filtered on 0.22 µm cellulose ester filter Millex® (Merk, Massachusetts, USA), and was used as a crude extracellular enzyme.
The lipase activity was determined according to **Jensen, (1983)** titrimetric analysis method with some modifications in volumes. 1 mL of enzyme was mixed with 10 mL of 10% olive oil emulsified in gum arabic (Sigma Chemical, St. Louis, USA). The reaction mixture was incubated on a shaker at 150 rpm for 30 min. 20 mL of ethanol-acetone (1:1v/v) was then added to stop the reaction. Released fatty acids were neutralized with 0.05 mol/L NaOH at pH 10.0 using phenolphthalein (Sigma Chemical, St. Louis, USA) as a color indicator. The blank reaction was similarly manipulated outside the enzyme. The unit of lipase was the amount that released one µmol of fatty acids per minute and was expressed as U/min/mL.

Partial purification and molecular weight estimation

The proteins with lipase activity were precipitated at 75% ammonium sulfate (Sigma Chemical, St. Louis, USA) of saturation; the precipitate was suspended in phosphate buffer (pH 7.0) and was, then, dialyzed. The enzyme preparation was loaded on diethylaminoethyl (DEAE) Sephadex column (10 x 1cm) (Sigma Chemical, St. Louis, USA), and eluted with a linear gradient from 0 to 1.5M NaCl (0.05M). The active fractions were collected, pooled and ultracentrifuged with 5 kDa cut-off using Amicon® filters (Merk, Massachusetts, USA). The total protein was determined by **Lowry *et al.* (1951)** method using bovine serum albumin from 0 - 0.1 mg/mL, as the standard. However, throughout DEAE-chromatography, the protein elution profile was controlled spectrometrically at 280nm. The pooled active fractions were subjected to electrophoretic migration in Tricin SDS-polyacrylamide gel electrophoresis at 12% **(Gagaoua *et al.*, 2014)**. The molecular weight was assessed using control proteins as markers: ß-Lactoglobulin (18.4 kDa), Actin (42 kDa), Glutamate dehydrogenase (53 kDa), Transferrin (76 kDa) and β-Galactosidase B (116 kDa). After separation, the gel was stained with 4.9

mM Coomassie Brilliant Blue R-250 (Bio-Rad, Strasbourg, France) diluted in 50% (v/v) ethanol and 7.5% (v/v) acetic acid, and then destained until a clear background was obtained.

Effect of pH and temperature

The enzyme was incubated at pH values of 5.0, 6.0, 7.0, 8.0, 9.0, and 10.0, adjusted with 0.2 M buffer solutions: citrate phosphate, sodium phosphate, and glycine NaOH. The lipase activity was, then, determined. The pH stability was determined in the above-mentioned buffers by incubation for 1 hour; the residual activity was calculated considering the initial activity as 100%. Similarly, the enzyme activity was determined at temperatures ranging from 40 to 70°C, and thermal stability was monitored for 1 hour **(Zerizer *et al*., 2016)**.

Effect of metal ions, inhibitors, oxidants, and surfactants

KCl, NaCl, $CaCl_2$, $CoCl_2$, $CuCl_2$, $FeCl_2$, $MgCl_2$, $MnCl_2$, and $ZnCl_2$ (Biochem Chemopharma, Cosne sur Loire, France) were tested for their effects on the enzyme activity by incubating the enzyme with 1 and 10 mM (final concentration) for 1 hour. Likewise, the lipase activity was assayed by incubating the enzyme for 1 hour with 1 and 10 mM (final concentration) EDTA and SDS, and with 1 and 10% (v/v) H_2O_2, Triton x100, and Tween 80 (ProLabo, Paris, France). The activity was calculated by its comparison with the additive-free control **(Zerizer *et al*., 2016)**.

Effect of organic solvents

The enzyme was maintained at an optimal pH and temperature for 1 hour and, additionally, exposed to 50% (v/v) organic solvent: butanol, chloroform, ethyl acetate, isopropanol, isobutyl acetate, hexane, methanol, petrol ether, and toluene (ProLabo, Paris, France). The lypasic reaction is then carried out according to the method described above, and the relative activity is calculated while considering the control without organic solvent treatment as 100% **(Zerizer *et al*., 2016)**.

RESULTS AND DISCUSSION

Strain identification

Comparative analysis of the partial sequence of 16S rDNA of the isolate (accession number OQ842263) with NCBI Blast assigned the isolate to the genus *Streptomyces* with up to 96% of similarities. Figure 1 shows the phylogenetic tree with the most closely related species. The strain is most closely related to *Streptomyces coelicoflavus* NBRC 15399T with 93% bootstrap support.

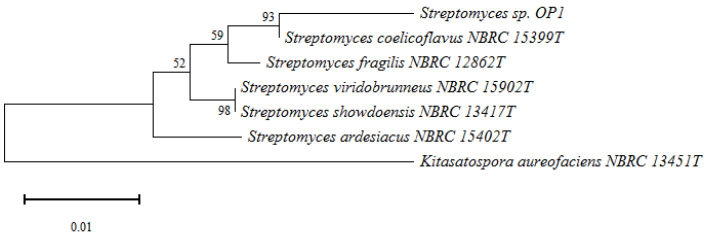


Figure 1 *Streptomyces* sp. OP1 phylogenetic tree bases on partial sequence 16S rDNA. Bootstrap supports are based on 100 replicates. *Kitasatospora aureofaciens* NBRC 13451T is used as outgroup.

Partial purification profile and molecular weight

A single peak was obtained after ammonium sulfate precipitation, DEAE-Sephadex chromatography and ultrafiltration purification steps (Figure 2A). A single band is also obtained by Trycin SDS-PAGE corresponding to 72 kDa (Figure 2B); this is probably the molecular weight of the lipolytic enzyme. The results of lipase purification (Table 1) show 62.5 fold, 14.83% of yield, and 337.5 U/mg of specific activity. In other studies, lipases were partially purified with similar steps, without ultrafiltration, *Trichosporon asahii* lipase provided 2.14 fold, 15.64% of yield, and 108.26 U/mg specific activity **(Kumari and Gupta, 2012)**, *Acinetobacter haemolyticus* TA 106 lipase provided 10.44 fold, 6.36% of yield, and 70 U/mg specific activity **(Jagtap and Chopade, 2015)**, *Pseudomonas aeruginosa* LX1 lipase gave 4.3 fold and 41.1% of yield **(Javed *et al*., 2018)**. Elsewhere, lipases from *Streptomyces bambergiensis* OC 25-4, *Streptomyces* sp. OC 119-7, *Streptomyces lienomycini* 350-2 have been purified by ammonium sulfate precipitation and gel filtration chromatography, from 1.56 to 5.52 folds, with 0.53-3.43% overall yield, and 92.12-1466.8 specific activity U/mg **(Ayaz *et al*., 2014; Ugur *et al*., 2014)**. Lipases are reported to be in general monomeric proteins **(Sharma *et al*., 2011; Chandra *et al*., 2020)**. *Streptomyces* lipases have a very wide range of molecular weights, and they are most in the range of 27 to 63 kDa **(Zhang *et al*., 2008; Cho *et al*., 2012; Yuan *et al*., 2015; Rajanikanth and Damodharam, 2017; Gao *et al*., 2020; Wahidi *et al*., 2021)**. Whereas, the molecular weight of bacterial lipases ranges from 16 to 120 kDa **(Saxena *et al*., 2003; Javed *et al*., 2018)**. The high levels of lipase molecular weight were isolated from, *Bacillus* sp.THLO27 (69 kDa) **(Sharma *et al*., 2011)**, *Pseudomonas gassardii* (92 kDa) and *Halobacillus* sp. strain LY5 (96 kDa) **(Javed *et al*., 2018)**.

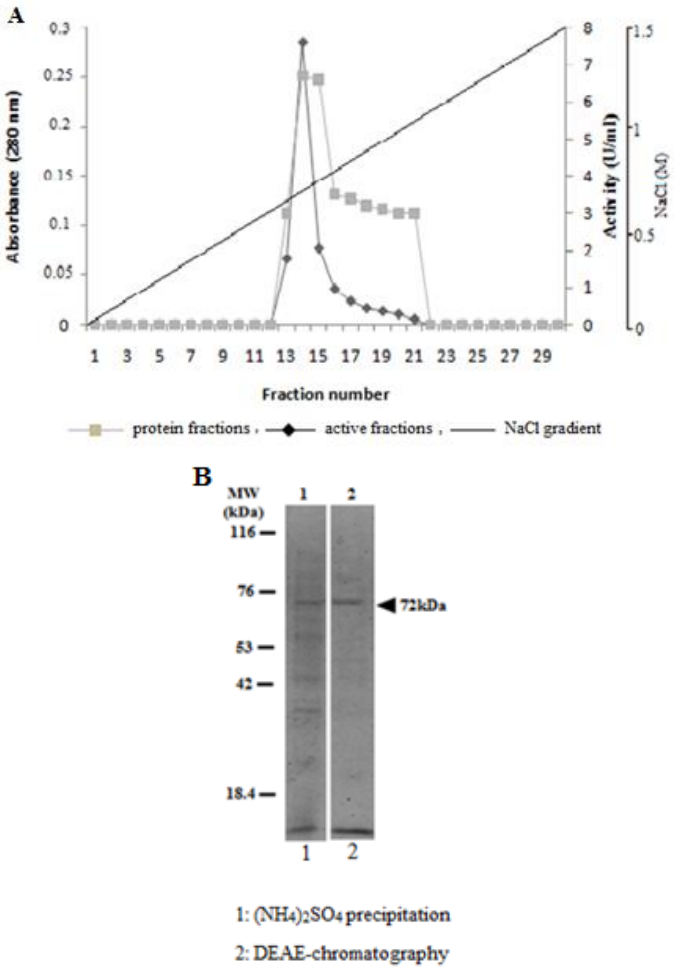


Figure 2 Lipolytic enzyme profile on ion exchange chromatography and electrophoresis

Table 1 Proteins and lipolytic activity of purification steps

Step of purification	Total protein (mg)[a]	Total activity (Units)[b]	Specific activity (Units/mg protein)	Recovery (%)	Fold
Supernatant	370	2000	5.40	100	1
75 % $(NH_4)_2SO_4$ and dialysis	37.28	1857.6	49.83	92.88	9.23
DEAE Sephadex and ultrafiltration	0.88	297	337.5	14.85	62.5

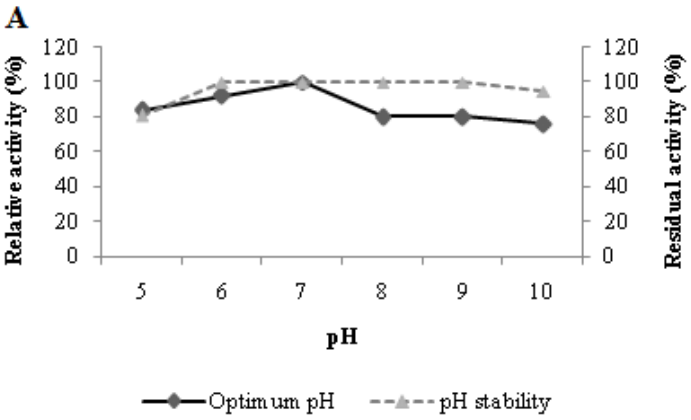
[a]Protein quantification method of **Lowry *et al*. (1951)**, [b]Activity quantification method of **Jensen (1983)**

pH, temperature, optimum, and stability

The *Streptomyces* sp. OP1 lipase shows an optimum activity at pH 7.0. The lipase enzyme preserves 100% of their activity at pH between 6.0 to 9.0 and retains over 80% of residual activity at pH 5.0 and 10 (Figure 3A). The maximum activity of *Streptomyces* sp. OP1 lipase is at 60 °C. Although, 73 to 93% of residual activity remains after 1 hour of the incubation period at 40, 50, 60 and 70 °C (Figure 3B). The best activity at pH 7.0 reveals that this enzyme is member of the neutral lipase group. The optimum pH of *Streptomyces* lipases is usually neutral or alkaline; *Streptomyces* sp. CS326 and *Streptomyces* sp.W007 produces neutral lipases with an optimum at 7.0 **(Cho *et al*., 2012; Yuan *et al*., 2015)**. Lipases derived from *Streptomyces bambergiensis* OC 25-4, *Streptomyces* sp. OC119-7, *Streptomyces* sp. AU-1, *Streptomyces violascens* OC125-8, and *Streptomyces pratensis* MV1 have an optimum pH of 8.0 **(Ayaz *et al*., 2014; Ugur *et al*., 2014; Boran *et al*., 2019; Wahidi *et al*., 2021)**. Similarly, *Streptomyces fradiae* var. k11, *Streptomyces cellulosae* AU-10, and *Streptomyces bacillaris* lipases have an optimum value of 9.0 **(Zhang *et al*., 2008; Boran, 2018; Gao *et al*., 2020)**; *Streptomyces* sp. SBLWN_MH2 produces a highly alkaline lipase with an optimum level at 10. In

this study, the lipase remains most stable under all the tested pH values. Moreover, in the acidic to neutral range, (pH 5.0, 6.0 and 7.0) the stability is better. The same results are achieved with *Streptomyces fradiae* var. k11, *Streptomyces* sp. OC119-7, *Streptomyces violascens* OC 125-8, and *Streptomyces pratensis* MV1 that maintains between 60-100% of their residual activity at interval pH acid to alkaline **(Zhang *et al*., 2008; Ayaz *et al*., 2014; Boran, 2019; Wahidi *et al*., 2021)**. The enzymes with these characteristics are attractive and have high potential for different industrial applications **(Rajanikanth and Damodharam, 2017)**. On the other hand, in most literature cases, stability was not preserved at a wide pH range, the enzymes were generally stable in the optimal average value; this is the case of *Streptomyces fungicidicus* RPBS-A4 and *Streptomyces pratensis* MV1 both retain a whole activity at the optimum pH **(Rajanikanth and Damodharam, 2017; Wahidi *et al*., 2021)**. Likewise, *Streptomyces bacillaris* and *Streptomyces coelicolor* A3 (2) lost a maximum activity outside the optimum pH **(Côté and Shareck, 2008; Gao *et al*., 2020)**.

The lipolytic enzyme from *Streptomyces* sp. OP1 is thermoactive. Previously extracellular lipases from *Streptomyces* sp. were reported with the optimum activities extending from 25 to 55°C and were found stable until 100% in their optimum temperature and maintained from 40 to 80% of residual activity at 40-70°C **(Côté and Shareck, 2008; Zhang *et al*., 2008; Cho *et al*., 2012; Ayaz *et al*., 2014; Ugur *et al*., 2014; Yuan *et al*., 2015; Rajanikanth and Damodharam, 2017; Boran, 2018; Boran *et al*., 2019; Gao *et al*., 2020; Wahidi *et al*., 2021; Ali *et al*., 2023)**. The authors have, hence, recommended thermoactive lipases in diverse industrial applications; it has been proposed as detergent ingredients, since oily stains are difficult to remove at low temperatures **(Boran, 2018; Boran *et al*., 2019)** ; as an alternative for the concentration of essential polyunsaturated fatty acids in human diet from vegetable oils **(Wahidi *et al*., 2021)**; as a modern clean energy generator in the biodiesel industry **(Ugur *et al*., 2014; Saadoun *et al*., 2023)**; and for flavor cheese concentration when the liberation of fatty acids is higher **(Chandra *et al*., 2020)**.



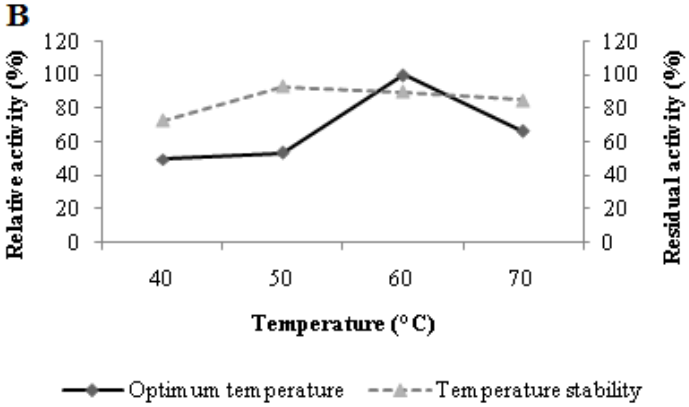


Figure 3 Lipase activity and stability at different pH (A) and temperature (B)

Metal ions, inhibitors, oxidants, and surfactants preserved activity

The catalytic activity of *Streptomyces* OP1 lipase was reserved for more than 87% with the tested ions, including K^+, Na^+, Ca^{2+}, Co^{2+}, Cu^{2+}, Fe^{2+}, Mg^{2+}, Mn^{2+}, and Zn^{2+}; however, it did not increase above 100%. Moreover, approximately 100% of the activity was retained after incubation with the oxidizing agent H_2O_2, inhibitors (EDTA and SDS) and surfactants (Triton x100 and Tween 80) (Table 2).

It has been reported that for the expression of the lipase's activity, metal ions are not required as cofactor **(Chandra *et al*., 2020)**, and can, however, alter the activity **(Yao *et al*., 2021)**. In the literature, *Streptomyces* lipases have presented a varying tolerance to metal ions; Ca^{2+} stabilize lipolytic enzymes of *Streptomyces* sp. W007, *Streptomyces fungicidicus* RPBS-A4 and *Streptomyces bacillaris* **(Yuan *et al*., 2015; Rajanikanth and Damodharam, 2017; Gao *et al*., 2020)**; but, they inhibit significantly the lipase activity of *Streptomyces fradiae* var. k11, *Streptomyces bambergiensis* OC 25-4, *Streptomyces cellulosae* AU-10 and *Streptomyces violascens* OC 125-8 **(Zhang *et al*., 2008; Ugur *et al*., 2014; Boran, 2018; Boran *et al*., 2019)**; Zn^{2+} activated lipase of *Streptomyces bambergiensis* OC 25-4, but Co^{2+}, Cu^{2+}, Na^+, Mg^{2+}, and Mn^{2+} suppressed their activity **(Ugur *et al*., 2014)**; Fe^{2+} increases considerably the activity of *Streptomyces cellulosae* AU-10 and *Streptomyces violascens* OC 125-8 lipase **(Boran, 2018; Boran *et al*., 2019)**; K^+ improves the lipase activity of *S. bacillaris* **(Gao *et al*., 2020)**.

The *Streptomyces* sp. OP1 lipolytic activity is not suppressed by the chelating agent ethylene diamine tetra acetic acid (EDTA), which accords with ion effect results; therefore, metal ions do not bind an active site. It can be supposed that the enzyme is not metalloprotein, although some *Streptomyces* sp. produces metallo-lipases **(Côté and Shareck, 2008; Ugur *et al*., 2014; Boran *et al*., 2019;)**. Sodium dodecyl sulfate (SDS), as a choatropic agent, disrupts non-covalent interactions of protein; no significant effect of SDS on the lipase activity, assuming that the catalytic active site of lipase is essentially maintained by disulfide linkage. Triton x100 and Tween 80 reduce the interfacial region between water and oil; thus, increase the enzyme access to the substrate, and stabilize emulsification, especially on the insoluble substrate **(Côté and Shareck, 2008; Boran *et al*., 2019)**. However, different effects of Triton X-100 and Tween 80 on the lipase activity have been previously observed, *Streptomyces violascens* OC 125-8 lipase activity was resistant **(Boran *et al*., 2019)**. In contrast, *Streptomyces pratensis* MV1 lipase was stimulated **(Wahidi *et al*., 2021)**, *Streptomyces* sp.W007 and *Streptomyces bacillaris* lipases were decreased **(Yuan *et al*., 2015; Gao *et al*., 2020)**. *Streptomyces* sp. OP1 lipase stability in the presence of H_2O_2, EDTA and surfactants is an interesting fact, as these agents are active components in detergent formulation; they proceed in synergy with the enzymes to enhance detergency **(Bisht *et al*., 2013; Rajanikanth and Damodharam, 2017; Boran, 2018)**.

Table 2 Metal ions, inhibitors, and surfactants effect on the lipase activity

Agent	Concentration	Residual activity (%)	Agent	Concentration	Residual activity (%)
KCl	1 mM	94.26 ± 0.00	$MnCl_2$	1 mM	90.44 ± 0.00
	10 mM	94.85 ± 0.17		10 mM	95.59 ± 0.07
NaCl	1 mM	90.88 ± 0.05	$ZnCl_2$	1 mM	93.97 ± 0.02
	10 mM	93.40 ± 0.10		10 mM	93.38 ± 0.10
$CaCl_2$	1 mM	100.00 ± 0.00	EDTA	1 mM	102.50 ± 0.04
	10 mM	94.85 ± 0.10		10 mM	106.66 ± 0.07
$CoCl_2$	1 mM	91.17 ± 0.14	SDS	1 mM	98.33 ± 0.07
	10 mM	99.26 ± 0.10		10 mM	105.33 ± 0.08
$CuCl_2$	1 mM	95.14 ± 0.02	H_2O_2	1 %	104.16 ± 0.03
	10 mM	92.35 ± 0.01		10 %	103.50 ± 0.00
$FeCl_2$	1 mM	87.50 ± 0.10	Triton x100	1 %	102.50 ± 0.03
	10 mM	96.61 ± 0.02		10 %	108.83 ± 0.05
$MgCl_2$	1 mM	90.14 ± 0.09	Tween 80	1 %	100.00 ± 0.00
	10 mM	94.11 ± 0.14		10 %	104.66 ± 0.10

Activity without any effectors was considered as 100%, results are expressed as mean ±SD

Organic solvents stimulate activity

Streptomyces OP1 lipase activity is enhanced in the presence of 50% (v/v) butanol, chloroform, ethyl acetate, hexane, isobutyl acetate, isopropanol, petrol ether, and toluene, with a relative activity more than 113%; however, the activity decreases to 69% in methanol (Table 3). Lipases have a range of organic solvents reactivity; they are, typically, more inactivated in polar than non-polar ones **(Kumar *et al*., 2016)**. For this, much research has focused on screening the enzymes that possess high activity in various organic solvents. In this study, the stability and the increased reaction rate of *Streptomyces* sp. OP1 lipolytic enzyme can be, probably, explained by the impact of organic solvents on the disorganization of aggregates formed between lipids and enzymes, or between individual enzyme molecules **(Guncheva and Zhiryakova, 2011; Kumar *et al*., 2016)**, or by several hydrogen bonds forming with water in the relational area, resulting inflexibility and conformational mobility **(Bisht *et al*., 2013)**. Otherwise, the interaction of non-polar solvents with hydrophobic amino acids, available at the catalytic site, maintains the enzyme in an open conformation and also modifies the substrates' solubility in the reaction environment, which is favorable to catalysis **(Guncheva and Zhiryakova, 2011; Rmili *et al*., 2020)**. The active lipase in organic solvents provides numerous advantages **(Ali *et al*., 2023)**. We have particularly reviewed that the enzyme can be used directly within a chemical process, and it is possible to regain it after reaction since it is not dissolved. In addition, the organic solvent improves the solubility of the substrate and product. Similarly, it is simple to remove them from the reaction environment when they have a lower boiling point than water. Organic solvents can also remove microbial contamination **(Sharma and Kanwar, 2014; Kumar *et al*., 2016)**. Interesting organic solvents tolerant microbial lipases are documented. For example, in the presence of methanol, isopropanol, and ethanol *Staphylococcus capitis* SH6 immobilized lipase retains around 90% of its initial activity **(Rmili *et al*., 2020)**; lipases from *Bacillus* spp. and *Acinetobacter* sp. AU07 are resistant in non-polar organic solvents, and their activity tends to increase considerably in hexane and toluene **(Guncheva and Zhiryakova, 2011; Gururaj *et al*., 2016)**.

Table 3 Organic solvents effect on the lipase activity

Organic solvent	Relative activity (%)
Butanol	113.42 ± 3.27
Chloroform	136.10 ± 7.85
Ethyl acetate	296.77 ± 4.90
Hexane	119.91 ± 5.88
Isobutyl acetate	126.85 ± 5.23
Isopropanol	134.90 ± 9.56
Methanol	69.44 ± 3.27
Petrol ether	124.07 ± 1.31
Toluene	123.14 ± 2.22

Activity without organic solvents is considered as 100%, results are expressed as mean ±SD

CONCLUSION

Mill waste can host new strains with significant lipolytic activity. *Streptomyces* sp. OP1 isolate produce an extracellular lipase with many industrial desirable proprieties. The maximum activity at acid to neutral pH and at high temperature, the stability in thermal, acidic, and alkaline conditions, in addition to oxidizing, inhibitor, and surfactant agents resistance are all extremely attractive characteristics of this enzyme. Enzymes that operate effectively under difficult conditions, such as the wide range of pH, the elevated temperature, and the addition of denaturizing agents, are required to be exploited in a number of different processes. Therefore, *Streptomyces* sp. OP1 lipase can be applied, for example, to boost the flavor of cheese, to remove fat from meat products, or as a detergents component to remove lipid stains, or even as a surfactant or emulsifier in cosmetic and perfume preparation, etc. Furthermore, organic solvents tolerant feature is an interesting characteristic of this enzyme, since there are a lot of fatty acid esters employed in food, detergent, cosmetic, pharmaceutical, and chemical industries that are produced in organic media.

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