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Lipase and their different industrial applications: A review

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Abstract. Enzymes are also known natural catalysts. Lipases are flexible enzymes that are mostly used. These enzymes are found extensively all over the animal and plant kingdoms, likewise in molds and bacteria. Among all identified enzymes, lipases have concerned the mainly biotechnological attention. This review paper discusses the characteristic, microbial origin and application of lipases. The present review discussed about different characteristics and sources (fungal, bacteria's) of lipase. The present article also discussed about different bioreactors used for lipase production and different biotechnological applications (food, detergent, paper and pulp, biofuels etc) of lipases. An observation to considerate lipases and their applications as bulk enzymes and high-value of production, these enzymes are having huge impact in different bioprocesses.

Keywords: Lipase; Microbial source; Bioreactors; Industrial application.

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Introduction

The interest for biotechnological enzymes, especially from microbial origin, is constantly growing due to their applications in a broad diversity of processes. Lipases are a central group of biotechnologically applicable enzymes and they find enormous applications in detergent, pharmaceutical food and dairy industries.

Lipases (triacylglycerol acylhydrolases EC: 3.1.1.3) are produced from a variety of living organisms such as plants, animals, bacteria, fungi, yeasts, actinomycetes. Amongst all, microbial sources of lipase have gained industrial attention in past few years because of unique properties and chemical stability (Abada, 2008).

General characteristics of lipase

Lipase is a pervasive enzyme which is essential for physiological importance and industrial uses. Lipases accelerate the breakdown of triglycerides to glycerol and free fatty acids. In addition, lipase causes the breakdown and transesterification esters at the same time, synthesis of esters also take place (Thakur, 2012).

They are dissolved in water and unable to dissolve in polar lipolytic. In 1856, the 1st lipolytic enzyme was discovered by Claude Bernard and after that it was recognized from animals, plant and microorganism (fungus, bacteria and yeast).

Lipase catalyzed reaction

Lipases are the most pliant biocatalyst and convey broad series of biotransformation reactions, i.e., hydrolysis, esterification, interesterification, aminolysis, alcoholysis and acidolysis (Kumar and Gupta, 2008).

In inclusion, lipase has long-established ability to stimulate breakdown of lipid, they can also stimulate synthetic reactions, for example transesterification (Shariff et al., 2007). Lipase catalyzed reactions into three most important classes according to previous report (Devi et al., 2012):

(i) Hydrolysis: this reaction occurs in the presence of high amount of water, where breakdown of ester bond take place. This technique is currently used in the formation of fatty acids, monoglycerides, diglycerides, flavouring substance for detergents and dairy products.

(ii) Esterification: It happens under little aqueous environment (anhydrous solvents). High quantity of the products (esterified compounds) is achieved under regulated environment. Formation of Primary (oleic acid esters) and Secondary (aliphatic and terpenic) are most common example (Macrae and Hammond, 1985).

(iii) Transesterification: It includes the exchange of acid moiety between two or more compounds. According to the type of substrates, lipases are able to catalyze acidolysis (it involve ester and carboxylic acid), alcoholysis (it involve ester and an alcohol), aminolysis (ester is allowed to react with an amine) and interesterification (where two acyl groups are exchanged between two esters) (Bajpai and Tyagi, 2007).

Lipase properties

Since 1980s, the number of obtainable lipase has been increased and used as biocatalysts in industries because of their different properties like pH dependency, high catalytic state, temperature, bio-degradability and high specificity (Masse et al., 2001). Amyl, isoamyl, isobutyl and ethyl are mostly used as flavor ester (Vaseghi and Najafpour, 2014). Lipase investigated in two sources hydrolytic and synthetic activity. Lipase has ability to consume mono-, di-, triglycerides and free fatty acids which is the most preferred characteristics (Klibanov, 1997). On the other hand, in mild condition of pH and temperature reaction of lipases is carried out and in abnormal temperatures and pressures energy is reduced for direct reactions. Due to this, unbalanced products and reactants are confined from destruction. Other reasons of lipases are they can be activated without any cofactor and stability in organic solvent (Houde et al., 2004).

Occurrence of lipase

The microbial origin of lipase is characterized as the most widely used group of enzyme in organic chemistry and biotechnology. Table 1 showed list of different microbes for lipase production. These enzymes are generally found throughout the plant and animal kingdoms, besides in bacteria and mold (Hiol et al., 1999). Lipase enzymes are found in both prokaryotes (Archaea and Bacteria) as well as in eukaryotes (plant, animal and fungi). Lipase of the microbial origin and animal are used widely. Lipase produce from pig pancreas contain trypsin which is a bitter tasting amino acid therefore, due to above mentioned reason, microbes are the preferred choice for the production of 100 or so industries (Jaeger and Reetz, 1998)

Although many micro organism are recognized as potent producers of extracellular lipase including fungi, yeast and bacteria.

Table 1. List of different microorganisms for lipase production.

	Microorganism	Source	Reference
1.	*P. fluorescens*	Bacterium	Pandey et al. (1999)
2.	*P. pseudomallei*	Bacterium	Kanwar and Goswami (2002)
3.	*Staphylococcus aureus*	Bacterium	Simons et al. (1996)
4.	*P. luteola*	Bacterium	Arpigny and Jaeger (1999)
5.	*Rhizopus sp.*	Fungus	Thakur (2012)
6.	*Pencillium sp.*	Fungus	Shelatkar and Padalia (2016)
7.	*Geotricum sp.*	Fungus	Thakur (2012)
8.	*Aspergillus sp.*	Fungus	Cihangir and Sarikaya (2004)
9.	*Candida rugosa*	Yeast	Vakhlu and Kour (2006)
10.	*Yarrowia lipolytica*	Yeast	Amaral et al. (2007)
11.	*Rhodotorula mucilaginosa*	Yeast	Kumar and Gupta (2008)
12.	*C. cylindracea*	Yeast	D'Annibale et al. (2006)

Bacterial lipase

In Malaysia, thermostable lipase was reported by strain L2 which was thermophilic bacterium isolated from hot spring. The extracellular lypolytic activity was done by broth and plate assays at 70 °C after 28 h (Selvamohan et al., 2012) Bacterial lypolytic enzyme may be membrane bound, intracellular and extracellular. *Bacillus clausii* which can only grow on simple lipids and glycerol but not on triglycerides long chain are the intracellular lipase. *Bacillus* sp. can produces both extracellular and intracellular lipases (Sharma et al., 2016).

Among bacterial lipase, those obtained from *Bacillus* spp. Show attractive features that make them potent candidates for various biotechnology based industries. *Bacillus pumilus* (Coradi et al., 2013), *Bacillus subtilis* (Coradi et al., 2013), *Bacillus licheniformis* (Romo-Sanchez et al., 2010), *Bacillus stearothermophilus* (Abada, 2008) and *Bacillus amyloliquefaciens* (Sangiliyandi and Gunasekaran, 1996) are the potential sources of bacterial lipases. Further, *Pseudomonas aeruginosa*, *Pseudomonas* sp. (Mirón et al., 2010), *Bacillus cepacia* (Ogino et al., 2000), *Staphylococcus pasteuri* (D'Annibale et al., 2006), *Burkholderia multivorans* (Gopinath et al., 2002) and *Staphylococcus caseolyticus* (Verma et al., 2012) has also been reported as bacteria lipase producers (Sztajer et al., 1998).

Fungal lipase

Funguses are commercially used in the production of lipases. Industrial waste, dairy plants, Vegetable oil processing factories and oil seeds or contaminates soil are some common sources of the lipase producing microorganism. The maximum lipolytic activity is shown by Rhizomes species (Sharma et al., 2016). Filamentous fungi are preferred sources of lipase among the rest the lipase producing. micro organism the main producers of lipase are *Pencillium sp*. (Gupta et al., 2003), *Rhizopus* sp. (Marlot et al., 1985), *Aspergillus* sp. (Abrunhosa et al., 2013), *Mucor* sp. (Gutierrez et al., 1996), *Acremonium alcalophilum* (Pandey et al., 1999), *Candida rugosa* (Pereira et al., 2013), *Lipomyces starkeyi (*Sumita, 2012), *Cunninghamella verticillata (*Divakar and Manohar, 2007), *Trichoderma* sp. (Clark et al., 1984) and *Geotrichum candidum* (Buchon et al., 2000).

Yeast lipase

Candida rugosa is one of the most rapidly used industrially important enzymes due to its high activity in both hydrolysis as well as synthesis phase (Ramos-Sanchez et al., 2015) . In 1985, *Candida rugosa* is used by Japan for the production of fatty acids from castor bean (Macedo et al., 2003). Earlier it has been reported for use of microbial lipase in the flavoring of milk and creams production (Padilha et al., 2012). Organolephtic has proved that every lipase strains develop a characteristic flavor (Padilha et al., 2012). *Saccharomyces*, *Torulospora*, *Kluyveromyces*, *Candida*, *Pseudozyma*, *Pischia*, *Lachancea* and Z*ygosaccharomyces* are the lipase producing yeast belong to different genera (Rajendran et al., 2008).

Different bioreactor used in lipase production

Tray bioreactors (TB)

Tray bioreactors made up of a chamber where controlled air (cabin temperature, cabin flow, and cabin humidity) is distributed in different trays. Every tray combines a solid substrate layer, usually between 5 and 15 cm deep. These trays are generally open from the top and have holes in the bottom which support exchange of gases. With the help of hand steady mixing of the solid substrate could be carried out but it can be only perform once in a day (Menoncin et al., 2008). The drawback of tray bioreactors is their small volumetric efficiency in compare to column bioreactor. In the recent years, there is no significant use of tray design.

Packed-bed bioreactor (PBB)

Packed-bed bioreactor frequently have a static bed on top have holed plate from which conditioned air is blasted. In other packed -bed bioreactor design, air is blow through a hole rod placed in between of the bed. In last 25 years, PBB has get more modeling and experimental consideration. The most unique characteristic of PBB is that it has no moving parts (mechanical) hence it reduces the cost of maintenance, operation and construction. Axial dynamic temperature is observed in PBB processes (Aruna and Khan, 2014). Since, when the temperature the water carrying quantity of the air will increases, it provide the slope to rise evaporation inside the bed so when the water thoroughly soaked in the air is worn to aerate the column. Although, in cellular metabolism 65% of the heat produced is remove up by evaporation phenomenon, it decreases the moisture content of solid substrate as a result cellular growth is limited (Gutarra et al., 2009). In PBB, bed is not mixed due improper moisture regulation. Consequently, water jacket is the most advantageous to enhance heat removal.

The large-scale production are very less in grams of solids supports and insufficient in demands studied in recent reports (Berglund and Hutt, 2000). The production of lipase in various bioreactors design is noteworthy and yield is very low at small scale.

Applications of lipases

Lipase are broadly used in the different application like detergent making , detergents formulations, the synthesis of fine chemicals, food processing, cosmetics, bioremediation processes , pharmaceuticals, and paper manufacture (Verma and Kanwar, 2008). Lipase can be used to speed up the polyurethane and humiliation of fatty waste (Martinez-Ruiz et al., 2008). Table 2 showed different industrial applications of lipase. From fungi and bacteria mostly microbial lipases are produced.

Lipases in food processing

Oil and fat are main ingredients of food .in food processing industry, oil and fat modification is the most important part and it has great demand in green technologies (Gupta et al., 2007). Lipase has ability to enhance the place of fatty acid chains in the glyceride and replace them into new one.

In biotechnology, there are various industrial applications that result there are different biotech products that we are using in daily life at home. In food science application several enzyme exploit in the class of various foods. Lipases have vast application in food industries like in flavor development, EMC technology and cheese ripening (Martinez-Ruiz et al., 2008). It is also used in flavor and fragrance compounds which are addition in food to adjust flavor by synthesis of ester (fatty acids and alcohols) (Redondo et al., 1995).

Table 2 Industrial application of lipase.

Industry	Causes	Application
Bakery food, dairy food and food dressing	Flavor improvement, breakdown (cheese ripening, milk and fat), alteration of butter fat and Quality upgrade.	Prolongation phenomenon of flavoring agent in butter and milk cheese and mayo whippings and dressing.
Wellness food	Transesterification.	Health foods.
Cleaner	Minimize biodegradable strains.	Washing of strain from fabrics.
Surfactants	Replace phosphoilpases in the production of lysophospholipids.	In industrial detergent, carbohydrates fatty acid esters and polyglycerol used as stabilizer in food creation i.e., ice creams.
Pharmaceutical.	Breakdown of expolyester alcohols.	specialty lipids and digestive aids.
Agrochemicals.	Esterification.	Herbicides,i.e. phenoxypropionate.
Liquor.	Enhance aroma.	Undistilled fermented beverages such as beer, wine.
Fuel industries.	Transesterification.	Production of biodiesel.
Meat and fish.	Flavor development.	Removal of fat.
Pollution control.	breakdown and transesterification of oils and grease.	Removal of hard stains, and breakdown oil and greases.

Lipases in detergents

Enzymes are mostly used for formulation of detergent in developed countries. Different enzymes like protease, lipase, amylase, cellulose etc are used in detergent industries as they can split oil, fat, starch and protein. Due to capability of hydrolysis of fat and lipid, lipase is used in laundry industries and household detergent (Ashley et al., 1999). Laundry detergent instruct flexibility to fabrics, softness , dissolved in water, harsh to skins and eyes and anti staticness that's way it is commonly used in washing machine and becoming more popular.

Lipases in pulp and paper industry

Enormous amount of lignocelluloses biomass is processed every year by the pulp and paper industry. Historically, enzymes are used in the paper industry for limited uses such as modification of raw starch. In pulp technology, application of microbial enzymes is highly diverse and numerous opportunities. Pitch known as the insoluble components of wood (mostly waxes, triglycerides), which causes drastic troubles in pulp and paper manufacturing (Houde et al., 2004). Microbial Lipases are worn in removal of pitch since the pulp formed in paper manufacture. *Candida rugosa* fungal lipase is used in the hydrolysis of 90% of the wood triglycerides or waxes it is the pitch control method developed by Nippon Paper Industries, Japan.

Lipases in organic synthesis

In the synthesis of organic chemical the use of lipase is becoming more and more important. Chemo-, region- and stereoselective transformation in large variety can be accelerating by the use of lipase (Kademi et al., 2003). Microbial lipase worn as a catalysts in bulk amount in organic chemistry. The workings of these enzymes are based on hydrophilic - lipophilic interface. Previously it has been discussed about the enantiopure compounds, which are synthesized by the use of lipase (Balcão et al., 1996). These enzymes catalyze the breakdown of water-immiscible triglycerides at water-liquid interface.

In liquid interface, the enzymes accelerate the breakdown of water-immiscible triglycerides at water. Under these conditions, the direction of lipase catalyzed reaction depends upon the quantity of water in the reaction mixture. The process of transesterification and esterification is depending upon the amount of water. These processes occur when there is petite or no water and when there is excess of water then hydrolysis reaction take place (Kazlauskas and Bornscheuer, 1998). Catalysis of lipase is supercritical solvent based reaction (Thakur, 2012).

Lipase in bio fuels production

Bio fuels or biodiesels are the ester of long chain of fatty acids and small chain alcohols. Through direct transesterification of vegetable oils and fat with short chain of alcohols (i.e., ethanol and methanol) in the existence of proper catalyst synthesis of biodiesels molecules take place (Amaral et al., 2007).

Transesterification is process similar to breakdown of water but here instead of water, alcohol is employed. In this reaction, displacement of alcohol from an ester to alcohol takes place (Shirazi et al., 1998). In short chain reaction, methanol, alcohols and ethanol are frequently worn; above all, methanol is used regularly due to physicochemical compensation and low cost. This procedure is frequently worn to decrease the thickness of triglycerides, therefore increasing the physical property of renewable oil to perk up engine presentation (Cihangir and Sarikaya, 2004).

Lipases in pollution control

In biotechnology, lipase is Employment as the new aspect of bioremediation process. From different origins like restaurants and factories lipase can be used to clean up waste of lipid processing. Lipase could be used in both the ways either in situ or ex situ (Padilha et al., 2012). Environmental pollution and industrial is becoming critical more and more due to speedy development. In enzymological remediation, the lipase strains play important role in soil pollution (Lasoń and Ogonowski, 2010). Lipase shows unique properties in field of cold adaption like active compounds synthesis cold condition, bioremediation in fat contaminated and wastewater treatment in cold condition (Bhargav et al., 2008). At the same time in different regions, where temperature diminish, the competence microorganisms is degrading pollutants like lipids and oil. This enzyme is perfect for bioremediation process because it is active in moderate low temperature (Sztajer et al., 1998).

Conclusion

In industrial use of microorganism is still remaining partial by their production in small amounts, low performance in lipase mediated process and their high production costs. Even though lipases have different fascinating vast applications food processing, detergents, pulp and paper industry, organic synthesis and bio fuels production etc. the latest applications are still to be explore and the applications of lipases are expansion rapidly in these industries.

Conflict of interest

The authors declare that they have no conflict of interest in the publication.

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