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Original Article

Assay of Lipase producing bacteria isolated from cooking and automobile oil contaminated soils within Kaduna Metropolis

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SUMMARY

Lipases have become increasingly important in industrial biotechnology due to their broad range of applications in cosmetics, food, environmental remediation, and pharmaceuticals. Environmental factors like temperature and pH affect lipase activity, and more research is needed for cost-effective production and purification of lipase for industries. The production of daily life products consumes large amounts of raw materials and energy and generates large amounts of waste with an adverse impact on our environment and quality of life. The growing need for eco-friendly industrial processes and bioremediation strategies drives the search for effective lipase-producing microorganisms. Therefore, this review focuses on assessing the lipase producing potential of bacteria isolated from cooking and automobile oil-contaminated soil, leveraging environments rich in lipid substrates. By optimizing production methods and assessing pH and temperature effects, this research aims to offer a promising solution for green industrial processes, addressing pollution and sustainability challenges.

Keywords: Soil, Bacteria, Lipase, Assay, oil contaminated

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INTRODUCTION

The development of enzymology has been a breakthrough in biotechnology, enabling the widespread use of enzymes to support scientific and industrial innovation [1]. Enzymes serve as catalysts found in all living organisms; they are known for their high efficiency and specificity [2]. These protein molecules accelerate biochemical reactions by lowering activation energy, without undergoing permanent changes themselves [3]. Their role is essential in supporting life processes, as they convert substrates into products through catalytic action. Enzymes also contribute to sustainable practices by improving product quality, conserving resources, minimizing waste, and reducing environmental pollution [4,5]. Biotechnology provides a sustainable approach to enzyme production, offering an alternative to conventional chemical catalysts. This eco-friendly shift is reflected in the projected growth of the global enzyme market, expected to increase from $11.4 billion in 2021 to $17.88 billion by 2028 [4,6]. Enzymes are categorized based on their specific catalytic actions, with hydrolytic enzymes like proteases, amidases, esterases, and lipases dominating the commercial enzyme market [7]

Lipases have been isolated from diverse sources. However, there is limited understanding of lipases-producing bacteria in cooking and automobile oil-contaminated sites, and the untapped potential of their lipases for bioremediation and industrial applications. Therefore, this review focuses on assessing the lipase producing potential of bacteria isolated from cooking and automobile oil-contaminated soil within Kaduna metropolis using enrichment techniques. Identifying potent strains can enable large-scale, cost-effective lipase production, supporting cleaner industrial processes and bioremediation efforts.

Structure of Enzymes

The active site, a small region within this structure, is where substrate binding and catalysis occur. It includes a binding site for substrate recognition and a catalytic site where the reaction takes place [8].

Although most enzymes are proteins, some RNA molecules, called ribozymes, also have catalytic capabilities. The enzyme's function is largely determined by its amino acid sequence and structure [9].

Classification of Enzymes

Enzymes are categorized based on the reactions they catalyze, their structural features, or their site of activity. The most widely accepted classification is the Enzyme Commission (EC) system, developed by the International Union of Biochemistry and Molecular Biology (IUBMB), which divides enzymes into six main classes: Oxidoreductases (EC 1) – catalyze oxidation-reduction reactions, transferases (EC 2) – transfer functional groups, hydrolases (EC 3) – break bonds using water, lyases (EC 4) – remove groups to form double bonds or add groups to double bonds,isomerases (EC 5) – catalyze structural rearrangements within molecules, ligases (EC 6) – Join two molecules using energy from ATP. Other classification methods include substrate specificity (e.g., proteases, lipases, amylases), catalytic mechanism (e.g., covalent or metal-ion catalysis), and cellular location (e.g., cytoplasmic or membrane-bound enzymes) [9,10].

Lipases

Lipases (triacylglycerol acylhydrolases, E.C. 3.1.1.3) catalyze the conversion of triglycerides to free fatty acids and glycerol at an oil- water interface. They have esterolytic activity on distinct substrates [11]. They also play a crucial role in lipid metabolism, digestion, transport, and energy regulation [7]. They have gained significant attention recently due to advancements in enzyme technology [12]. Lipases are highly versatile, capable of performing various reactions including hydrolysis, esterification, alcoholysis, and interesterification. Lipases stand out due to interfacial activation at oil-water interfaces, high substrate specificity, functionality in organic solvents, pH and temperature stability and cofactor independence [4].

Biotechnological advancements have enabled the isolation and screening of microbial strains from diverse environments, such as oil-contaminated soils, for their lipase-producing potential. These enzymes are now widely used in food production, pharmaceuticals, cosmetics, and biofuel industries due to their efficiency and selectivity [13]. Types of lipases include Triacylglycerol lipases (TAG lipases), phospholipases (e.g., PLA1, PLA2), lipoprotein lipases (LPL), hepatic lipases (HL), endothelial lipases (EL). These enzymes are not only involved in digestion and energy metabolism but also play roles in inflammation, cell signaling, and environmental remediation [14].

Historical Development of Lipases

Lipase was first discovered by Claude Bernard in 1856 within pancreatic secretions. This discovery marked a foundational moment in the study of enzymes [3]. In the early stages of industrial application, lipases were extracted from plant seeds and animal pancreas. However, the rising demand and limitations in supply from animal sources prompted the exploration of microbial alternatives for large-scale production. This discovery marked a foundational moment in the study of enzymes [2,3].

Sources of Lipases

Lipases are sourced from plants (seeds, roots, leaves), animal tissues (pancreas, liver, etc.), and microorganisms [7]. Various microorganisms, including bacteria, yeasts, and molds, produce lipases with distinct enzymological features and specificities [15]. However, bacterial lipases are more economical and stable, gaining importance in biotechnology due to their ease of isolation, simplicity in genetic manipulation, potential for high yield production, absence of seasonal variations, ability to grow on low-cost media and minimal ecological toxicity [15].

Properties of Lipases

Since the 1980s, lipases have gained popularity as industrial biocatalysts due to their ability to act specifically on substrates, function in non-aqueous environments, and operate under moderate conditions. They are biodegradable, reusable (especially when immobilized), and resistant to alcohol and temperature fluctuations. Lipases are effective in transesterification reactions and show minimal inhibition by their products [3]. These characteristics reduce the need for extreme processing conditions, leading to energy savings and lower operational costs [3,4].

Conditions for Optimizing Lipase Production

The production of lipases is affected by nutritional and environmental factors, including carbon and nitrogen sources, lipid presence, salts, temperature, pH, and oxygen availability [16]. Carbon sources, especially oils and fatty acids, are major inducers of lipase synthesis. Nutrients such as sugars, amino acids, and trace elements also influence enzyme production. Optimizing these variables is crucial for maximizing yield and efficiency [3,4].

Chemical and Structural Characteristics of Lipases

Chemical perspective:

Lipases act on the ester bonds linking glycerol and fatty acid chains in triglycerides. The reaction involves the addition of water, which severs the bond and liberates fatty acids and glycerol [17].

Structural perspective:

Lipases exhibit a unique structural element—a "lid" or "flap"—which shields the enzyme's active site. This mobile structure, typically hydrophobic, plays a critical role in binding substrates like triglycerides and aligning them for catalysis. The enzyme's active site is composed of a catalytic triad—commonly serine, histidine, and either aspartic or glutamic acid—that facilitates the reaction. Lipases are known for their conformational adaptability, allowing them to interact with a broad range of substrate shapes and sizes [17, 18].

Applications of Microbial Lipases in Industry

The demand for industrial enzymes, particularly lipases and cellulases, is on the rise. Lipases are increasingly used in detergents and cosmetics, supported by advances in genetic engineering and recombinant DNA technology [1]. Their industrial relevance lies in their ability to drive biochemical transformations efficiently, making them valuable tools across a wide range of applications [7]. They primarily serve as biocatalysts in the synthesis of food ingredients and fine chemicals. Ranking third in sales after proteases and carbohydrases, lipases represent a multibillion-dollar global market [6]. Their ability to operate in both aqueous and organic environments has led to widespread use across numerous industries, replacing traditional chemical approaches and minimizing environmental impact. By using microbial enzymes, industries can reduce the consumption of raw materials, energy, water, and harmful chemicals, thus promoting cleaner and more sustainable production methods [19]

Lipases in Biosensors

Lipases are instrumental in biosensor technology and diagnostic assays due to their specificity. They are used to detect triglycerides and other biomarkers. Some inhibitors, such as CTAB, cadmium, and cobalt ions, have been found to reduce lipase activity from specific microbes. These enzymes are applied in environmental, food, and clinical diagnostics through electrochemical and optical sensing systems [7].

Lipases in Bioremediation

Bioremediation using lipases offers a sustainable method for dealing with pollutants like oil, plastics, and pesticides [20]. Microbial lipases help break down micro- and nanoplastics (MNPs), a growing ecological concern. Species such as *Bacillus amyloliquefaciens, Aspergillus clavatus, Pseudomonas fluorescens,* and *Candida cylindracea* are known to degrade plastics, making them promising tools in environmental cleanup efforts [1].

Emerging Trends in Lipase Engineering

Lipase engineering is advancing through genetic modification and scaling technologies. Techniques like rational design (RD) and molecular dynamics (MD) are used to improve enzyme stability, activity, and specificity. RD identifies critical amino acid residues for targeted mutation, while MD simulates enzyme behaviour under various conditions [7].

Directed evolution (DE), involving random mutagenesis and selection, allows the creation of lipase variants with improved traits such as heat resistance, pH tolerance, and catalytic performance. These advancements support the development of customized enzymes for industrial applications [4].

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