IMMOBILIZATION OF B-GLUCURONIDASE ON THE BIOMATERIALS/NANO PARTICLES AND IT’S INDUSTRIAL APPLICATIONS

Muhammad Waqas 1*, Zahid Mehmood 1, Khalid Mahmood 1, Mohammad Azam 1, Ghulam Mustafa Khan 2, Mohamamd Ibrahim 1, Amir Rasool 1 and Sheikh Ahmed 1.

1Institute of Bio-Chemistry, University of Balochistan, Quetta Pakistan.
2Department of Chemistry University of Balochistan Quetta Pakistan.

Abstract:
β-glucuronidase enzyme belongs to the hydrolase family which perform breakdown of complex carbohydrates. β-glucuronidases are large biomolecule consists of polypeptide chains and are widely included eukaryotes and prokaryotes, however their parasitic sources are extremely constrained. β-glucuronidase from fungal source could be profoundly valuable as a result of its high selectivity and stability over extensive variety of pH and temperature which makes it exceptionally reasonable for mechanical applications. Number of studies have been conducted to test the enzyme stability by immobilization which can easily be detached from biomaterials and nano particles for repeated use in the chemical reaction. β-glucuronidase is also comprehensively use in industry for protein catalyzed reactions. For this purpose enzyme was isolated and cloned from many sources such as Penicillium purpurogenum Li-3 ZnO-NP, sodium alginate, sepabeads etc. These studies revealed that β-glucuronidase shows maximum catalytic activity at pH below 7 after immobilization on biomaterials and on divalent metal ions. The catalyst turns out to be more effective and has higher fondness for the substrate.

Key words: β-glucuronidase, immobilization and polypeptide chains

Corresponding author:
Muhammad Waqas,
Institute of Bio-Chemistry,
University of Balochistan,
Quetta Pakistan

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INTRODUCTION:
The protein atoms formed by the body that includes in the chemical reaction to produce a particular desired product in minimum time by consuming enzymes. These protein particles are very much particular; they are required to coordinate a specific reaction with a particular result. They are arranged in an extensive variety of shapes and sizes because of its induced and frame fit properties. In the living organism enzymes are assumed as a dynamic and active role in regulating daily life. Enzymes are basic in the capacity that they are proteins proposed to increase the rate of chemical reaction and without them chemical reaction would continue at very slow rate. β-glucuronidase is arranged in the group of hydrolases enzyme. The known name of this class of enzyme is glycyrrhizinate glucuronosyl hydrolase. Diverse other basic names incorporates glycyrrhizin beta-hydrolase, glycyrrhizin hydrolase, hydrolase and glycyrrhizinic acid.

In the intestine glucuronidated metabolites are hydrolyses to their toxic form by the intestinal bacterial β-glucuronidase which results the damage of intestine. The advancement of a technique to restrain β-glucuronidase is in this manner critical however has been constrained by the trouble of straight forwardly surveying protein movement in live animals (1).

β-glucuronidase enzyme can be isolated from large variety of sources, such as prokaryotes and eukaryotes, fungi, bacteria, plants, animals and humans (2).

β-glucuronidase hydrolyses glucuronidated mixes, freeing glucuronic acid and the aglycone type that can be an alcohol, imine, or thiol. It is co-encoded with a glucuronide transporter, permitting glucuronide section in the microscopic organisms and its utilization as carbon source. Among them a huge number of animal varieties display in the human gut microbiota, a modest number (around 50 species) conveys qualities encoding β-glucuronidases. Two gatherings of glucuronidases are observed in view of amino acid successions (3) (4). The following chemical reaction is catalyzes by the enzyme glycyrrhizinate β-glucuronidase in enzymology.

Along that, two substrates of this compound are glycyrrhizinate and H2O, and its two items formed are 1,2-β-D-glucuronosyl-D-glucuronate and glycyrrhetinate. The best study of β-glucuronidase enzyme is extracted from Escherichia coli and also the crystallographic structure of this enzyme has been determined. The studies explained further about the β-glucuronidase is that biomaterials are important in obtaining maximum catalytic efficiency (5).

Applications of β-glucuronidase
β-glucuronidase is the enzyme that catalyzes the hydrolysis of the 1,2-β-D-glucuronosyl-D-glucuronate and glycyrrhetinate. In food industry, this enzyme is utilized to expel certain unwanted segments coming from the production procedure, and additionally to increase the value of the product. As well as it also upgrade the relative sweetness of the product (4). Apart from the above function in the food industry β-glucuronidase also uses in waste treatment systems (6). Human β-glucuronidase is a kind of glucuronidase that catalyzes hydrolysis of β-D-glucuronic acids delivered from the non-polar end of muco polysaccharides for instance, heparin sulfate. Human β-glucuronidase is arranged in the lysosome. In the gut, brush line β-glucuronidase changes the conjugated bilirubin to the unconjugated for re-digestion. The Immobilization of β-glucuronidase on nano particles or on biomaterials, for example, gelatin-cellulose bearer framework, epoxy /glyoxyl /Br CN gatherings, DEAE agarose gel, glutar aldehyde, silicon surface, polyelectrolyte surfaces, amino di acidic, metal chelates, and ethylene di amine increases its stability and reuse for Industrial purpose. In β-glucuronidase activity study there is an increasing economic interest (6) (7), this enzyme is used as one of the ingredient of toothpaste. It is also utilizes as the part of eye drops In pharmaceutical industry as well as in creams as antiseptic, In cosmetics industry, it is utilized for skin refresher, emulsions and facial cream. In food industry, it is used as sweetening, and as energy drinks, in
mammalian and plant cells as a reporter gene β-glucuronidase is used to monitor gene expression (8).

β-glucuronidase is used as a reporter gene for the expression in plant cells and mammalian cells. β-glucuronidases hydrolyze to expel the glucuronic corrosive from glycoside. It has been utilized as a part of the analysis of disease, pharmacotherapy and hereditary control. Its characteristic in change of the typical glucuronidase has pulled in unprecedented contemplations (10).

**Immobilization**

An immobilized biological catalyst is characterized as the enzyme joined to an inert, insoluble biomaterial or nano molecule, for example, sodium alginate (made by the blend of sodium alginate with calcium chloride)[9] this can give outright absorbability to change in conditions, for example, pH or temperature. It moreover permits strength in the compound response, taking after which they are effectively disconnected from the immobilized material or nano particles and might be utilized once more. It is additionally exhaustively utilized as a bit of industry for protein catalyzed responses. Another elective stable immobilization is entire cell immobilization (9).

A thermo stable β-glucuronidase gene bgaB have been cloned and expressed by the Chen et al. in from *B. stearo thermophilus* in *B. subtilis* WB600. The maximum temperature for this β-glucuronidase enzyme activity was about between 60 to 70°C. (10). it was additionally clarified that on the zinc oxide nano particles (ZnO-NP) the cleansed recombinant β-glucuronidase (PGUS - E) from Penicillium purpurogenum Li-3 was immobilized for the biotransformation of glycyrrhizin (GL). It was watched that PGUS-E regular chipping away at ZnO nano molecule was 6.52 U/mg with an ordinary of 85.83% immobilization item. The adsorption of the PGUS-E on ZnO-NP was confirmed by using the filtering electron magnifying instrument (SEM) and infrared (FTIR) spectroscope. The examination of synergist activity of the immobilized PGUS-E was surveyed in the ionic fluids (ILs) media and cushion. The more prominent reactant viability of the immobilized PGUS-E was recorded in the IL co-dissolvable medium than the support; and after 8 times of reuse of ideal 30.54% and 7.42% of its reactant activity was held. The recuperation rate of the IL medium ([Bmim] PF6) was as high as 76.11%. The estimation of the compound action parameters and active vitality also communicated the perversiveness of the IL co-dissolvable media over the mono phasic media (10).

Lauro have cloned, communicated, purged and described a β-glucuronidase (Aaβ-lady) from thermo acidophilic microscopic organisms *Ali cyclobacillus acido caldarius*. The recombinant Aaβ-lady is ideally dynamic and stable at 65°C (11).

Immobilization has appeared to enhance β-glucuronidase's stabilize and reuse. The immobilization of the β-glucuronidase of *Thermus sp.* was performed utilizing ionic adsorption onto two distinct backings: another anionic exchanger tar, in light of the covering of Sepabeads inside surfaces with polyethylenimine (PEI) polymers, and regular DEAE-agarose. Immobilization continued abnormally in the two cases, however the adsorption quality was considerably more prominent on account of PEI-Sepabeads than in DEAE-bolsters at both pH 5 and 7. (12).

For β-glucuronidase distinctive immobilizing biomaterials were examined for the enzymatic sensibility of the whole cells of *Penicillium* .p. The ideal polymers found for the biotransformation of glycyrrhizin (GL), into glycyrrhetic corrosive 3-O-mono-β - D-glucuronic (GAMG) were Li-3 (w-PGUS) and alginate gel. The activity of compositionally advanced w-PGUS alginate dabs were assessed in various ionic fluids (ILs) and synthetic responses were inspected and shown to express biocompatibility with alginate gel bolster. The ideal biotransformation conditions of the immobilized (w-PGUS) were analyzed, detached and after that contrasted and ionic fluid co-dissolvable media and support with dabs of (w-PGUS)quantity of 70 g/lit versus 80g/lit, substrate fixation (5mMvs.4mM), temperature (45 °C vs.45 0C), pH(5.4vs. 5.6), and shaking speed (180rpm vs.160rpm) were individually .(13)

There are constrained investigations has been done on the use of immobilized chemicals. Albeit more worthwhile utilization of immobilized proteins are there in which following two essential centered points of interest are

1. Simple separation of the compound from the thing.
2. Reuse of the chemical.
Immobilized chemicals are used as a piece of regular compound. Immobilization expands the unit of the steady protein and can cut down the costs of catalyst essentially. This is legitimate for immobilized protein courses of action that give all finished balanced execution, for example, if there should be an occurrence of immobilization produces, less mass trade restrictions, and expanded working steadiness. There are different systems accessible for immobilizations which are utilized to immobilize the catalyst on the biomaterials or on nano particles. The electrostatic restricting power between chemical to be immobilized and biomaterials or nano particles vary between frail different and single adsorptive connection through covalent bond (14).

The most amazing immobilization comes about were accomplished by utilizing glutaraldehyde as helpactivator and protein stabilizer. The streamlined chemical focus for immobilization was 15-20 mg g-1 of help (15). The use of calcium alginate (CA) and gellan - xanthan (GX) gel dots for entanglement of cells of Streptococcus thermo philus containing β-glucuronidase upgraded the security of compound at higher temperatures (>55 °C) (16). Hence, immobilization has appeared to enhance the dependability of β-glucuronidase and diminishes the handling time in sustenance and different industries.

The improvement of technological procedures and devices based of β-glucuronidase has caught the consideration of analysts and industry. While metals are activators of the compound and enhance its catalytic activity, also their effect on stability decides the best performance of such devices (17). The interest for modifying of the working conditions for this compound makes the closeness of metal an important factor (18)(19). The point of the recent study was to research the impacts of some biomaterials on the movement and compound and thermal stability of β-glucuronidase. (20)

CONCLUSIONS:
This literature mining study revealed the immobilization of the enzyme that is β-glucuronidase on the different biomaterials and the changes occur in activity of enzyme after immobilization. The various possible materials for the immobilization include calcium alginate, gelatin etc. Based on the valuable data about the above presented activities, clinical studies of the enzyme should be conducted to explore its reuse.

REFERENCES:


