

# Immobilised Enzymes and Its Application in The Food Industry

<sup>1</sup>Priya V. N. and <sup>2</sup>Usha Ravindra

<sup>1</sup>Ph.D. Scholar, Department of Food Science and Nutrition, UAS, GKVK, Bengaluru-560065

<sup>2</sup>Head of the Department, Department of Food Science and Nutrition, UAS, GKVK, Bengaluru-560065

\*Corresponding Author: [priyanagaraja1998@gmail.com](mailto:priyanagaraja1998@gmail.com)

Enzymes are the biological catalysts that promote chemical reactions in living organisms. Enzymes catalyse reactions in different states, individual molecules in solution, in aggregates with other entities and as attached to surfaces. In recent years, the application of enzymes in different industries is continuously increasing. Industrial applications of enzymes include food (baking, dairy products, starch conversion) and beverage processing (beer, wine, fruit and vegetable juices), biosensors, cosmetics, wastewater treatment, health care and nutrition, pharmaceuticals and chemical manufacture and bio fuels such as biodiesel and bio-ethanol. Traditionally, enzymes in free solutions (*i.e.* in soluble or free form) react with substrates to result in products. Such use of enzymes is wasteful, particularly for industrial purposes, since enzymes are not stable, and they cannot be recovered for reuse.

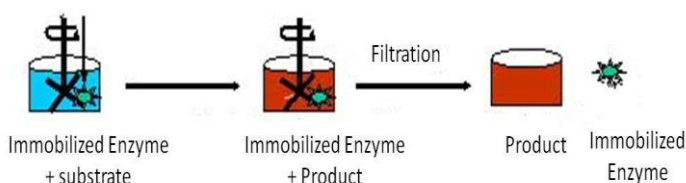
However, the applications and desirable traits of enzyme are often hampered by their sensitivity to process conditions, low stability, their cumbersome from recovery and reuse and tendency to be inhibited by high concentrations of reaction components. These drawbacks which were listed above can be overcome by a technique called by immobilization of enzyme or immobilized enzymes which results in the development of stable, robust and preferably insoluble biocatalysts.

Immobilized enzymes mean fixing of an enzyme within or onto an insoluble matrix. Enzyme immobilization may be defined as the confinement of an enzyme to a phase (matrix/support) other than the substrates and products. Inert polymers and inorganic materials are usually used as carrier matrices. The term immobilized enzymes refer to enzymes physically confined or localized in a certain defined region of space with retention of their catalytic activities, and which can be used repeatedly and continuously. Moreover, the attachment of an enzyme to a solid support can increase its resistance to various environmental changes such as pH or temperature.

## Preparation of immobilized enzymes

Preparation of an immobilized enzymes involves: An enzyme, support matrix and mode of attachment.

Immobilized Enzyme



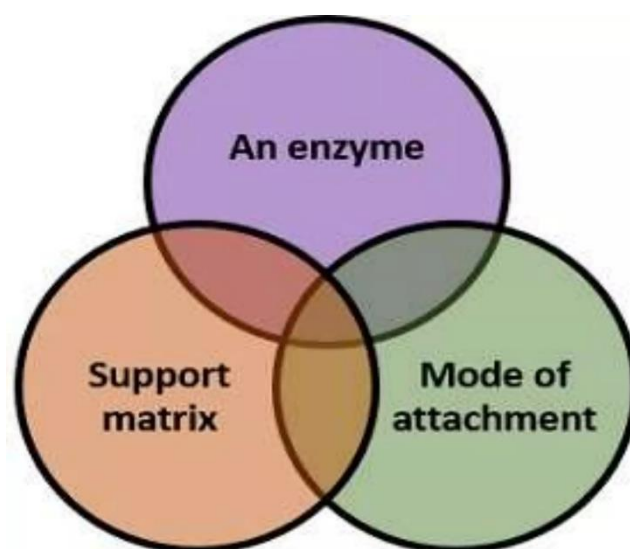
**Fig. 1 Enzyme immobilized**

**A. Support matrix:** Support matrix that holds the enzyme should be: Cheap and easily available and should not react with medium and enzyme. Three types of matrixes are used:

1. Natural polymers: alginate, chitosan and chitin, collagen, carrageenan, gelatin, cellulose, starch, pectin
2. Synthetic polymers: ion exchange resins/polymers [polyvinyl chloride (PVC), UV activated Polyethylene glycol (PEG)]
3. Inorganic materials: ceramics, silica, glass, activated carbon, charcoal.

**B. An enzyme:** Different enzymes such as lipase, oxidoreductase, protease, alpha amylase and lactases are used for enzyme immobilization.

**C. Mode of attachment:** Various methods of mode of attachment of enzymes are used for immobilization such as entrapment, adsorption, covalent binding and



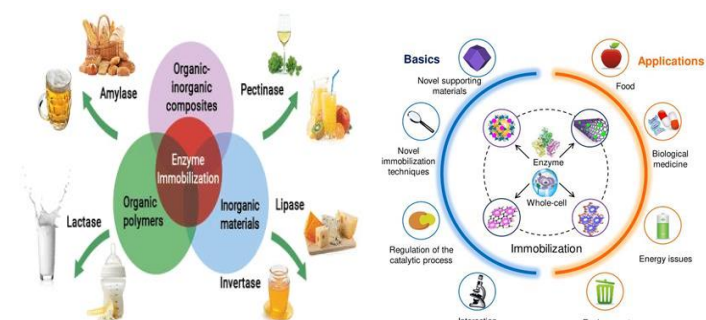
cross binding *etc.*

## Fig. 2 Preparation of immobilized enzymes involves – An enzyme, support matrix and mode of attachment

### Application of immobilized enzymes in food industry

In recent years the use of immobilized enzymes has increased considerably in various industries like pharmaceuticals, detergents, chemicals and mainly in the food industry. Food industry is invariably searching newer and advanced technologies to fulfill the consumer demands. The use of enzymes for transformation of raw materials into useful products has been practiced from early times. Enzymes are generally regarded as safe (GRAS) from the legal point of view that has aroused their extensive use in food processing. Addition of few enzymes in purified state to food preparations enhances flavor, aroma, appearance and nutritional value of foods.

But many main problems were associated with the industrial application of enzymes are their cost and unstable nature that can be overcome by immobilization of enzymes. Applications of immobilized enzymes in food industry are continuously increasing.



**Fig. 3 Different enzymes used for enzyme immobilization and their applications**

The main applications of immobilized enzymes in the food process industry are as follows

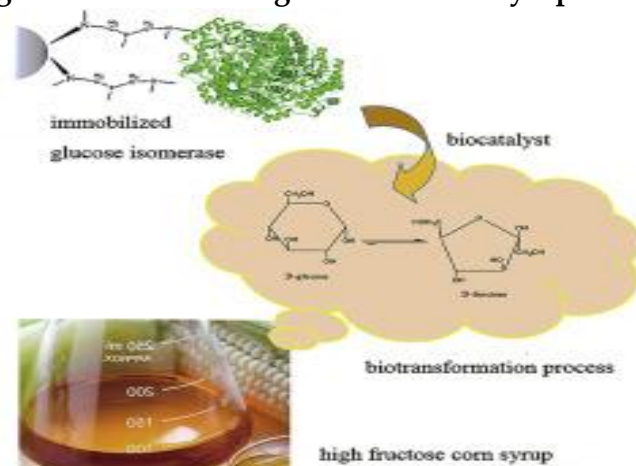
### Sugar industry

Most important application of immobilized enzymes in food industry especially in the sugar industry is the conversion of glucose syrups to high fructose syrups by the enzyme glucose isomerase. High-fructose corn syrup (HFCS) is used as a sweetener in beverages and foods. It has fewer calories and is cheaper than sucrose. Immobilized glucose isomerase is used for better quality and consistency.

Immobilization technique is used in the brewing industry by the entrapment of yeast cells to increase their concentration; this reduces the process time. Immobilized yeast cells reduce processing time without affecting the product quality. Most alcoholic

beverages, such as whiskey, vodka, and brandy, are produced from sugar containing raw materials.

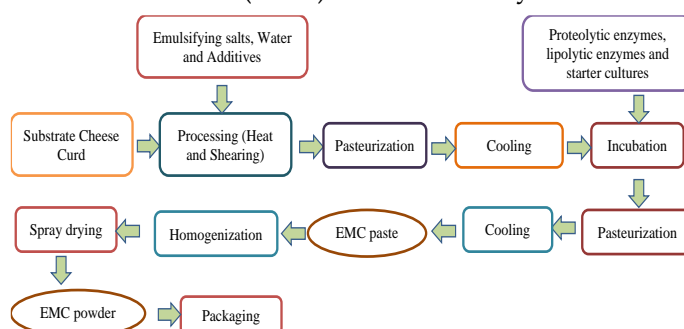
**Fig. 4. Production of high fructose corn syrup using**



**immobilized enzyme glucose isomerase**

### Dairy industry

Lipases are commonly used in the dairy industry to hydrolyze milk fat and current applications of lipases in the dairy industry includes cheese ripening, flavour enhancement, manufacturing cheese-like products and lipolysis of cream and butter fat. Cheese texture is dependent on fat content so lipases that release short-chain fatty acids, develop the sharp and tangy flavor, whereas release of medium-chain fatty acids causes a soapy taste in the product. To reduce above components immobilized enzymes are more helpful. Lipases are also used for enzyme-modified cheeses (EMC) to liberate fatty acids.



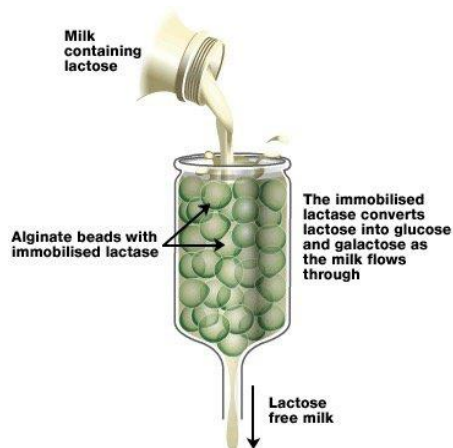
**Fig. 5. Process flow chart for the production of enzyme modified cheese powder**

### Immobilized enzymes-In the production of lactose free milk

Lactose is a disaccharide composed of glucose and galactose units linked by  $\beta$ -1,4 glycosidic bond. Lactose is main sugar found in milk and milk products. Lactase/  $\beta$ -galactosidase enzyme produced by walls of small intestine hydrolyzes milk sugar lactose into glucose and galactose. Lack of lactase enzyme in some people leads to a condition called lactose intolerance characterized by decreased ability

to digest lactose. High lactose content present in milk products like ice-cream, condensed milk crystallizes resulting in inferior quality products.

Milk containing lactose was passed through the vessel containing immobilized lactase and alginate beads. Immobilized lactase converts lactose into glucose and galactose as the milk flows through it. Alginate beads with immobilized lactase passes through the vessel as lactose free milk.

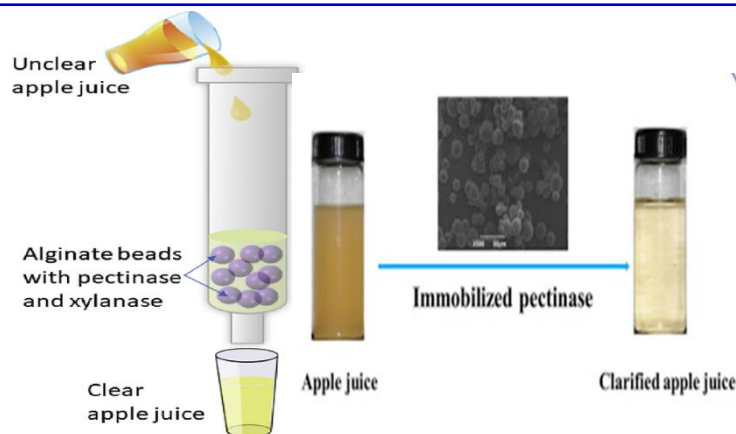


**Fig. 6. Process flow diagram for the production of lactose free milk**

#### Clarification fruit juices

Clarification is an important step in juice processing. On industrial level juice clarification is mainly achieved by making use of pectinolytic enzymes. Immobilized pectinase in ultrafiltration serves as an alternative to conventional processes (use of chemical finings) for clarification of fruit juice. Pectinase and cellulase immobilized on suitable carriers are used for the production of jams, jelly syrup form fruits and vegetables.

Unclear apple juice was passed through the vessel where alginate beads with immobilized enzymes pectinase and xylanase get immobilized with the juice and passes through the vessel as clear apple juice.



**Fig. 7. Process flow diagram for the production of lactose free milk**

#### Future Prospects

- Immobilized enzymes can be of great help in national security, for example, biocatalysts may be incorporated into air filters, masks and clothing to neutralize chemical gases or vapors.
- In food industry immobilized enzymes like glucose oxidase can be employed for removing oxygen from beverages thereby reducing oxidative deterioration by converting it to gluconic acid.

#### Conclusion

Enzyme immobilization is a highly efficient and cost-effective technique used in various fields but mainly in food industry. This technique is continuously replacing conventional approaches in both lab scale and industrial levels. In food industry these immobilized enzymes can be a great help in producing different products like lactose free milk, sweeteners, clarified wines *etc.* More research should be focused on overcoming the limitations and expanding the range of applications of these immobilized enzymes.

\* \* \* \* \*