Frequently Asked Questions

1. **Write a note on the scope of Amylase**

Amylases are important hydrolase enzymes which have been widely used since many decades. Among amylases α-Amylase is in maximum demand due to its wide range of applications in the industrial front. α-Amylase can be produced by plant or microbial sources. Due to the advantages that microbial production offers, α-Amylase from microorganisms has been focused upon and preferred to other sources for production. The ubiquitous nature, ease of production and broad spectrum of applications make α-Amylase an industrially important enzyme.

2. **What are amylases?**

Amylase is a digestive enzyme that aids in the breakdown of carbohydrates by breaking the bonds between sugar molecules in polysaccharides through a hydrolysis reaction. It can be found in animals, plants, and bacteria.

3. **Name the different classes of amylases**

Amylase can be classified into three types: alpha-amylase, beta-amylase, and gamma-amylase. The three types differ in how they hydrolyze the polysaccharide bonds. Neither beta nor gamma amylase is found in animal tissue. This information sheet will focus on alpha-amylase, which is an important enzyme in digestive and metabolic processes.

4. **Who discovered the amylase for the first time?**

In 1815, Kirchhoff preformed an experiment, which converted four parts of water, two parts of starch, and malt into a starch paste. This paste began to liquefy into sweet syrup. His results showed that gluten had the capacity to convert a larger quantity of starch into sugar. Thus, Kirchhoff laid the foundation for the discovery of Amylase.

5. **Who isolated the amylase?**

In 1833, Anselme Payen and Jean-François Persoz further describe and isolate diastase (amylase) in powder form from barley malt, showing it to be heat labile. They postulated the central importance of what would later be named "enzymes" in

biology.

6. **Explian the structure of amylase**

To the right is a ribbon diagram of alpha-amylase composed of beta-sheets and alpha-helices. It consists of 496 amino acids, one calcium ion, one chloride ion and 170 water molecules. The Ca(2+) ion, which is required for its function, is bound to an aspargine, argine, asparate, histidine and three water molecules. In addition, the Cl(-) ion is bound to an argine, asparagine and argine and one water molecule.

7. **Write a note on the function of amylase in the body**

Amylase is critical in the digestion of starch into sugars to make them available energy sources for the body. Amylase is found in two primary places within the human body, and the two types are classified according to where they are found. Salivary Amylase is a component of saliva, and breaks starch into glucose and dextrin. It hydrolyzes the bonds between long-chain polysaccharides found in food, breaking compounds such as glycogen and starch into their useful monomers, glucose and maltose. Pancreatic Amylase is added to the small intestine to further digest starches; amylase is denatured in the acidic stomach. Amylase is also present in blood where it digests dead white blood cells.

8. **What are the uses of amylase?**

Amylases can be used in the production of high-fructose corn syrup, as well as in alcohol production and brewing industries. Agriculturally, amylase has been used to develop a more digestible feed for animals. In addition, Amylase is found in yeast, which is used in bread making. It is important when breaking down the flour (starch) into simple sugars to feed to yeast. The yeast then breaks down the sugars into alcohol and carbon dioxide to cause the bread to rise and give it its tasty flavor.

9. **What are the sources for amylase production?**

α-Amylase can be isolated from plants, animals or microorganisms. The enzyme has been isolated from barley and rice plants. It has been found that cassava mash

waste water is a source of α-Amylase which is active in wide range of pH and temperature. In the recent past, there has been extensive research on microbial production of α-Amylase.

10.Microbes are the important source for the amylase production. Why?

There are 2 major reasons for the increasing interest in microbial sources:

1) The growth of microorganisms is rapid and this will in turn speed up the production of enzyme. Microorganisms are easy to handle when compared to animals and plants. They require lesser space and serve as more cost effective sources.

2) Microorganisms can be easily manipulated using genetic engineering or other means. They can be subjected to strain improvement, mutations and other such changes by which the production of α-Amylase can be optimized. Also, the microorganisms can be tailored to cater to the needs of growing industries and to obtain enzymes with desired characteristics like thermostability.

11.Name he bacterial species involved in amylase production

α-Amylase is produced by several bacteria, fungi and genetically modified species of microbes. The most widely used source among the bacterial species is the Bacillus spp. B. amyloliquefaciens and B. licheniformis are widely used for commercial production of the enzyme.

12.Explain the Purification process of α-Amylase

Enzymes used for industrial applications are usually crude preparations and require less downstream processing. Whereas the enzymes used clinical and pharmaceutical industry need to be highly purified. Also when used for study of structure function relationships and biochemical properties the enzymes have to be in purified form. Purification methods commonly employed are precipitation, chromatography and liquid-liquid extraction depending on the properties of the

enzyme desired. A combination of the above methods is used in a series of steps to achieve high purity. The number of steps involved in purification will depend on the extent of purity that is desired. The crude extracellular enzyme sample can be obtained from the fermented mass by filtration and centrifugation. In the case of intracellular enzymes, raw corn starch may be added followed by filtration and subsequent steps. The crude amylase enzyme can be precipitated and concentrated using ammonium sulphate precipitation or organic solvents.

The precipitated sample can be subjected to dialysis against water or a buffer for further concentration. This can be followed by any of the chromatographic techniques like ion exchange, gel filtration and affinity chromatography for further separation and purification of the enzyme.

13.What are applications of amylase in bakery industry?

α-Amylase is added to the dough in bread baking process. This causes the starch to hydrolyze into small dextrins which can further be fermented by yeast. This increases the rate of fermentation. Also the starch hydrolysis decreases the viscosity of the dough, thus improving its texture and increasing loaf volume by rising of dough.

Once the baking is done, there may be changes during storage of baked products. All undesirable changes like increase of crumb firmness, loss of crispness of the crust, decrease in moisture content of the crumb and loss of bread flavor together are called staling. The enzyme is also used as an anti staling agent to improve the shelf life and softness retention of baked goods.