

## YEASTS

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### Contents

1. Introduction
  2. Properties of Yeasts
    - 2.1. Morphological Characteristics.
    - 2.2. Reproduction.
    - 2.3. Physiological Characteristics.
  3. Classification of Yeasts.
    - 3.1. Ascomycetes (true yeasts)
    - 3.2. Fungi imperfecti
  4. Industrially Use of Yeasts
    - 4.1. Baker's yeast.
    - 4.2. Brewer's Yeast.
    - 4.3. Wine Yeasts
    - 4.4. Distillers Yeast
    - 4.5. Yeasts in Other Fermented Products.
  5. Production of Nutrients and Enzymes with Yeasts
    - 5.1. Yeast Biomass
    - 5.2. Production of Yeast Protein Preparations.
    - 5.3. Fats
    - 5.4. Vitamins
    - 5.5. Enzymes
  6. Yeast Autolysates
- Glossary  
Bibliography  
Biographical Sketch

### Summary

According to botanical classification yeasts belong to the division fungi and the yeasts found in food are divided to classes: *Ascomycetes* and *Fungi imperfecti*. These in turn are divided into Orders, Families, and Genera. Most yeasts used industrially belongs to the class *Ascomycetes* and the most important is the genus *Saccharomyces*. *Saccharomyces cerevisiae* is the leading species used, for example in the baking industry for leavening of bread, in production of beer and wine, and for production of ethanol and several other products used in the food industry. *Saccharomyces cerevisiae*

var. *ellipsoideus* is known as wine yeast because this is a high-alcohol-yielding variety. It is also used in industrial ethanol production with fermentation technology.

Among *Fungi imperfecti*, film yeasts such as genus *Candida* and *Mycoderma* which grow on wine, beer, cheese, pickles, sauerkraut, and other fermented products and take part in their spoilage, are of commercial significance. The manufacture of yeast started in the second half of the nineteenth century. Strains of *Saccharomyces cerevisiae* for use in production of baker's yeast are grown on a molasses-mineral salts medium. During growth of the yeast the medium is aerated at a rapid rate. The yeast is centrifuged out in the form of „cream”, which is put through a filter press or drum filter to remove excess liquid. The mass of yeast is made into cakes of different size after incorporation of small amounts of vegetable oil. Active dried yeast is made under a carefully controlled temperature regime.

Most strains used in breweries are *Saccharomyces cerevisiae*. Yeasts may be carried in pure culture in brewery laboratory or obtained when needed from specialized laboratories. Traditional wine making was based on the natural yeast flora of the grape. The grapes have a variety of micro-organisms on their surfaces, including yeasts and bacteria. To suppress the growth of unwanted microbes, either sulfur dioxide or sulfite was added or the grapes were pasteurized. In modern wine technology carefully selected yeast species are added to the grapes. These strains are varieties of *Saccharomyces ellipsoideus*, a high-alcohol-yielding strain that contributes to the specific flavor of famous wine types. For champagne production, specific yeasts, tolerant of high alcohol content and carbon dioxide pressure, are selected.

Distiller's yeast is ordinarily a high-alcohol-yielding strain of *Saccharomyces cerevisiae* var. *ellipsoideus* adapted to the medium or mash generally used in distillery. Malted grains (barley, wheat, maize, rye), potato and molasses are the common raw materials in industrial ethanol production by fermentation.

The incidental consumption of microbes by humans in fermented foods and that of distiller's and brewer's spent grains by domestic animals is quite old. However a conscious attempt to grow micro-organisms for human consumption started in the twentieth century. Due to their high protein and vitamin content, yeasts may be used for food purposes. Considerable amounts of yeast are now produced for feed and rather less for food purposes. Yeasts produced directly for such purposes are termed *primary* and those recovered as by-products of a fermentation process, e.g. brewing, are called *secondary*.

Protein concentrates and isolates, often called *single-cell protein* may be produced and used for protein enrichment of low-protein foods. Proteins are rich in lysine, but generally poor in methionine. An additional problem is the high nucleic acid content of concentrates. For humans an upper limit of daily intake of 2g nucleic acids has been generally accepted.

Certain yeasts synthesize lipids in appreciable amounts. Yeasts can synthesize many of the vitamins and some provitamins. Concerning the concentration level of vitamins in yeast biomass, it should be mentioned that yeasts can absorb thiamine, niacin, biotin,

and, to lesser extent, pyridoxine, inositol, and other vitamins. The level of these vitamins in the substrate in which the yeasts are grown is, therefore, a factor in determining the vitamin content of yeast cells.

From different yeasts several enzymes and biologically active compounds are produced on a commercial scale. Some examples are alcoholdehydrogenase, hexokinase, L-lactate-dehydrogenase, glucose-6-phosphate-dehydrogenase, glyceraldehyde-3-phosphate-dehydrogenase, inorganic phosphatase, invertase.

Yeast autolysates (self digests) are produced by the action of intracellular enzymes, principally proteases, on polymeric proteins and other polymers in the yeast cell. The process results in the formation of degradation products of proteins (polypeptides, peptides, and amino acids) which have a characteristic meat-like flavor.

Yeast autolysates are widely used in soups, gravies, meat dishes, and generally as condiments.

## 1. Introduction

Yeasts are without doubts the most important groups of microorganisms exploited by man. No other group of micro-organisms has been associated with the progress and well-being of humans than yeasts. The association has been based primarily upon the ability of certain yeasts to rapidly and efficiently convert sugars into alcohol and carbon dioxide, thus effecting an alcoholic fermentation of sugary liquids such as fruit juices and grain extracts. Evidence from archeologists shows that apparently all of the ancient civilizations utilized alcoholic fermentation, very much as we do at the present time. In addition to alcoholic beverages, yeasts were widely used in production of baked goods and some specific fermented foods.

While man used the fermentative capabilities of yeast, the concept of yeasts *per se* may be considered to have had its beginning when Leeuwenhoek observed the yeast cells in a droplet of beer in 1680 using a hand-ground lense. Recognition of the importance of this finding was delayed more than a hundred years. In 1818 Erxleben expressed the view that yeasts consisted of living organisms responsible for fermentation. This view received little attention in the following decades when the vitalistic theory proposed that if yeasts are introduced into a sugar-containing solution, they use the sugar as a food and excrete the non-utilized parts as alcohol and carbon dioxide. Studies by Pasteur finally proved that fermentation is due to the activities of living cells.

Since the end of the nineteenth century, yeasts have been produced industrially and the annual production, including that formed during brewing and distilling practices, is in excess of a million tons. Finally it may also be mentioned that yeasts are undesirable if they cause spoilage of, for example, fruit juices and other fruit products.

Although the term *yeast* is used extensively in scientific literature, it has been difficult to state a precise definition of yeasts based on common morphological, physiological and other characteristic properties. If we omit the numerous exceptions, it may be stated most yeasts are *single-celled, colorless, and bud forming; the cell shape is round or*

*oval, and they are able to grow in the absence of air.* This chapter will concentrate on industrially important yeasts without attempting to give an overview of the full world of yeasts. The complexity of classification of yeasts may be illustrated by the fact that the monograph of Lodder and Kreger van Rij represented the evaluation of 1317 strains of yeasts as being classified into 165 species with 17 varieties. The total number of described species is at present estimated to be over 500.

## **2. Properties of Yeasts**

### **2.1. Morphological Characteristics.**

Yeast cells have been intensively investigated because of their practical importance. Microscopic investigations revealed the morphological characteristics of the cell, and later the fine structure of cells was studied by electron microscopy. Biochemists are now able to relate many metabolic functions to the ultrastructure of the cell. The shape of yeast cells varies from spherical to ovoid, lemon-shaped, pear-shaped, cylindrical or even elongated. Parts of the structure which can be seen are the cell walls, cytoplasm, vacuoles of water or fat and granules. Electron micrographs show the membrane structures, the nucleus and structure of organelles.

### **2.2. Reproduction.**

Most yeasts reproduce asexually by budding. The term budding means a process in which some of the protoplasm bulges out the cell wall, the bulge grows in size and finally walls off as a new yeast cell. Bud formation can occur at different sites on the surface of the cell (multi-lateral), exclusively at the two opposite sites (bipolar), or at one pole only (monopolar). Some yeasts (true yeasts, Ascomycetes) may reproduce sexually by means of ascospores. The formation of ascospores follows conjugation of two cells in some species of yeasts. Other species form ascospores without conjugation, but later ascospores or small daughter cell may conjugate. The usual number of ascospores per ascus is a characteristic of the species. Differences in method of conjugation and ascospore formation are used in the classification of yeasts. Asporogenous yeasts don't produce sexually derived spores such as ascospores ; all genera reproduce by budding (or fission).

*Cultural characteristics.* The characteristics of yeast cultures, like formation of sediment ring, islets or pellicles in stationary liquid media are easily detectable and valuable in species characterization. Yeasts are divided into two groups based on the type of growth in or on liquid media. Some yeast species form a film or scum on the surface of liquid. These are called film-yeasts or oxidative yeasts. The other group, the fermentative yeasts, grow throughout the liquid. The appearance of massed yeast is not, for the most part, useful in the identification of yeasts. The appearance of the growth is important when it causes colored spots on foods. However the differentiation between bacterial colonies and those of yeast normally needs microscopic investigation. Most young yeast colonies are moist and somewhat slimy, but they may appear mealy; most colonies are whitish, but some are cream-colored or pink. Some colonies change little with age, but others become dry and wrinkled.

### 2.3. Physiological Characteristics.

Yeasts differ considerably in their physiology. Here only the physiological characteristics of industrially important yeasts will be discussed. Fortunately this group has enough physiological characteristics in common to permit generalizations, but it should be borne in mind that there will be exceptions to every statement made.

The plentiful supply of available moisture is one of the most important conditions of growth of yeasts. In comparison to the other two groups of micro-organisms important in food microbiology, it may be stated that many yeasts grow better in the presence of greater concentrations of solutes, like sugars and salts, than do most bacteria. This means that yeasts require less moisture than bacteria, but most yeasts need more moisture than moulds. The requirements are often expressed in values of water activity ( $a_w$ ). Lower limits of water activity for ordinary yeasts tested thus far range from 0.88 to 0.94. (The so-called osmophilic yeasts can grow at significantly lower water activity and this may of importance in spoilage caused by such type of yeasts). It should also be kept in mind that the water activity values, mentioned above, may vary with the nutritive properties of the substrate, pH, temperature, availability of oxygen, and presence of inhibitory substances. The optimum range of temperature for growth of most yeasts is around 25 to 30 °C and the optimal range of acidity is pH 4 to 4.5.

Although yeasts are classified by some specialists as plants, they lack chlorophyll and are unable to manufacture by photosynthesis, from inorganic substrates, the organic compounds required for energy supply and growth, so they need organic carbon sources. In general, sugars are the best nutrient for yeasts. All fermentative yeasts are able to ferment glucose to produce ethanol and carbon dioxide. This fermentation process is used in practice for producing beer, wine, industrial alcohol, and carbon dioxide produced by baker's yeasts accomplishes the leavening of bread. Some yeasts, e.g. film yeasts, can metabolize other organic carbon sources such as organic acids.

Yeasts can utilize both simple inorganic (ammonia, some species also nitrate) and organic (urea) compounds as a nitrogen source, but also amino acids, peptides and polypeptides. In addition to sugars (sources of carbon, oxygen and hydrogen, and N-containing compounds, the yeasts need several minor, biologically important compounds commonly known as growth factors.

The requirement of yeasts for an exogeneous source of vitamins varies widely. Some yeasts can synthesize all of their required vitamins, whereas other yeasts have multiple requirements. In some cases the requirement is not absolute; the yeast can grow without supplementation of medium with a given vitamin, but its growth rate is low. Biotin is the most commonly required vitamin to be supplemented in the medium whereas riboflavin and folic acid are apparently synthesized in sufficient quantities by all yeasts. Addition of high levels of vitamins can „enrich” yeasts by taking advantage of their ability to concentrate vitamins, particularly vitamins of the B-group, from the medium into the yeast cell.

Eventual supplementation of medium with minerals depends on the type of medium. Generally phosphorus and sulfur is added.

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