



## CHAPTER V

### PRODUCTION OF YEAST EXTRACT

Technically extracts from yeasts can be produced in various ways. The processes differ both in their basic and in the functional properties of the products obtained (Hill, 1981; Sugimoto, 1976).

#### 5.1 Methods of production

The three variants for yeast extraction are plasmolysis, autolysis and hydrolysis (Hill, 1981; Robbin et al, 1975; Sugimoto, 1976). Hydrolysates are prepared by the controlled cooking of yeast in acid solution. Plasmolysates are prepared by extracting the cellular materials from the yeast cell with high concentrations of salt, sugar or certain acetate esters. Autolysates are prepared by inducing the self-digestion of the cytoplasmic materials in the whole cell followed by recovery of the solubilized material (Robbin et al, 1975).

##### 5.1.1 Autolysis

Autolysis, or self digestion, is a complex process. The reactions develop as the cream is heated to a temperature of  $40^{\circ} - 60^{\circ} \text{C}$  (Peppler, 1982). Autolysis occurs when endogenous enzymes, mainly protease and ribonuclease, digest the intracellular high molecular weight components of the yeast cells (Chao et al, 1980). The process can be induced by heating yeast to a temperature (about  $50^{\circ} \text{C}$ ) (Peppler, 1982) where the cell is killed, but the enzymes systems are active. During autolysis, macromolecules are hydrolyzed and the soluble degradation products of small molecular size, such as peptide, amino acids,

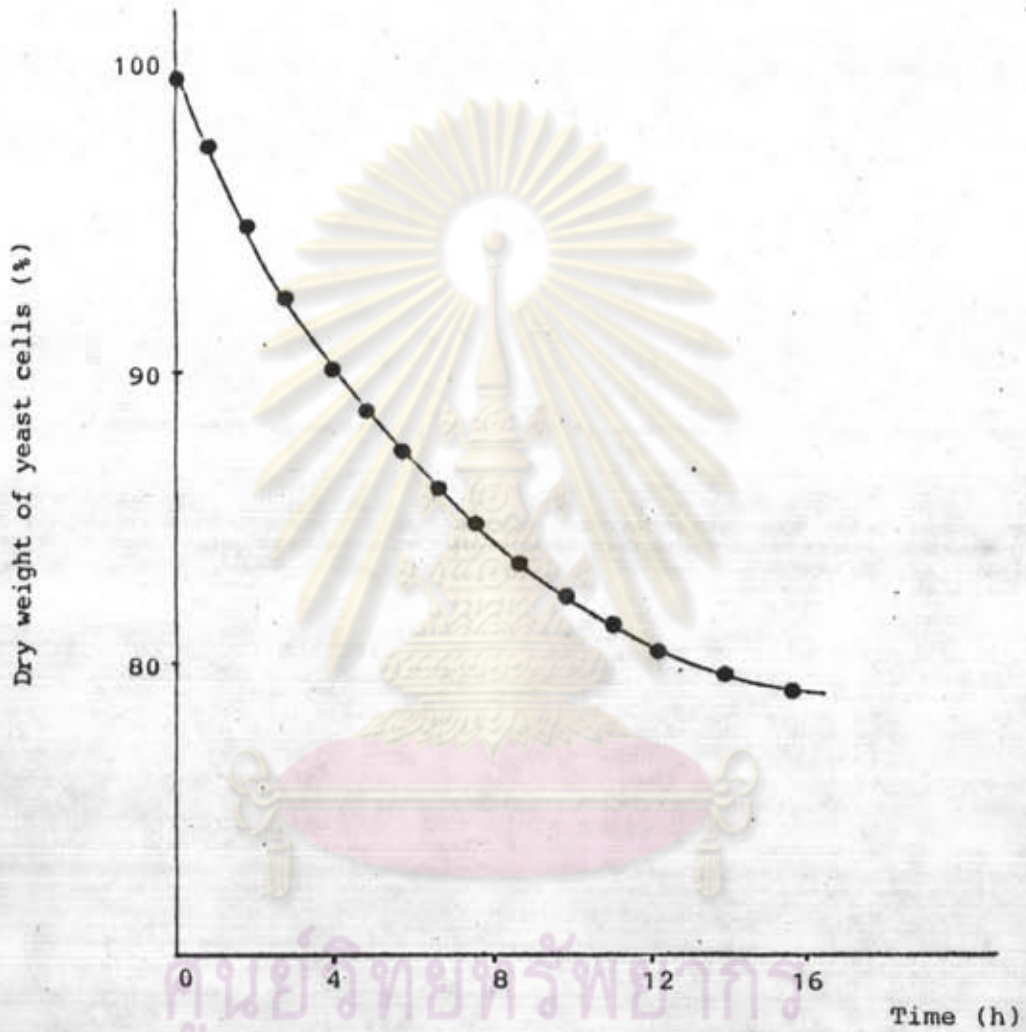
nucleotides etc., diffuse out from the cell (Peppler, 1982). Figure 5-1 illustrates the changes in yeast cell weight as leakage of cell constituents and their derivatives occur during autolysis (Hough, 1970). The course of changes in yeast protein and amino acid content during autolysis are shown in Figure 5-2 (Hough, 1970). The rate of yeast autolysis depends on temperature, pH value, time, type of yeast and conditions of culture (Peppler, 1982) (Table 5-1).

#### 5.1.1.1 Mechanism of yeast autolysis

Cell death arises from either starvation or from other adverse circumstance (Hough, 1970) such as high temperature, the yeast will almost certainly be present as a mixture of living, dying and dead cells. The cells will be starved of nutrients and some of them will become moribund, leading to a change in the permeability of the plasma membrane. Intracellular material will be released into the medium. The living cells will respond to the presence of protein in the medium (some of which is derived from the dead cells) and will secrete proteolytic enzymes to utilize it. These proteolytic enzymes are probably derived from inside the cell vesicles or lysosomes and released by reverse pinocytosis. Hence the first stage of autolysis is an attempt by the yeast cell to secure nutrient material. Some of the proteolytic activity in the medium, however, will be derived from dead yeast cells in which general disorganisation has caused the release of enzymes within the vesicles. Hydrolytic enzymes other than proteases liberated at the same time will degrade macromolecules such as nucleic acids. This process proceeds within the yeast mass until eventually all the cells die and complete autolysis will occur (Hough 1970, 1971).

#### 5.1.1.2 Proteases in yeast autolysis

It is known that there are four proteases release



**Figure 5-1** Solubilization of yeast solids during autolysis in water at pH 6.5 and 45 C (Hough, 1970)

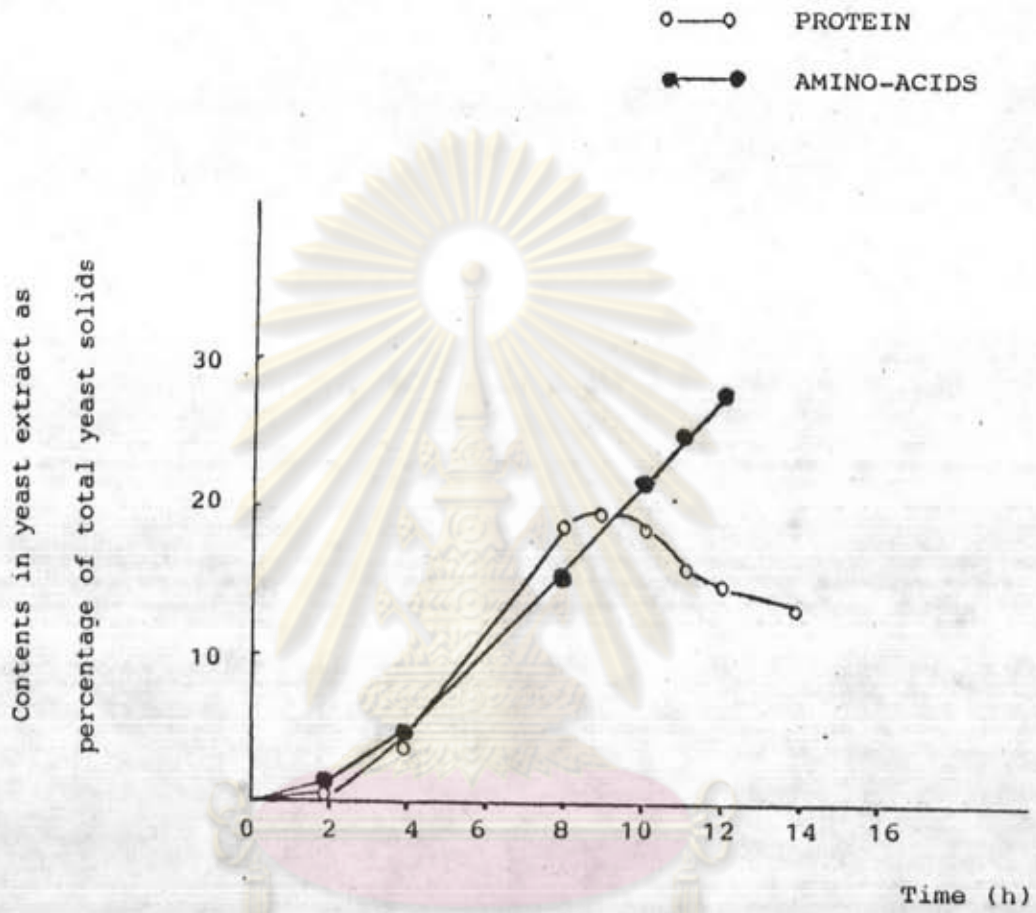


Figure 5-2 Changes in protein and amino acid concentrations (as percentage of total solids) in yeast extract during autolysis (Hough, 1970)

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**Table 5-1** Comparison of proteolytic enzymes from three different autolysing yeasts (Hough, 1970)

	Enzymes	Baker's yeast <u>S. cerevisiae</u>	Top fermenting brewer's yeast <u>S. cerevisiae</u>	Bottom-fermenting brewer's yeast <u>S. carlsbergensis</u>
pH optimum	A	6.8	7.3	7.5
	B	4.9	4.5	6.2
	C	4.5	6.6	6.2
	D	4.3	6.3	3.5
Temperature optimum (°C)	A	37	35	35
	B	51	50	50
	C	59	50	50
	D	66	60	60

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during autolysis (Table 5-2), they are referred to as A, B, C and D, of which A is by far the most abundant. The characteristics of these enzymes in Table 5-2 show that A is less stable at temperatures over 30° C than the others and has a pH optimum which is more alkaline than that of the others. The products of the activity of A are amino acids or peptides of low molecular weight, where those of B, C and D are mainly of molecular weight greater than 5,000. Possibly, enzyme A acts on the products of the other three enzymes. The proteases are glycoproteins, containing both glucose and mannose residue in various proportions (Hough 1970, 1971).

Autolysis may arise from excessive permeability of membranes so that compartmentation of the cells break down and lysosomes release their enzymes. Alternatively, autolysis may be due to cessation of anabolism without loss of catabolic activity (Hough, 1970).

#### 5.1.1.3 Exogenous enzyme assisted autolysis

Yeast autolysis is enhanced by the addition of certain exogenous-enzymes to the yeast slurry. (Chao et al, 1980). The use of exogenous enzymes to enhance the autolysis of yeast such as protease enzyme (Papain) (Peppler, 1982), is an initiator for liberating the cell contents. The efficiency of yeast autolysis has been found to be greatly improved by adding at least one protease enzyme and/or mixture of enzymes containing protease, nuclease, lipase, and amylase as the digestive aid. Papain, an FDA acceptable food-use protease is most effective in their synergistic effect on the autolysis (Farnum, 1975).

There are two possible mechanisms that added exogenous enzymes work in a synergistic fashion with the yeast endogenous enzymes to achieve the autolytic degradation of cellular material. One mechanism would be the direct attack on the yeast cell wall and/or membrane by the added enzyme(s) to disrupt the structure of the barrier.

**Table 5-2** Characteristics of the four proteolytic enzymes release during yeast autolysis (Hough, 1970)

Enzyme	Optimum temperature for activity ( $^{\circ}\text{C}$ )	Maximum temperature for 100% stability ( $^{\circ}\text{C}$ )	Optimum pH for activity	Optimum pH for stability
A	35	0	7.5	6.0-6.2
B	50	55	6.2	6.0-6.2
C	50	55	6.2	6.0-6.2
D	60	65	3.5	6.0-6.2



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The cytoplasmic content of the cell, including endogenous enzymes, would be leached out to permit better interaction between the substrates and the enzymes. A second possible mechanism could be the situation wherein the autolysis was initiated from the action by the exogenous enzymes. Enzymes of small size (molecular weight of approximately 13,000 such as ribonuclease and 23,900 such as papain) are known to be able to penetrate through the yeast cell wall and membrane and act hydrolytically on the intracellular components (Chao et al, 1980).

#### 5.1.2 Plasmolysis

Also, the popular methods of autolysis in industry are the use of plasmolysing agent. The common commercial process, non polar organic solvent such as toluol, chloroform, ethyl acetate or amyl acetate or inorganic salts such as sodium chloride are often used to accelerate autolysis for yeast (Knorr et al, 1979).

#### 5.2 Typical production of yeast extract

During autolysis, the solid pressed yeast is changed into a liquid of the consistency of thin cream. The solubilized protein is separated from insoluble residual cell material by centrifugation. The clear supernatant from the centrifuges is next concentrated in vacuum evaporator prior to spray-drying (Pepler, 1982).

#### 5.3 Composition of yeast extract

Chemical analyses pertaining to five extracts produced by five different manufacturers are compared in Table 5-3 to 5-5. The gross composition of selected commercial autolysed yeast powder (Table 5-3) varies markedly in protein and ash content. This indicates the degree of variability in initial yeast composition and differences in process



conditions. The nucleic acid components found in two commercial autolysates (Table 5-4) include GMP (5'-guanylic acid) , a highly desired flavor booster derived from yeast RNA. The nutritional importance of yeast extract centres about contents of the B-vitamins (Table 5-5).



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Table 5-3 Proximate analysis of five commercial autolysed yeast extracts (Peppler, 1982)

Component (% w/w)	A	B	C	D	E
Moisture	3.4	3.1	3.4	3.3	5.8
Carbohydrates	27.3	19.3	20.5	12.0	6.2
Protein (total nitrogen times 6.25)	55.5	50.8	65.1	57.0	67.2
Ash	9.3	22.7	9.7	25.8	10.9
Lipids	0.2	0.3	0.1	0.1	0.5
Organic acids	1.6	3.9	2.6	2.0	4.8
Ammonium chloride	0.7	0.9	1.0	0.7	2.9

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**Table 5-4** Nucleic acid components found in two commercial autolysed yeast extracts (Pepler, 1982)

Component	Content (percentage of product)	
	A (96.6% Dry weight)	E (96.7% Dry weight)
Adenosine monophosphate	ND	0.93
Adenosine	0.48	0.18
Adenine	0.50	ND
Cytidine monophosphate	0.40	0.65
Cytidine	0.57	0.18
Cytosine	0.03	ND
Guanosine monophosphate	0.28	1.32
Guanosine	0.85	0.15
Guanine	0.15	ND
Uridine monophosphate	0.39	1.14
Uridine	0.33	0.24
Uracil	0.65	0.30
Xanthine	ND	0.19
Total	4.63	5.28

ND indicates that the component was not detected.

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Table 5-5 Range of vitamin content of commercial autolysed yeast extracts (Peppler, 1982)

Vitamin	Range of content (mg/100 g)
Thiamin	1.0 - 1.5
Riboflavin	1.5 - 7.5
Nicotinamide	12.5 - 55.0
Calcium pantothenate	3.0 - 12.0
Pyridoxin	1.0 - 25.0
Biotin	0,005 - 0.2



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The principle steps in a typical production are detailed in Figure 5-3.

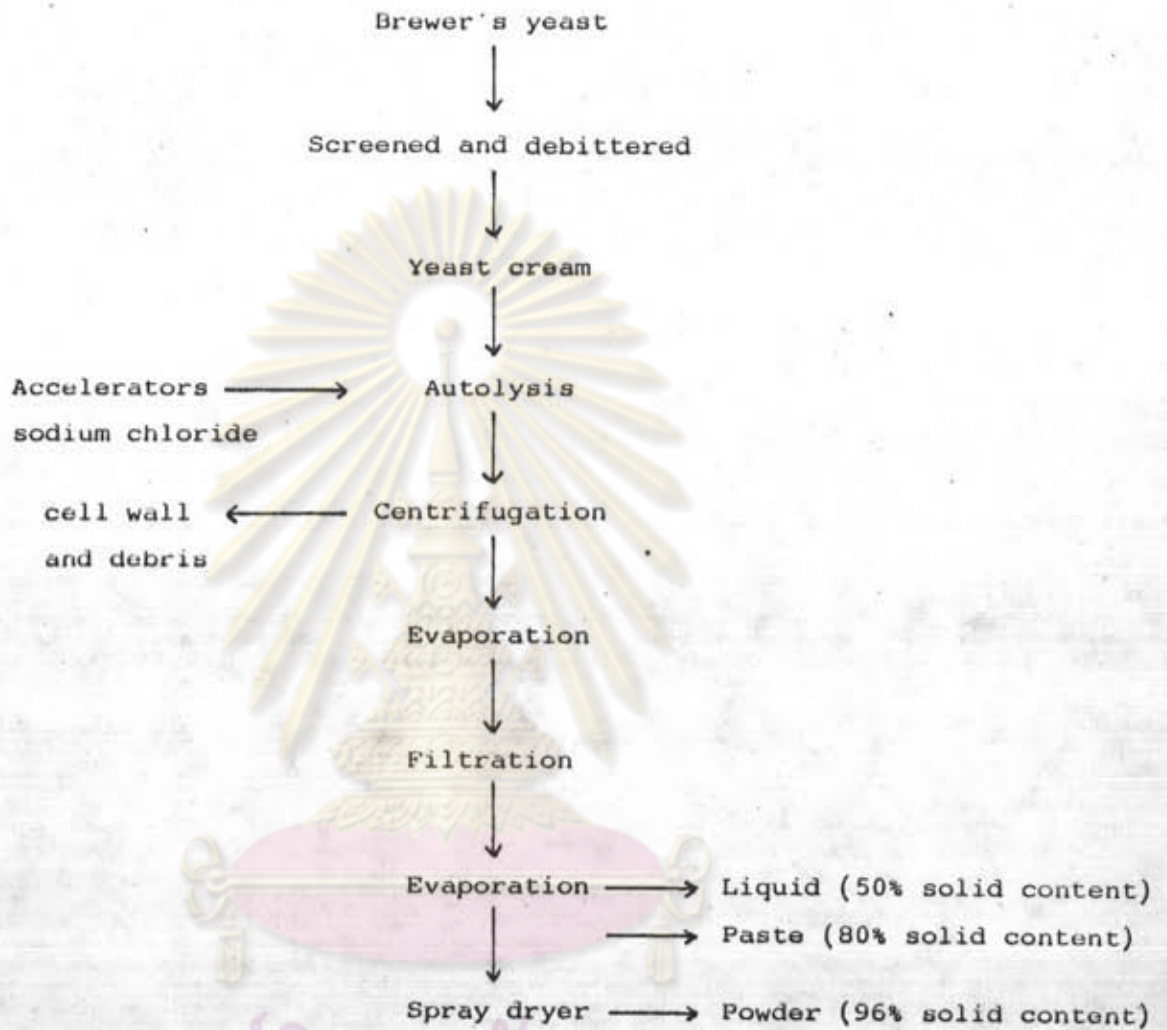


Figure 5-3 Process routes in the manufacture of yeast extracts

(Peppler, 1982)

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