



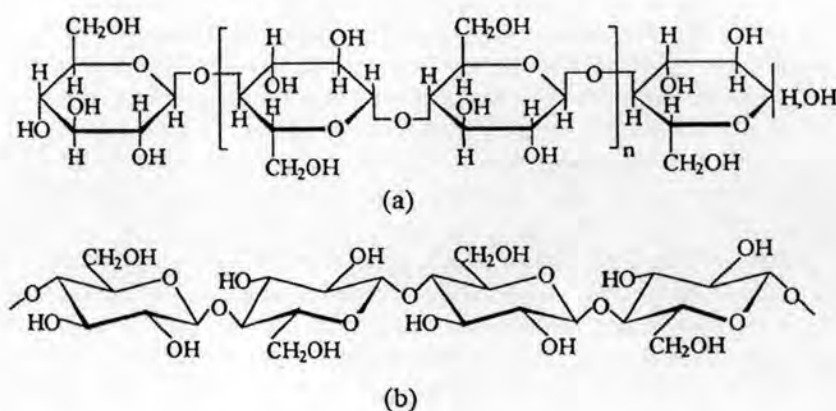
## CHAPTER II

### LITERATURE REVIEW

Cellulose has been the primary component of man's diet since the beginning of human history. It is the most abundant natural polymeric raw material. All forms of plant life contain cellulose. Based on its nearly ubiquitous distribution in nature, and humankind's long exposure to cellulose, cellulose and its derivatives are generally recognized as the safest and most acceptable polymer class for use in food and pharmaceutical products. As supplied in nature, it is free of chemical contaminants (e.g., residual monomers, initiators, or catalysts) commonly present in the synthetic polymer sources. The cotton fiber is the purest source of cellulose. However, a large portion of cellulose for industrial use is obtained from wood sources. The composition of cellulose in wood varies from one wood source to another. In general, wood contains cellulose between 40-55%, on a dry weight basis. The other major components of wood are lignin (15-35%) and hemicelluloses (25-40%) (Kumar and Banker, 1993).

#### Chemical structure and chain length

Cellulose is a polydisperse, linear syndiotactic polymer. Its basic monomeric unit is D-glucose, which links successively through a glycosidic linkage in the  $\beta$ -configuration between carbon 1 and carbon 4 of adjacent units to form long chain 1,4-glucans. Figure 1 shows a structural diagram of a portion of a cellulose chain.



**Figure 1** Cellulose structure: (a) Haworth formula; (b) chair formula. (Hon, 1996)

Because of the  $\beta$ -configuration of the intermonomer links, the glucose units effectively alternate up and down in the chain. Hence, scientists consider cellobiose as the repeating unit of cellulose, on which a syndiotactic configuration of the macromolecule is formed. The size of the cellulose molecule occurring in nature, indicated by its degree of polymerization (DP) or chain length, is dependent heavily on its source (Table 2). In some cases, the DP may exceed 25,000. Pure cellulose

**Table 2** Degree of polymerization of cellulose from various sources. (Hon, 1996)

Source	Degree of polymerization
Acetobactor xylinum	2,000-3,700
Acetobactercellulose	600
Bacterial cellulose	2,700
Bagasse	700-900
Bast fibers	1,000-5,000
Cotton fibers	8,000-14,000
Cotton linters	1,000-6,500
Flax fibers	7,000-8,000
Ramie fibers	9,000-11,000
Wood fiber	8,000-9,000
Valonia	25,000-27,000
Pulp cellulose (bleached)	500-2,100

inevitably loses some of its DP or molecular weight (MW) during isolation and purification. Existing experience indicates that valuable and interesting properties of cellulose and its derivatives can be obtained only if their DP or MW is high. The DP or MW distribution in many cases profoundly influences the mechanical, solution, biological, and physiological properties of cellulose and has given very useful clues to the design of effective cellulose derivatives.

## Hemicellulose

Hemicellulose are not forms of cellulose. They contain several sugar units and exhibit a considerable degree of branching. The most abundant sugar units are 1, 4-linked  $\beta$ -D-xylopyranose (some bearing short side chains of 1, 3-linked  $\alpha$ -L-arabinofuranose or esterified 1,2-linked  $\alpha$ -D-glucuronic acid) and 1, 4- $\beta$ -D-glucomannans with or without 1, 6-linked  $\alpha$ -D-galactopyranose units as side chains. The glucose and mannose units are also present but their distributions are random (Kumar and Banker, 1993).

## Lignin

Lignin is a phenolic polymer that imparts strength and decay resistance to the plant secondary cell wall and is thought to have been essential to the evolution of terrestrial plants. Precursors for lignin biosynthesis are synthesized from L-phenylalanine via the phenylpropanoid pathway that provides ferulic acid (4-hydroxy-3-methoxycinnamic acid) and sinapic acid (3,5-dimethoxy-4-hydroxycinnamic acid) for the synthesis of guaiacyl- and syringyl-substituted lignin monomers, respectively. In angiosperms, lignin biosynthesis requires the activity of at least two cytochrome P450-dependent monooxygenases, cinnamate-4-hydroxylase (C4H), and ferulate-5-hydroxylase (F5H), although the activity of F5H is required only for the synthesis of syringyl monomers (Meyer et al., 1998).

Lignin is responsible for adding rigidity and strength to cell walls and for providing barriers to diffusion and infection. Lignification of the cell wall results in a great decrease in the permeability of the wall to water and to dissolved solutes. The lignin replaces water throughout the bulk of the wall matrix, leaving few narrow pores through which water can penetrate. Once lignification of the cell wall has occurred, the wall becomes impenetrable to all but a very few fungi. The deposition of lignin in the cell wall is one of the most important methods by which the cell wall can defend itself from infection by pathogens. It is part of the hypersensitive response in which cells surrounding the point of parasitic infection rapidly undergo lignification and cell death. The point of infection is then effectively sealed off from the rest of the plant by a layer of lignified cells, then the parasite can not penetrate any further. This response is triggered by both fungi and bacteria. Thus, lignin provides a physical barrier to attack by pathogens (Brett and Waldron, 1990).

### Sources of cellulose

Cellulose in its native form is distributed throughout the plant kingdom, in trees, shrubs, and grass. It incorporates about 40% of the carbon in plants and its total amount in the vegetable world is estimated at over  $10^{11}$  t. The cell walls of green algae and some flagellates and the membranes of most fungi also contain cellulose. Cellulose never occurs in a pure form: in softwood and hardwood, it constitutes about 40-50% of the weight, in flax 70-85%, whereas cotton seed hairs, which are the purest source, contain more than 90% (Table 3). In these materials, the cellulose macromolecule serves as a structural material within the complex architecture of the plant cell walls.

**Table 3** Natural sources of cellulose. (Hon, 1996)

Source	Cellulose content (%)
Bagasse	35-45
Bamboo	40-55
Cotton	90-99
Flax	70-75
Hemp	75-80
Jute	60-65
Kapok	70-75
Ramie	70-75
Straw	40-50
Wood	40-50

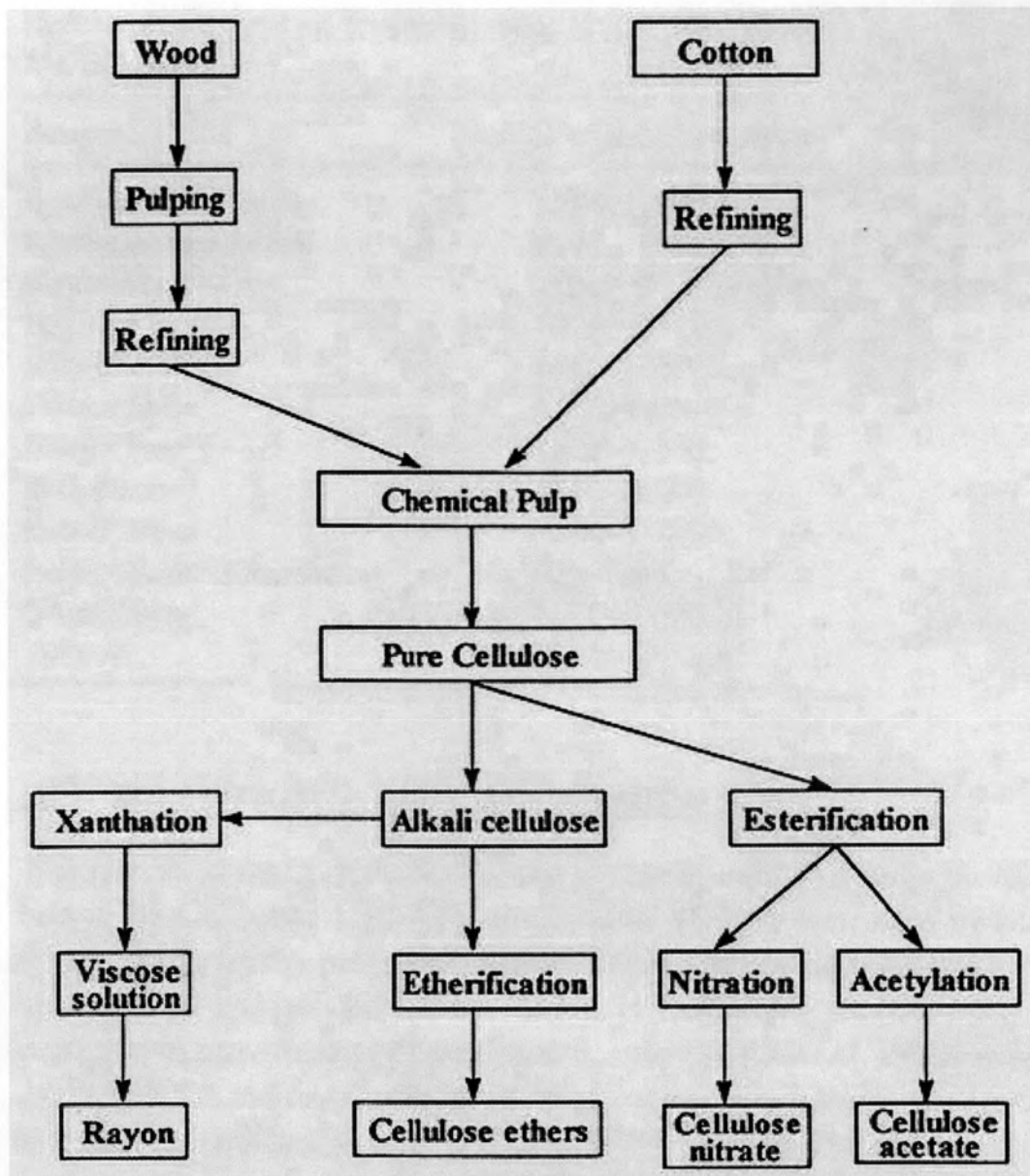
Commercial cellulose production concentrates on the highly pure sources such as cotton or the easily harvested sources such as wood. The isolation of cellulose from wood is more difficult than its isolation from most of its other sources. However, chemical pulping processes to isolate cellulose from wood are among the important chemical industries throughout the world. About 224 million tons of chemical cellulose was produced from wood in the world in 1990. Over one million tons of the chemical



cellulose is used annually by chemical industries in the manufacture of textile fibers and various cellulose derivatives. A general scheme for production of chemical cellulose from wood and cotton is depicted in Figure 2 (Hon, 1996). Cellulose can also be produced from various other plant fibers, such as corn cobs or stalks, soybean hulls, bagasse (sugar cane stalks), oat hulls, rice hulls, wheat straw, sugar beet pulp, bamboo, and fibers such as jute, flax, and ramie among others.

Different sources of cellulose are used for different purposes, for economic reasons. Pulp and paper are usually produced from wood, while textile fibers are generally not isolated from woody fibers. Cotton fibers are a biological source of almost pure cellulose, but this is not usually used in food grade cellulose and are used instead for various cellulose derivatives, pharmaceutical, or chemical engineering uses, such as chromatography, paints, and explosives. Bacterial sources of cellulose have also been developed using *Acetobacter xylinum* that ferment substrates of glucose from corn. These produce cellulose of small particle diameter with more surface area than powdered cellulose derived from wood pulp. They have high tensile strength and water holding capacity and currently are used as a high-value specialty chemical with applications ranging from acoustical speakers, high quality paper, diet foods, and artificial skin.

Most commercial sources of cellulose are from cotton linters and wood. Cellulose obtained from cotton linters needs only a treatment with a hot sodium hydroxide solution that removes the protein, pectic substances, and wax to produce high quality cellulose. Wood requires more extensive processing to solubilize the hemicelluloses and lignins (delignification). In addition, there are relatively small quantities of bacterial-derived cellulose commercially available, but reportedly have not been used for food applications (OMRI, 2001).



**Figure 2** Production scheme for production of pure cellulose and cellulose derivatives from wood and cotton. (Hon, 1996)

## **Pulping process**

Wood contains approximately 50% cellulose, 30% hemicellulose, and 20% lignin. The pulping process separates the cellulose from the lignin and hemicellulose (structurally unrelated polysaccharides), leaving it in a fibrous form that is purified, dried, and shipped in large rolls.

In the wood pulp processing, timber is debarked and cut into chips. These are mechanically ground and then digested (cooked) chemically using either a sulfite or alkali process at elevated temperatures in pressure vessels or digesters. Either process can be modified to produce higher purity cellulose that is not only free of lignins and hemicellulose, but also further degraded to result in reduced molecular weight products and derivatives. Various sulfite processes are used for delignification of the pulp, including the bisulfite process -which uses calcium bisulfite in the presence of sulfur dioxide at a pH range of 2-6 on various wood species (OMRI, 2001).

### **1. Chemical Pulping Process**

This is the cooking of fibrous material into pulp using chemical reagents. The major chemical pulping processes are:

1.1 Alkaline sulfite process involves the use of either sodium hydroxide (NaOH) with sodium sulfite ( $\text{Na}_2\text{SO}_3$ ) or sodium sulfite ( $\text{Na}_2\text{SO}_3$ ) with sodium carbonate ( $\text{Na}_2\text{CO}_3$ ) in solution under pressure and higher temperature in rotary or vertical digester. In this process, pulp yield is from 65-75% and pulp produced is light colored which can easily be bleached.

1.2 Kraft or sulphate pulping process involves the use of caustic soda (NaOH) and sodium sulfite ( $\text{Na}_2\text{S}$ ) to form the white liquor. The fibers are cooked at a temperature of  $170^\circ\text{C}$  and a pressure of 110 psi. Cooking lasts from 1-1½ to 3 hours or more depending on the concentration of the cooking liquor. Pulp yield in this process is about 55%.

1.3 Sulfite or acid process involves the treatment of fibers with solution of sulphurous acid ( $\text{H}_2\text{SO}_2$ ) and water and any of the bisulfites namely, calcium bisulfite [ $\text{Ca}(\text{HSO}_3)_2$ ], magnesium bisulfite [ $\text{Mg}(\text{HSO}_3)_2$ ], and sodium bisulfite ( $\text{NaHSO}_3$ ). The fibers are heated for about 6-12 hours under a temperature range of  $170$ - $176^\circ\text{C}$  and a pressure range of 90-100 psi. With a pulp yield of 60% or more, this pulp is also used for dissolving pulps in the manufacture of rayon and cellulose film or various kinds.

1.4 Soda process involves the use of sodium hydroxide (NaOH). The pulp produced is always bleached and is commonly used for the production of printing paper.

## **2. Semi-Chemical Pulping Process**

The fibrous materials are subjected to a relatively mild chemical treatment, followed by mechanical fiberization. The types of semi-chemical pulping are:

2.1 Cold soda process involves the use of caustic soda (NaOH) solution at room temperature using either atmospheric or hydrostatic pressure followed by defiberization of the fibers in a disk attrition mill. The pulp yield ranges from 85% to 95%.

2.2 Hot soda process is practically the same as the cold soda process except for the application of heat at a certain temperature and pressure during the pulping process.

2.3 Neutral sulfite semi-chemical (NSSC) process is a modified process wherein the fiber is partially digested with a neutral sulfite liquor (consisting of  $\text{Na}_2\text{SO}_3$  and  $\text{Na}_2\text{CO}_3$ ) in a rotary, vertical or continuous digester and later difiberized in attrition mills. Pulp yield ranges from 70-80%.

## **3. Mechanical Pulping Process**

This is the simplest and cheapest method of pulping. It consists of pressing the material (by hydraulic or other means) against a revolving grindstone. The stream of water softens the material and cools the stone at the same time. The types of mechanical pulping are:

3.1 Stone grinding is where the wood or logs are ground against a rotating stone/stone wheel to produce clumps of fibers, or pulps with many broken fibers, fines and shives.

3.2 Pressurized stone grinding is similar to stone grinding, except that this is under raised atmospheric pressure, allowing steam heating, and higher temperatures than stone grinding to soften the lignin. This process enables the fibers to be pulled apart more easily with less damage to fiber length.



3.3 Refiner mechanical pulping is where the wood or chips are defibered in a disk refiner producing better fibers than in the stone grinding process. Refining is usually conducted in 3 stages:

Screw feeds - 1st stage (20-30% consistency)

Screw feeds - 2nd stage (15-25% consistency)

Screw chest - 3rd stage (6% consistency)

3.4 Thermomechanical pulping (TMP) is where the chips are washed at 70°C, then heated in steaming tube. The chips pass down a screwfeed being pressurized against a pneumatic piston and they then expand into an impregnating vessel absorbing hot water uniformly, allowing easier refining. First stage refining in disc refiners takes place under pressure at raised atmospheric pressure. Pulp then passes a cyclone where steam is removed. Pulp is diluted then goes to 2nd and 3rd stage refiners. This pulping process separates the fibers with very little shortening due to the heat softening of lignin. The high lignin content on the outside makes the resultant fibers stiffer and less easily beaten and bonded in the paper.

3.5 Chemithermomechanical pulping is similar to TMP but with cooking chemicals. E.g. Sodium sulphite ( $\text{Na}_2\text{SO}_3$ ) added to the impregnating vessel to soften the lignin (Pulping process, 2005).

After pulping, several steps are required, including bleaching and alkali extraction, to develop products with the desired molecular weight and physical length of the fibers. Bleaching may include up to twelve steps of chlorination, hypochlorite bleaching, chlorine dioxide bleaching, and extraction with concentrated sodium hydroxide along with intermediate alkaline extraction (washing) after each oxidative stage. It must be purified and bleached to meet the specifications of Food Chemicals Codex. Purification may involve addition of surfactants in another hot weakly alkaline extraction step after chlorination. Another option is to treat pulp with another bath of 6-10% sodium hydroxide after the bleaching is finished. Final stages in most purification plants include the use of sulfuric acid and optional chelating agents (OMRI, 2001).

### **Powdered cellulose**

Manufacturers of powdered forms receive the wood pulp in the dried form from pulp mills and further mechanically process it to attain the desired fiber length, varying from 0.5 to 4 mm.

Cellulose quality is measured by the content of alpha-cellulose, which is that portion insoluble in 18% alkali. Highly purified forms (over 99% alpha cellulose) are used to make derivatives such as the cellulose gums, including sodium carboxymethylcellulose, methylcellulose and hydroxypropylmethylcellulose. However, powdered food grade cellulose is not required to reach that level of purity, because all cellulosic cell-wall materials are components of all fruits, vegetables, and cereals.

### **Microcrystalline cellulose (MCC)**

Microcrystalline cellulose is a purified, partially depolymerized cellulose that occurs as a white-colored, odorless, tasteless, crystalline powder composed of porous particles. It is commercially available in different particle size grades, which have different properties and applications (Wheatley, 1994).

Microcrystalline cellulose is prepared from native cellulose by controlled acid hydrolysis, which preferentially attacks amorphous regions, followed by back-neutralisation with alkali prior to recovery by spray-drying. The material produced is composed largely of aggregated bundles of crystallites of cellulose formed as porous particles, the various grades of which can be influenced by the source of the wood pulp used, the severity of the processing employed, co-processing with other additives such as sodium carboxymethylcellulose, and both the particle size range and residual moisture content as affected by the spray-drying conditions employed. Avicel<sup>®</sup> PH-101 (FMC) is among the most widely used pharmaceutical grades, has an average particle size of 50 mm approx, and is employed mainly as a filler:disintegrant:binder in tablets and capsules, and as an aid in extrusion:spheronization. Attempts to size reduce this and other grades by mechanical and other means are resisted by strong hydrogen bonding associated with surface groupings, which also promote re-aggregation during drying procedures (Levis and Deasy, 2001).

**Table 4** Pharmacopeial specifications of microcrystalline cellulose (Wheatley, 1994).

Test	JP	PhEur	USP
Identification	+	+	+
Characters	+	+	-
pH	5.0-7.0	5.0-7.5	5.0-7.0
Bulk density	+	-	+
Solubility	-	+	-
Loss on drying	≤ 7.0%	≤ 6.0%	≤ 7.0%
Residue on ignition	≤ 0.05%	-	≤ 0.05%
Conductivity	+	-	+
Sulfated ash	-	≤ 0.1%	-
Ether-soluble substances	≤ 0.05%	≤ 0.05%	≤ 0.05%
Water-soluble substances	≤ 0.24%	≤ 0.25%	≤ 0.24%
Heavy metals	≤ 10 ppm	≤ 10 ppm	≤ 0.001%
Starch	-	+	-
Organic volatile impurities	-	-	+
Microbial limit	+	+	+
Assay	-	-	97.0-102.0%

The utility of pulverized or powdered cellulose is limited due to the highly fibrous form that results in poor “mouthfeel”. Cellulose in its more native state has both amorphous and crystalline regions, and the amorphous regions are attacked first by solvents and chemical reagents. MCC production uses an additional step involving hydrolysis of the purified wood pulp, using hydrochloric acid to reduce the degree of polymerization. This leaves only the tiny, acid-resistant crystalline regions. It can be

spray-dried, and is then termed “powdered MCC”. This produces average particle sizes ranging from about 20-90  $\mu\text{m}$ . Another form is colloidal MCC, which is water dispersible and has properties similar to water soluble gums. It requires the use of mechanical energy after hydrolysis to tear apart microfibrils and provides a major proportion of colloidal sized aggregates (less than 0.2  $\mu\text{m}$  in diameter) (OMRI, 2001).

### **Scale-up**

Scale-up of a process or batch is studied to determine the operating conditions applicable to large-scale production batches, with the goal of obtaining products of the same quality based on previously optimized laboratory-scale. Scale-up of processes that involve powder handling is especially difficult because the dynamic behavior of powder is not very well understood. Therefore, although scale-up processes of materials in the solid state can be based on dimensional analysis, mathematical modeling, and computer simulation, most of the work in this field still depends on trial and error and the principles of geometric similarity. The latter describes the interrelationships among system properties on scale-up; thus, the ratio of some variables on small-scale equipment should be equal to that of similar variables in equivalent larger-scale equipment (Chuwumezie et al., 2002).

Ju and Chase (1992) classified the scale-up of a bioprocess into three stages: (1) *laboratory scale*, where elementary studies are carried out; (2) *pilot scale*, where the process optimizations are determined; and (3) *plant scale or production scale*, where the process is brought to economic fruition. Scale-up is a procedure for the design and construction of a large scale system on the basis of the results of experiments with small-scale equipment.

Kossen and Oosterhuis (1985) divided the approach for scale-up into four widely recognized steps: 1) fundamental methods; 2) semifundamental methods; 3) dimensional analysis; and 4) rules of thumb.

#### **1. Fundamental methods**

These methods are based on the application of turbulence models for description of the influence of operating conditions and geometrical design of the bioreactor on the flow pattern in the reactor. Solution of the microbalance equations for momentum and mass transfer is required. These methods are very complicated, and frequently, many simplifications are required. Nevertheless, these fundamental models



allow us to obtain a complete description of the process that is beneficial both for scale-up and for optimal operations at the production scale.

## **2. Semifundamental methods**

In these methods, simplified equations are applied to obtain a practical approximation of the process. The parameters obtained will be scale dependent, and thereby the influence of scale on the process can be examined by model simulations. However, despite of the extensive simplification of the problem when changed from the fundamental models to semifundamental models, the complexity of the model is still substantial.

## **3. Dimensional analysis**

Dimensional analysis is based on keeping the values of dimensionless groups of parameters constant during a scale-up. In physical meaning, the dimensionless groups, often called dimensionless numbers, are ratios of rates or time constants for the different mechanisms involved. Then, if all the dimensionless groups are kept constant, the relative importance of the mechanisms involved in the process will not change during scale-up.

It is often impossible to keep all the dimensionless groups constant during scale-up, hence one has to determine the most important group and deemphasize the rest. Moreover, noncritical application of the approach may lead to technically unrealistic values, e.g., for power consumption and stirrer speed in the fermenter.

## **4. Rules of thumb**

The rules-of-thumb method is the most common method. The scale-up criterion most frequently used are; 1) constant specific power input ( $P/V$ ), 2) constant  $KLa$ , 3) constant tip speed of the agitator, and 4) constant dissolved oxygen concentration. The different scale-up criteria normally result in entirely different process conditions on production scale. It is impossible to maintain all the parameters in the same ratio to one another.

Application of the rules of thumb method is very simple, but it is also a very weak method. There may easily occur a complete shift in the limiting regime above a certain scale.

In practical application, all four methods are used in combination with each other and sometimes the trial-and-error method must also be included. The success of scale-up processes are usually confirmed by experimental results which show that there is no difference between small and large scale production carried out under the same operating condition.