1. INTRODUCTION

A. Methods of Disruption

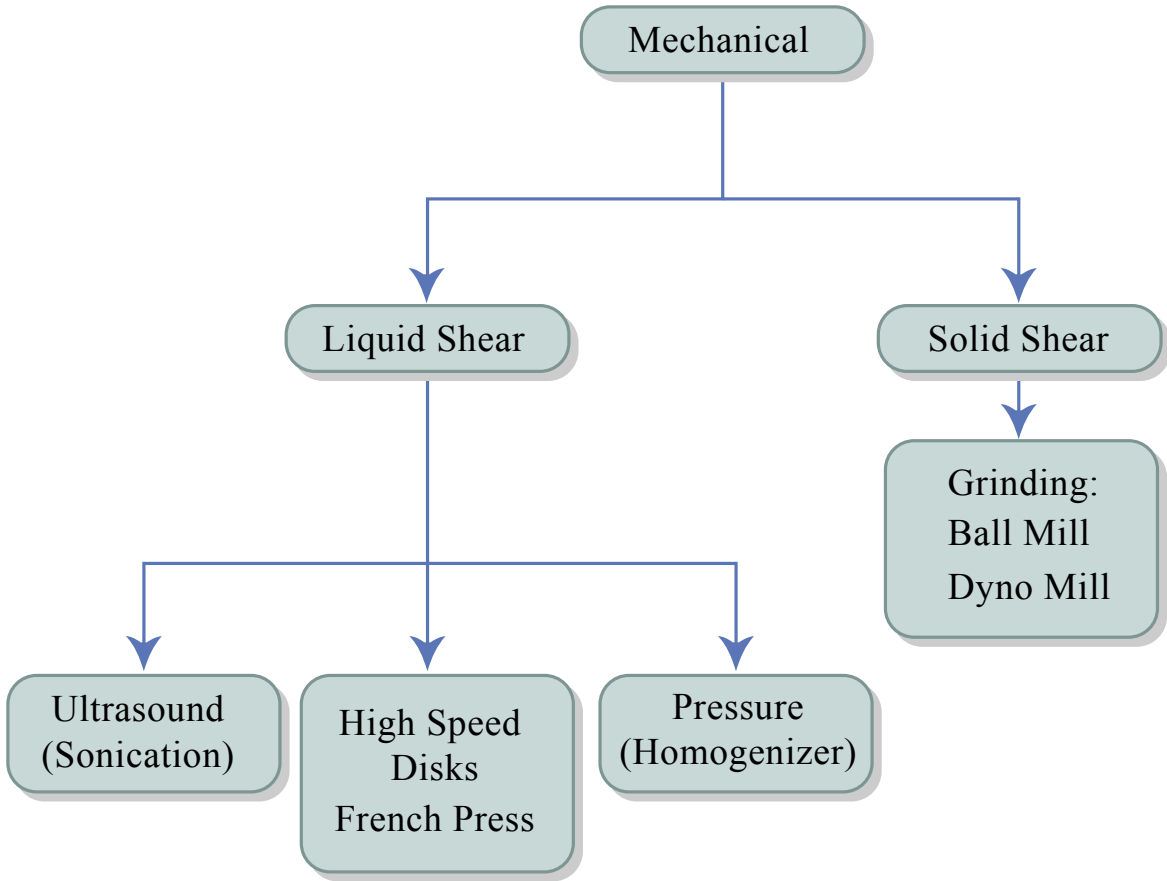


Figure by MIT OCW.

B. COMMENTS ON CELL DISRUPTION BY LIQUID SHEAR

- **Ultrasound: Sonication**

Advantages:

- Efficient (~ 95% Disruption)
- Rapid (Seconds to Minutes)
- Effective Against Many Organisms
- Simple to Operate
- Small Volumes (Milliliters)
- Ease of Containment (No Moving Parts)

Disavantages:

- Large Amount of Heat Generated
- Laboratory Usage (Not Scaleable)
- Low Cell Concentration

- **High Speed Disks:**

Advantages:

- High Disruption Efficiency
- Moderate Cell Concentration
- Available in Different Sizes

Disadvantages:

- Heat Generation
- Difficult for Containment
- Safety Issues: High Speed Rotating Disks

- **French Press:**

Advantages:

- Easy to Use
- Small Samples
- Moderate Concentrations

Disadvantages:

1. Not Effective for All Cells
2. Small Samples
3. Laboratory Tool

- **Pressure: Homogenization:**

Advantages:

- High to Reasonable Efficiencies Using Multiple Passes
- Effective Against Multiple Organisms
- Moderate Cell Concentrations
- Excellent on Scale-Up: Definable Scale-Up Parameters
- Reasonably Simple to Operate
- Continuous Operations
- Capable of Large Volume Operations

Disadvantages:

- High Pressures Needed
- Liquid Leakage: High Pressure Seals
- Multiple Passages for High Efficiency
- Containment Difficulties: Seal Leaks
- Moderate to Low Cell Concentrations
- High Viscosity: Release of DNA
- Maintenance: Erosion of Valve

C. COMMENTS ON CELL DISRUPTION BY SOLID SHEAR

- **Grinding: Dyno Mill:**

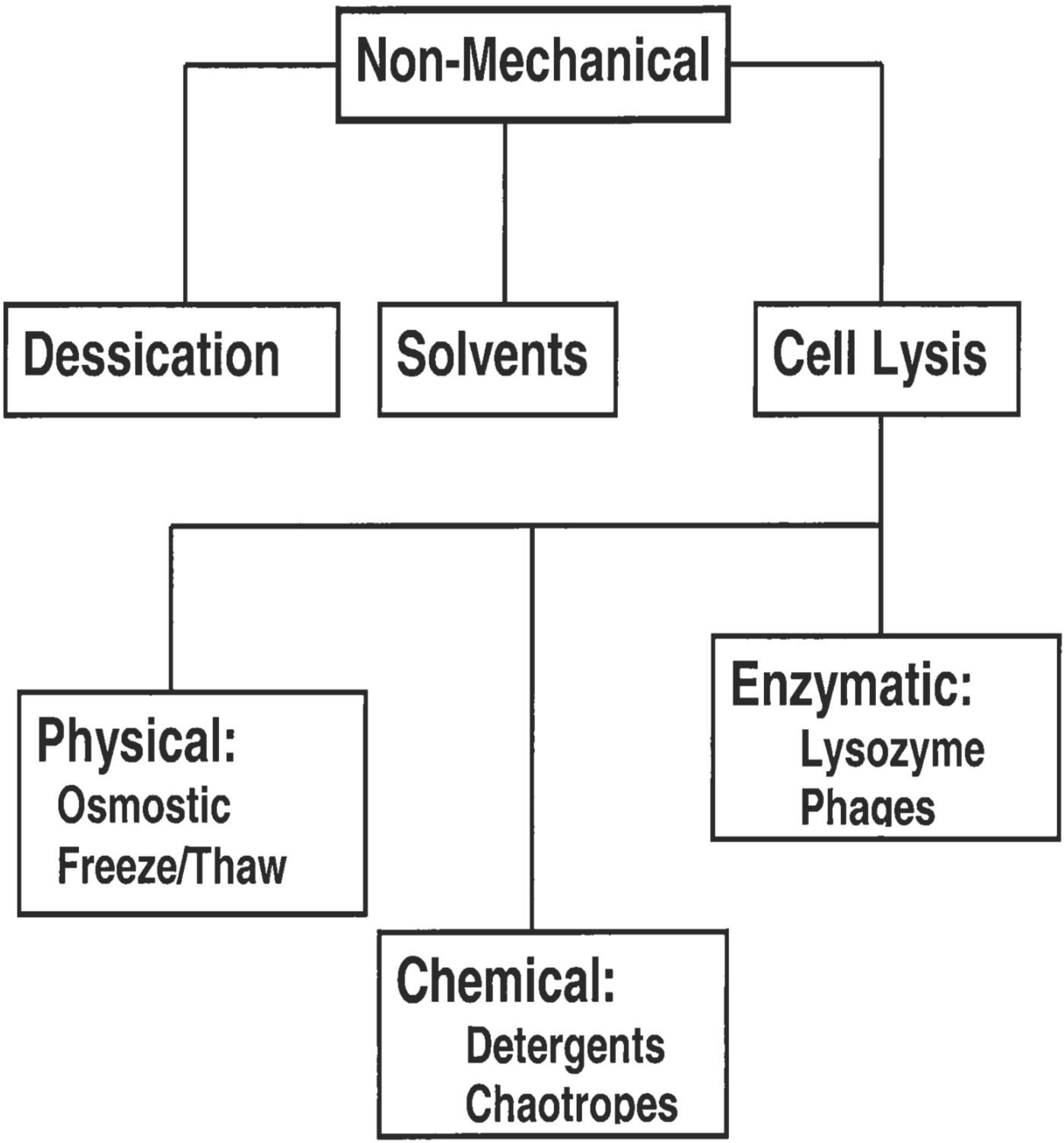
Advantages:

- High Disruption Efficiency
- Effective Against Multiple Organisms
- Low Pressure: Ease of Containment
- High Cell Concentrations:
  - Slurries up to 500 Grams/Liter
- Continuous and Batch Operations
- Available in Various Sizes
- Definable Scale-Up Parameters
- Low Maintenance: No High Pressure Seals, Minimal Number of Moving Parts

Disadvantages:

- Attrition of Beads
- Contaminating Products
- Parts and Service: European → USA

D. Non-Mechanical Cell Disruption



E. COMMENTS ON CELL DISRUPTION BY NON-MECHANICAL METHODS

- **Dessication:**

❖ Reasonably Simple
❖ Effective for Large Samples: e.g.Yeast and Vegetable Extracts

♦ *Additional Process Steps Needed: Cell Concentration, Dehydration*
♦ *Not Always Effective Against All Cells*
♦ *Difficult to Predict Performances*

- **Solvents:**

❖ Organic Solvents: Hexane, Benzene
❖ Effective to Permeabilize Cell Membrane
❖ Used Frequently in Cell-Free Systems

♦ *Toxicity of Solvent*
♦ *Potential Damages to Proteins*
♦ *EPA and OSHA Considerations*

- **Physical: Osmostic Shock:**

Concept:

- Inducing a Rapid Change in Salt Concentration
- Types of Salt: NaCl, Glycerol, Sucrose
- Disrupting Cell Wall to Release Intracellular Contents

Advantages:

- Reasonably Simple
- No Complex Equipment
- Operate at Low Temperatures
- Does Not Effect Protein Structure
- Effective Against Animal Cells and Red Blood Cells

Disadvantages:

- Not Always Effective Against Microorganisms
- Introduction of High Salt Concentration and Subsequent Removal is Necessary

- **Freeze/Thaw:**

Concept:

- Freezing Cell Paste and Followed by Thawing
- Permeabilizes Cell Wall
- Intracellular Contents Released Upon Thawing

Advantages:

- Cell Paste Can be Handled in Aseptic System
- Ideal Way to Schedule Timing to Hold Product
- Relatively Simple and Convenient
- Relatively Mild Method
- Able to Handle Large Samples

Disadvantages:

- Not Always Effective Against All Microorganisms
- Protein Release Efficiency Organism Dependent