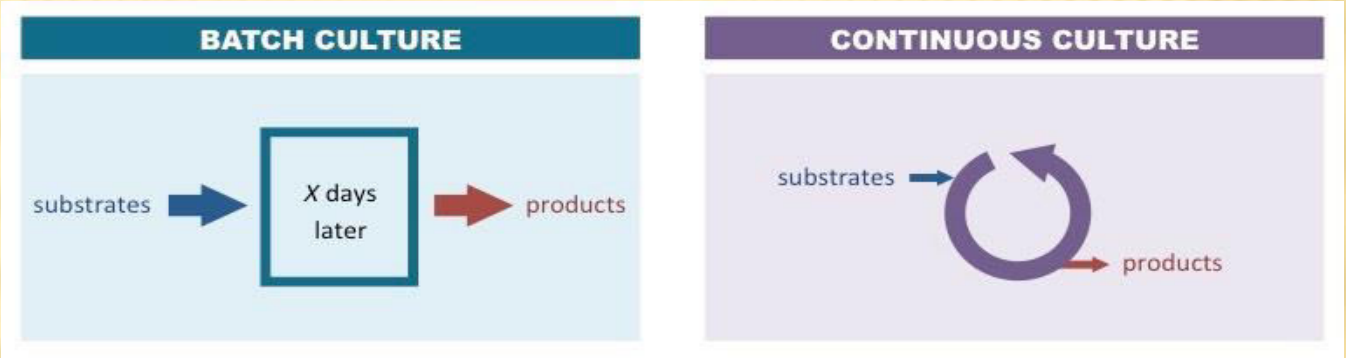
## Batch Culture

•Fermentation is carried out in a closed fermenter, with nothing added or removed during the process

•Microorganisms and nutrients are left for a set period of time, during which the nutrient stock is depleted

•Advantage the fermenter can be used for different reactions with each separate use

•Disadvantage : in significant periods of idle time between use, resulting in higher costs




## Continuous Culture

•Fermentation is carried out in an open fermenter, with nutrients added and product removed at a steady rate throughout

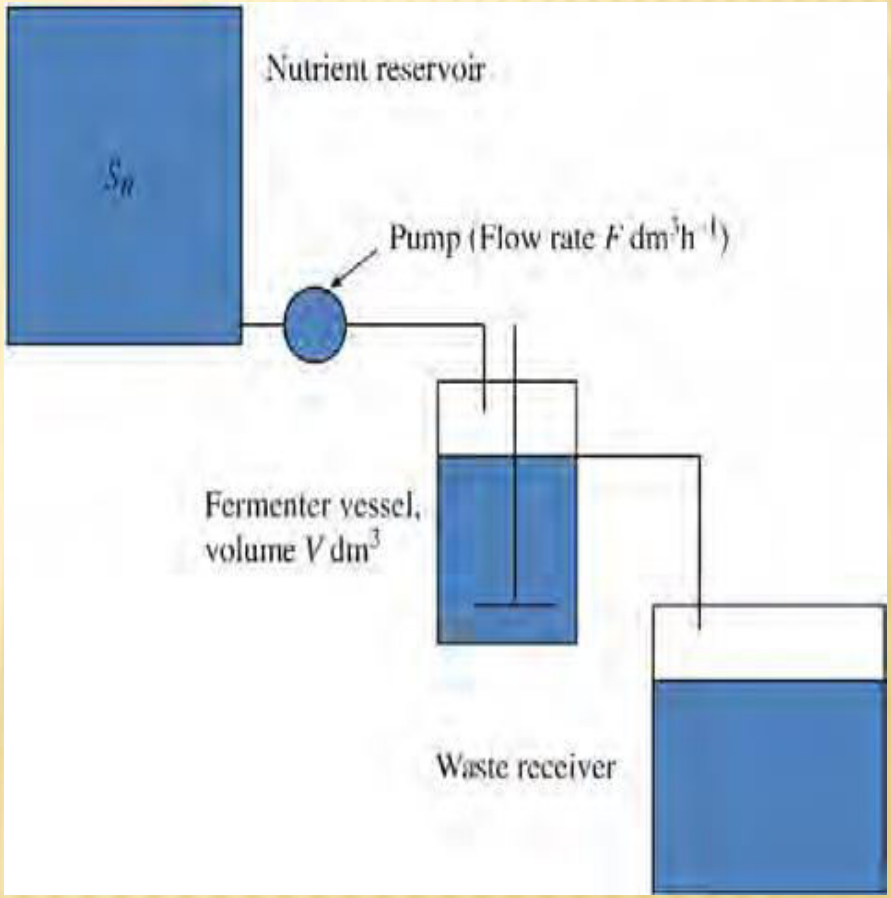
•Advantage: This results in a continuous reaction with no idle time, reducing labour costs and increasing product yields

•Disadvantage : higher risk of contamination due to the constant adjustments

•Continuous fermentation is feasible only when the inoculated cells are genetically stable

# CONTINUOUS CULTURE

- Batch culture may be prolonged by adding fresh medium to the vessel.
- Growth is substrate limited (ie, by some component of the medium), and not toxin limited,
- Exponential growth will proceed until the additional substrate is exhausted



The flow of medium into the vessel is related to the volume of the vessel by the term dilution rate, *D, defined as:*

$$D = F/V$$

where *F is the flow rate ($dm^3\ h^{-1}$) and V is the volume ($dm^3$).*

Thus, *D is expressed in the unit $h^{-1}$.*

The net change in cell concentration over a time period may be expressed as:

$dx/dt$= growth- output

or

$= \mu x - Dx$

Under steady-state conditions the cell concentration remains constant, thus *$dx/dt = 0$ and*

$$\mu x = Dx$$

and

$$\mu = D$$

under steady-state conditions the specific growth rate is controlled by the dilution rate. the dilution rate may be used to control the growth rate of the culture

continuous culture of this type is controlled by the availability of the growth limiting chemical component of the medium and, thus, the system is described as a chemostat, and described by Monod in 1942

$$\mu = \mu_{max}\, s/(K_s + s)$$

At steady state, $\mu = D$, *and, therefore,*

$$D = \mu_{max}\, \acute{s}/(K_s + \acute{s})$$

where $\acute{s}$ *is the steady-state concentration of substrate in the chemostat, and*

$$\acute{s} = K_s D/\, \mu_{max} - D$$

substrate concentration is determined by the dilution rate

An alternative type of continuous culture to the chemostat is the turbidostat, where the concentration of cells in the culture is kept constant by controlling the flow of medium such that the turbidity of the culture is kept within certain, narrow limits.

This may be achieved by monitoring the biomass with a photoelectric cell and feeding the signal to a pump supplying medium to the culture such that the pump is switched on if the biomass exceeds the set point and is switched off if the biomass falls below the set point.

If substrate is depleted below the level that supports the growth rate dictated by the dilution rate

1. **The growth rate of the cells will be less than the dilution rate and they will be** washed out of the vessel at a rate greater than they are being produced, resulting in a decrease in biomass concentration.

2. **The substrate concentration in the vessel will rise because fewer cells are left in** the vessel to consume it.

3. **The increased substrate concentration in the vessel will result in the cells** growing at a rate greater than the dilution rate and biomass concentration will increase.

4. **The steady state will be re-established.**

Thus, a chemostat is a nutrient-limited culture system in which the concentration of cells can be describe as

$$\bar{x}=Y(S_R- \acute{s})$$

$\bar{x}$ is the conc. of cell at steady-state in the chemostat

The basic chemostat may be modified in a number of ways but only two are very common extra stages (vessels): **MULTISTAGE SYSTEMS**
and
the feedback of biomass into the vessel: **FEEDBACK SYSTEMS**

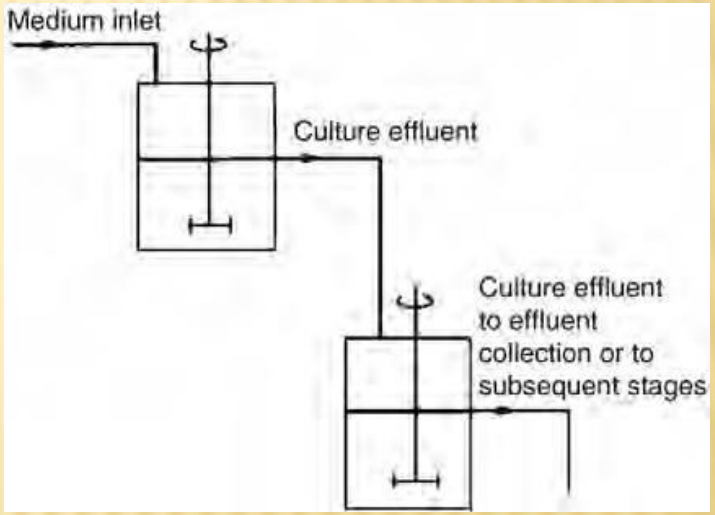
## MULTISTAGE SYSTEMS

Different conditions prevail in the separate stages.

Advantageous in the utilization on multiple carbon source and production of secondary metabolites

In dual system the second stage act as a holding tank where the growth rate is much smaller and thus producing secondary metabolites as in the case of *Klebsiella* were it was grown in a mixture of glucose and maltose.

Glucose was utilized in first stage whereas maltose in second. Experiment by Harte and Webb (1967)



# FEEDBACK SYSTEMS

Here, the biomass conc. is greater than $Y(S_R\text{-}s)$

Biomass conc. may be achieved by

Internal Feedback: Limiting the exit of biomass

External feedback: returning a portion of the concentrated biomass to the grown vessel

| BATCH CULTIVATION | CONTINUOUS CULTIVATION |
|---|---|
| 1. The bacteria are inoculated into the bioreactor (always stirred tank bioreactor).<br>2. Then, under certain conditions (temperature, pH, aeration, etc.) the bacteria go through all the growth phases (lag, exponential, stationary). | 1. The fresh medium flows into the fermentor continuously, and part of the medium in the reactor is withdrawn from the fermenter at the same flow rate of the inlet flow.<br>2. The bacteria is grown under certain conditions (temperature, pH, aeration) |
| Advantages:<br>• can be used for diff reactions every day.<br>• Safe: can be properly sterilized.<br>• Little risk of infection or strain mutation<br>• Complete conversion of substrate is possible | Advantages:<br>• Works all the time: low labor cost, good utilization of reactor<br>• Often efficient: due to the autocatalytic nature of microbial reactions,.<br>• the productivity can be high.<br>• Automation may be very appealing.<br>• Constant product quality |
| Dis-advantages:<br>•High labor cost<br>•Much idle time – Sterilization, growth, cleaning<br>•Safety – filling emptying, cleaning. | Dis-advantages:<br>•promised continuous production for months fails due to a. infection. b. spontaneous mutation of microorganisms to non producing strain |