IB01x - 4.3 - Fermenter operation

Until now we have seen some large scale fermenter designs. They have different shapes and come with different geometries. How they are being used for the process depends on the mode of operation. Typically there are three different ways how large scale fermenters can be operated.

These are the batch operation, the continuous operation and something in between called fed-batch operation. In this unit I will treat these one by one describing the pros and cons, followed by a mutual comparison. Let's first start with the batch process. This is the most simple mode.

At the start of the fermentation all the medium with the salts and the nutrients that the microorganism needs are present inside the reactor, including the renewable feedstock that the cell will convert into product. There is no additional inflow of substrate and nutrients and no outflow of fermentation broth. In case of an aerobic process oxygen can be supplied via the gas phase.

These graphs represent the amount of biomass on a logarithmic scale and the growth rate during a typical batch process. Before start, the media and bioreactor are prepared and sterilized. Fermentation is started by adding a small amount of cells, called the inoculum . After a short adaptation period, also called the lag phase, they will start to consume the feedstock and grow. Because all the nutrients are in excess , they will grow at their maximum growth rate μ_{max} and in the meantime make product. There is no supply of substrate, so after a certain time the substrate is depleted , or another factor has become limiting, for example the nitrogen or phosphorus source. Growth stops and the fermentation enters a stationary phase. This point determines the end of the fermentation and in practice the process will be stopped. If you continue, the cells will start to die, which is called the death phase, resulting in a decrease in biomass.

Here we plotted the biomass amount on a normal scale and included the amount of substrate and product in the fermenter during the batch process.

It illustrates that during the exponential growth phase the actual conversion to product can take place. This is logic if the cells themselves are the product, for example bakers' yeast or lactic acid cultures. But it could also be a metabolite or enzyme that is produced by the microbes and excreted.

A big advantage of this mode of operation is that it's very simple. You have a sterilized reactor, and start by entering fresh microbes and then you more or less wait until the batch is completed. The cells can grow with high speed, in an exponential way, usually under optimal growth conditions so the process is relatively short and can be completed (depending on the microorganism that you have) in one day or maybe a couple of days. The disadvantage is that you cannot very well control the environment of the microbes, except pH and temperature, and maybe the dissolved oxygen concentration. The concentration of cells, substrate and product change all the time. Further, the cells usually grow close to their maximum speed and it is not a given that these are also the optimal conditions for product formation. Moreover, this is very often not the case, and the key performance indicators for product formation (titer, productivity, yield) can be poor.

Let's have a look at the biomass balance during the batch phase as presented before in the second week by Sef. Here you see in mathematical terms that you can describe the growth of cells over time via the change term and this should equal the reaction term. There is no in- and outflow to the vessel. You can solve this expression and it says that the amount of biomass increases over time, depending on the initial value and the maximum specific growth rate, following an exponential profile. For product a similar expression can be set-up, using the product balance.

Now let's move to the chemostat. This is a continuous operation characterized by a constant inflow of medium, containing nutrients and feedstock for the microorganism. In a continuous operation mode there is also a flow going out of the reactor containing fermentation broth. Under industrial conditions, cells are not able to grow at maximum growth rate because it is determined by the dilution rate as I will show later in this unit.

To start a chemostat experiment you begin with a batch mode with little product formation. When sufficient biomass is formed, you start feeding and removing broth with a certain dilution rate and set the growth at the optimum for product formation. This is the steady state phase and can last up to a few weeks. In this phase, all the q-rates and all the compound concentrations in the fermenter remain constant! In the last phase typically the microorganism loses its ability to make product, due to degeneration of the strain and the process is stopped.

Continuous operation is in theory the preferred way, because there is a steady state, at which you operate the process under optimal conditions and you have very little disturbing dynamics. So from a control perspective this is very beneficial: it is optimal and relatively simple. In particular, you can set an optimal environment for the cells, so that they produce most efficiently the product that you want to have. A disadvantage is that it may take a few days before you arrive at the optimal working point with optimal performance. And with the currently used microorganisms it is often observed that they are not capable of keeping that optimal activity for too long, and usually after one or two weeks you will have to stop the fermentation, and restart afterwards. In addition, there are higher risks of contamination. So in practice you will not have a long-term continuous operation, and for this reason this mode is only rarely encountered in industry.

For the mathematical expression of the continuous phase, the accumulation term in the biomass balance is equal to zero, or the process is in steady state. When solving the balance you will see that under ideal conditions the specific growth rate equals the outflow rate of biomass divided by the total amount of cells present. This is what we call the dilution rate, D.

This relation clearly shows that the specific growth rate in a chemostat ONLY depends on the dilution rate, which you can set by tuning the outflow of the fermenter. This makes the chemostat a very powerful instrument.

Combining the needs for simplicity and high performance, a compromise has emerged in the industry, which is called the fed-batch mode of operation. A Fed-batch usually starts with a batch phase where you try to grow an initial, sufficient amount of cells. After the batch phase you start feeding the limiting substrate in such a way that the conditions for the microbes are approaching optimality for product formation. This can also take a couple of days or even weeks, and it needs to be said that a steady state is never achieved and product formation is mostly sub-optimal. The fed-batch process is run until the capability of cells to make the product is decreasing, just like in a continuous culture. Whenever this occurs one needs to stop the fermentation, empty the tank, harvest the product via downstream processing and start the cycle all over again.

Let us take a look at the biomass growth in the fed-batch process. First there is a batch phase, during which the growth rate is maximal. As soon as the substrate is depleted, the feed will be started. This results in a much lower specific growth rate, usually lower than 0.02 per hour, which is maintained by an exponentially increasing feed rate. However, soon the substrate conversion rate will become limited by a transport step, for example oxygen transfer, removal of CO_2, cooling or substrate mixing. As a result, the feed rate cannot be increased further, and the specific growth rate will drop, down to values where product formation will become too low to continue.

The fed-batch is a very convenient mode of operation and will result in higher productivities, biomass concentrations and product titers than a batch process, and not so much below the optimal conditions in an ideal continuous process. For these reasons, this has been the preferred mode of operation in industry so far. More than 80% of the industrial processes are operated in this way. I will not show the biomass balance for the fed-batch. This is more complex and beyond the scope of this course, but can be solved well with computer simulation programs.

In conclusion, when you compare the different modes of operation you can again recognize a number of pros and cons. Important aspects of operation mode are: fermentation time, product titer, productivity, ease of operation and maintaining sterility. These are different criteria that you can use to make the best choice for your system. But all depend on the microorganism and the product that you want to make. For the PDO case we have chosen continuous operation, because that is the best mode of operation, once you have optimized the microbe. And, it also simplifies the mathematics for the design.

That’s all for now. See you next unit!