## IB01x - 2.4 - Learning about the process: Gas phase balances

Last time we have discussed how to calculate conversion rates of compounds in the broth in mol/h using broth balances, and why these rates are economically relevant.

However the calculations of rates from broth balances was not yet complete, because volatile compounds in the broth can also be found in the gas phase in the fermenter.

As stated before, substrate, product and biomass rates cannot be measured, they are calculated from balances over the broth and measurements of concentrations, flow rates and volumes in the same broth.

Previously we have set up the balances for glucose and PDO in broth and we calculated the corresponding rates.

Here I also added the biomass broth balance, using a similar approach. Here the boundaries of the system are the broth volume in the fermenter.

But now we are going to look for compounds in the gas phase, so the boundaries will change.

Let's take for example oxygen which the organism could consume.

This oxygen first needs to dissolve into the broth of the fermenter, before it can be taken up by the microorganism. So oxygen goes from the gas phase to the liquid phase.

Let's draw our system. We have a broth phase, blue, with dissolved oxygen but now we also have a gas phase, which consists of all the gas bubbles present in the fermenter. We have an inflow and outflow of broth which contains oxygen. The oxygen in the gas phase will dissolve in the broth. This is the transfer rate of oxygen, named TN,O in mol O2 /h. The dissolved oxygen will be converted by the organism with rate RO, also in mol O2 /h So our broth O2 balance can be written as indicated. It has the dynamic term, the conversion term and inflows and outflows, but we also have the transfer term. This balance of oxygen, should allow us to calculate RO in mol O2/h. And if we inspect the balance, we can see that we can measure outflows, inflows and their dissolved oxygen concentration. However, we have now also this transfer term TN,O. This is again a rate which we cannot measure as such. It needs to be calculated from a balance. And the logical choice is of course the O2 balance over the gas phase, because TN,O comes from the gas phase into the broth phase.

In the gas phase a certain amount of gas is present, called NG in mol gas, and that gas contains a mole fraction of oxygen yO. We have an inflow of gas and an outflow of gas, all in mol gas /h. These gasses contain mole fractions of oxygen and in addition we have the transfer term, TN,o. The oxygen balance for the gas phase is shown below. It has a dynamic term, the 0 indicates that there is no conversion of oxygen in the gas phase, and we have a flow in of oxygen from air, and a flow out of oxygen. And there we see the transfer term, but now in a negative fashion.

The system is operating in steady state, so the accumulation term can be put to zero. And like I said, there is also no conversion of oxygen, which explains the second zero. So now we can rewrite the gas phase oxygen balance in steady state as indicated, and we can rewrite that equation to obtain our TN,O in measurable quantities, as we can see from this balance.

So now we have our measurement strategy to obtain RO, which we can see from the combined O2-balances for broth and gas phase. Once again if we assume steady-state, we can combine these two balances and we get the following expression for the oxygen consumption rate.

Another important rate is the CO2 production rate RCO2.

In a similar way we can set up the steady-state CO2 balances in broth and gas phase.

These balances show what we need to measure to calculate RO and RC.

However, there is one measurement problem, which is the outflow of gas FN,out.

Gas flows are measured by mass flow meters, of which the calibration line depends on the gas composition. This is no problem for FN,in, which has a fixed composition e.g. air.

The composition of the off-gas is variable, because it depends on the consumed O2 and produced CO2. The solution is to use the N2 gas phase balance to calculate FN,out.

So summarizing, we can obtain the production rate of CO2 and oxygen from 3 balances, measuring O2 and CO2 fractions in gas and liquid flows and measuring the gas inflow rate.

From broth and gas phase balances we have obtained important information to understand and improve the fermentation process. Now suppose we have calculated, using balances and measurements, the production rate of CO2, RC, for a given system.

We have already calculated the substrate consumption rate Rs, product formation rate Rp, biomass production rate Rx. So now we can now check our system for inconsistencies. Because the consumption rate of carbon in the form of glucose should equal the sum if the carbon present in the product rate, biomass rate and carbon dioxide rate as indicated in this sheet, which is called carbon conservation.

What if you find out that the calculated carbon intake from Rs is higher than the carbon going out in the form of product, biomass and carbon dioxide? What does this suggest?

There are multiple possibilities. For example, the cell converts your precious carbon from the substrate into something else of which you were not aware. Suppose that you are missing 20% carbon. Well, stick out your flag! Because now you have a target to work on, offering possibilities to improve the process economy.

But now suppose that the carbon output is larger than the carbon input in the form of substrate, what does that mean?

Well there could be many reasons for that, but the most obvious reason is that you have measurement errors in your procedures. Because it is impossible that there is more carbon coming out of the system than what you are feeding.

So therefore, certain measurements must have systematic errors. And I hope you understand that it is very important to find these errors, because these measurement errors compromise all the rates and economic indicators which you have calculated from these measurements and proper balances.

So summing up we are interested in consumption and production rates of microorganisms for all nutrients and products. These rates are calculated using a compound balance for each individual compound, for this we need to define the system boundaries, such as gas and broth boundaries. Setting up the balances tells you what you need to measure to calculate these rates. And it is very much advisable, to measure also the compounds for example oxygen and carbon dioxide in the gas phase to check, using element conservations, if the calculated rates violate for example these conservation principles. For example carbon as we have here shown. But there are more conservation principles such as conservation of the atoms N, O, H leading to even more checks!

Such consistency checks usually tell you that you don't have the full understanding of the process, there are hidden mechanisms, or you measurements are wrong.

With these balance calculations and consistency checks you understand your process much better, resulting in actions to improve the economy. And, of course finally you can tell your boss how much money he is making.