Hi guys,
Welcome to this week's online session, which you can watch whenever you want. My name is "Jenny" and I will guide you through today's session which packed with lots of interesting information regarding VO2max testing. As you might notice, my voice is generated using artificial intelligence. Everything I am saying was transcribed from Jasper. Also notice, that the entire transcribt of the session is uploaded on moodle.
Here you can see the slide where we left off last week. In the previous session, we wanted to understand the biophysical inner workings of metabolic carts. We looked at how the airflow rate passing through the system is measured, as well as the different systems that are used to determine the fractions of O_2 and CO_2.
Based on the percentages of O_2 and CO_2 together with the overall airflow, the system can then calculate VO_2 and VCO_2. From a technical perspective, we now have a general understanding of how these systems work, and I think it is always important to know these details.
I often keep this in the back of my mind when thinking about situations such as applying to an institute that works with spirometric devices. During an interview, they will eventually ask whether you have any questions to them regarding the job etc. This is a good time to distinguish yourself from other applicants by asking smart questions. A very good question to ask would be: "What kind of devices do you work with?"
When thinking about the different systems available for spirometric testing, you could ask questions such as:
Are you working with breath-by-breath systems?
Are you using a mixing chamber?
How are O_2 and CO_2 analyzed in your system?
What kind of measurement error do you typically observe?
How often do you calibrate these systems?
By asking such questions you can not only show interest but also demonstrate the depth of your knowlege. To my mind this would probably make a very good impression in an interview.
Now we want to examine how accurate these systems actually are.
There was a very interesting publication a couple of years ago — I do not remember the exact year — that investigated the accuracy of respiratory gas variables, substrate utilization, and energy expenditure measured from 15 CPET systems during simulated and human exercise.
CPET stands for Cardiopulmonary Exercise Testing.
In this case, when we talk about "simulation," we are not referring to the bioenergetic model we developed in previous sessions. Instead, a machine was used to provide controlled amounts of O_2 and CO_2 at predefined flow rates to the gas analyzers.
The advantage of this setup is that, because we know exactly how much O_2 and CO_2 are being delivered to the systems, we can validate the measurements obtained from the CPET devices.
In other words, normal breathing behavior is simulated mechanically, and the CPET systems then measure VO_2 and VCO_2. Since the exact reference values are known beforehand, researchers can directly evaluate how accurately the systems perform.
Validating these systems directly in humans is much more difficult because there is always biological variability involved.

For example, you could perform a VO_2max test on day one and repeat the same test on day two, then compare the VO_2 curves from both sessions. If the curves overlap well, this would indicate good reliability — meaning the system produces similar measurements repeatedly. However, this still would not necessarily prove validity.
Reliability can also be influenced by biological variability. The athlete may be in a different nutritional state, glycogen depleted, fatigued, or affected by many other uncontrolled variables. Therefore, even if the machine itself works perfectly, differences between tests may still occur because of human physiology rather than technical measurement error.
This makes it very challenging to determine how valid these systems truly are.
That is why studies using simulation machines are so valuable. By delivering known amounts of VO_2 and VCO_2 to the CPET systems and comparing the measured outputs with the known reference values, researchers can properly evaluate system accuracy.
The study examined several dependent variables, including:
VE: expiratory ventilation
BF: breathing frequency
VO_2: oxygen consumption
VCO_2: carbon dioxide production
RER: respiratory exchange ratio
The researchers compared different CPET systems and evaluated how accurately these variables were measured.

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On this slide, you can see some of the different CPET devices that were evaluated in this study.
In the bottom row, you can see the different manufacturers, and above that, several technical characteristics of the systems are listed.
If we start with the first column, the Vyntus CPX, we can see that this device is manufactured in the USA. The system type is breath-by-breath, and several technical specifications are shown as well. Volume is measured using a turbine, O_2 is measured using a chemical fuel cell, and CO_2 is measured using a non-dispersive infrared sensor.
All of these components are very similar to the systems we discussed last week.
An interesting observation appears when we compare the different analyzers. For example, for the Vyntus CPX in the first column, the O_2 measurement system comes from Teledyne, California, USA. This means that the company does not manufacture its own O_2 sensor but instead integrates a commercially available sensor into its system.
If we move one column to the right and look at the Oxycon Pro, we can see that the manufacturer of the chemical fuel cell is actually the same. This is something we observe for many of these devices: the key components responsible for the measurements often come from the same manufacturers.
So even though these systems are sold by different companies, there is a good chance that many of the central components originate from the same factory.
The same pattern can be observed for the CO_2 measurement systems. Looking again at the Vyntus CPX, we can see that the non-dispersive infrared sensor is manufactured in the USA. The same is true for the Oxycon Pro.
Let’s also briefly look at the MetaLyzer 3B and the MetaMax systems. These are the third and second-to-last systems from left to right. Both are produced by the German manufacturer Cortex Biophysik.

Both systems are breath-by-breath analyzers and have very similar technical specifications. I specifically mention these devices because, to my knowledge, this is the type of system that is also used here at Ruhr University.
Finally, if we look at the VO_2 Master system, we can see that it only measures O_2 and does not measure CO_2. Some systems focus exclusively on oxygen measurements and therefore do not provide VCO_2 values.
This is important to keep in mind if you are considering purchasing such a device. Not all systems are capable of measuring CO_2, so it is necessary to carefully review the specifications beforehand.
You will also need a considerable budget. In the lowest row, you can see the approximate system costs. Prices range from around €6,100 for the VO_2 Master — which only measures VO_2 — up to approximately €70,000 for the Omnical V6, which also measures VCO_2.
This demonstrates that these systems can become very expensive.
Above the pricing information, you can also see the manufacturer-reported accuracy for volume, VO_2, and VCO_2 measurements. Most manufacturers claim an accuracy within ±3% or ±50 mL, depending on which value is larger.
This means that for very small measurements, the ±50 mL criterion becomes relevant, whereas for larger values, the measurement error should remain below ±3%.
Now let's take a look at what exactly was done in the study and how these systems performed in terms of measurement accuracy.

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Thank you, Jasper! You are sooo clever...
Here we can see the percentage error for all dependent variables, including ventilation, breathing frequency, VO_2, VCO_2, and the respiratory exchange ratio. On the far right side, the authors also provide the overall percentage error for each system.
The results are shown for every device, ranging from the Vyntus system in the first row all the way down to the Calibre system in the final row.
To better understand what these numbers actually mean, we should focus on the red box at the bottom of the slide. The authors defined specific thresholds for evaluating measurement agreement.
A system was considered to show:
good agreement if the error was below 3%
acceptable agreement if the error was below 5%
Based on these thresholds, the systems were classified accordingly.
In the red boxes that I added to the VO_2 column, I highlighted all devices that showed an average error below 3%. However, as you can see, none of these systems received two asterisks from the authors.
The reason for this is that not only the average error is important, but also the confidence interval around that error.
The rationale behind this is relatively straightforward. If we perform repeated measurements, overestimations and underestimations may cancel each other out, meaning that the average value becomes informative. However, if we only perform a single measurement, that measurement could theoretically fall anywhere within the distribution of possible error values.
Therefore, to classify a system as showing good agreement, both boundaries of the 95% confidence interval had to remain below 3% error.
Similarly, to achieve acceptable agreement, the entire 95% confidence interval had to remain below 5% error.

When we specifically examine VO_2 measurements, we can see that none of the 15 systems evaluated in this study achieved the criterion for good agreement between the simulated reference values and the values measured by the devices.
If we move to the right-hand side and look at the respiratory exchange ratio, we can see a somewhat different picture. For example, the Oxycon Pro MC — the third system from the top — achieved a rating of good agreement for the respiratory exchange ratio.
However, overall, especially when thinking about VO_2max testing, none of these systems demonstrated truly good agreement.
Importantly, this outcome did not appear to depend on whether the devices used a mixing chamber approach or a breath-by-breath approach.
In fact, when comparing the systems directly, there does not seem to be a clear difference in performance between breath-by-breath systems and mixing chamber systems.
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On this slide we can see a graphical representation of the numbers we have just seen in the table on the slide before.

In the upper row we can see the VO2 in percentage error for all the 15 systems that are tested and then if we were moving from the top left to the right we can see the VO2 and then last in this row the RER in percentage error.

In the bottom row we can see the percentage error for the calculation of the energy that is coming from carbohydrates on the bottom left side and then moving to the right side the energy that comes from fat in percentage error and lastly the overall energy expenditure in percentage error that we see in these systems.

You should now take a couple of minutes to look at these systems individually and work out for yourself why potentially the error that we see in the calculation of energy from carbohydrates and fats so the substrate utilization is so much higher in comparison to the error that we see in VO2, VO2 and RER in the top row.

Looking at the Y-axis for the energy from carbohydrates error for example it goes from minus 80 to 180 percent so very very right range whereas in the graph above so the VO2 percentage error only goes from minus 25 to 20.

So only from the scaling of the Y-axis we can see that the error is much higher in the calculation of the energy that comes from the substrates in comparison to the VO2, the VO2 measurement and also the measurement of RER.
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So far we have only looked at the average error that we see throughout all the steps that were tested.

Here we can see how the error behaves for different simulated VO2 values.

So we're looking at the left side of the graph here with 1000 ml per minute.

This would be the lowest age that has been simulated and on the Y axis we can see the percentage error.

Now for the different devices we can see how the error changes as a function of the simulated VO2.

Let's focus on the power cube Ergo which is the black line that we can see on this graph.

In the lowest age, so the 1000 ml per minute, the system underestimates the amount of VO2.

If we then go to the higher system it starts to overestimate the VO2 that has been brought to the system.

And then the error is a little bit lower but still there's an overestimation when we go to the final stage.

So the 4000 ml per minute.

This demonstrates that we cannot simply use a correction factor to correct our VO2 data.

But we need to keep in mind that the error that we see in this system might also be due to the function or to the amount of VO2 that is actually brought to it.

Also we can have a look at a different system.

For example the Metamax 3B which is in the pink color here on this graph which has a somewhat stable error.

So there's a little bit of an overestimation in the lower stages and then this overestimation goes a little bit down.

But there's still a little bit of an overestimation when we go to higher values but then the error becomes a little bit smaller.

But overall this system behaves a little bit more stable in comparison to what we see in the power cube Ergo.

Also we can see other systems.

For example the VO2 master which is in green on this slide which just behaves terribly.

So there's a higher underestimation of roughly minus 5 minus 6% in the lower stages and then as VO2 increases this error becomes even higher.

And then at the end we have an underestimation of roughly 15 or 16% here.

So definitely a very very high error when using this system.

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What does this now mean for practice?

Two questions I want you to answer.

First one is, would you perform a second-view or two-max test after a two-week long training period?

Second question, how would you interpret a change of 5% increase in VO2max for on the one hand the planar device and secondly the MetaLyzer 3B.

So in a fictitious scenario where you would test your athlete on day one and then you do a 12-week training program and then afterwards you want to test again you see a 5% increase how would you interpret this with regards to what you saw just in terms of validity of these systems.
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Now for the next slide we want to shift from the technical side of things more to the physiological side of things and how we can actually use these systems to test VO2 max individually for our athletes and for this I've made a very very short recap regarding the VO2 kinetics that we've already talked about to bring this back to our mind because we need to consider these kind of things and these characteristics of this metabolic pathway in order to make sure that we can actually test VO2 max.

So what we can see on the slide here is a sudden change in speed I showed you the slide before where a resting athlete starts to run at three meters per second and now the question is how does our VO2 system react to such a change in external load?
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So, view 2 takes some time to ramp up.

We can see this on the right hand here.

There's a view 2 on set time of roughly 2.5 to 3 minutes.

And afterwards, the view 2 will reach a steady state.

And then as soon as the exercise or the external load is terminated and this athlete goes back to resting, then view 2 will slowly go down again and has some inertia until it reaches the resting metabolic rate.
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We can divide the VO2 kinetics in different phases.

First we have this cardiodynamic phase where the VO2 will start to go up a little bit faster and then secondly it goes into the primary phase where we have this mono-exponential increase in VO2 until at some point the third stage it will reach a steady state.

Down in the lower part of this graph you can again see how we could potentially describe this mathematically so if you want to have a look at this function here or if you want to type it into Excel or maybe ask HBT to draw a graph when you use this type of function then you would see such a mono-exponential increase in VO2 where it takes roughly three to four times the tau value that you set in this function until a steady state is reached.
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Lastly, on this slide we can remind ourselves of the model that we have built.

So the ongoing on a metabolic cellular level that first the PCr system is becoming the phosphorylated and because of this then ADP and AMP or for the oxidative system mostly ADP will start to accumulate and this ADP is then a trigger for the oxidative system and as PCr system becomes more depleted, more ADP will accumulate and then there will be an answer of the oxidative system to use oxygen to actually produce ATP and then at some point in this example a steady state will be reached between the defaults for relation of the PCr system and the ATP that is actually used for local motor tasks.

And because of this we will then also see a steady state reaction in the VO2 at some point.
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You can go ahead and read through the summary here on the first bullet points.

We have the causal chain of VO2 increase, then say one time to reach the steady state, which I have explained.

And then the third one, which has not been a part of the summary, but we remember, for example, from the joiner model, or also from the calculation of VO2, that we can do this or we can approximate the VO2 if we know the efficiency and the external load.

So if we just multiply these two things with each other, then we can calculate the VO2 response that we are expecting.

Lastly, here we have the phases of the VO2 response in the summary slide.

Now we want to put this knowledge that we have gained to actual practical use, and we want to be able based on this knowledge to make up the characteristics of a test that we would need in order to actually measure VO2 max.

For example, what kind of external load would we need to measure VO2 max for a fictitious athlete?

So what kind of duration does this athlete need to perform this external load for so we can actually measure the VO2 max?

So before you go to the next slide, you should, for yourself, one or two minutes, think about how you could set these parameters in order to actually measure VO2 max, what kind of characteristics would the test need to fulfill in order to measure VO2 max?

Alright, so think about the fuselage ongoing inside the muscle and base on this extrapolate to a test design that would make the measurement of VO2 max possible.
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I hope you guys had some good ideas regarding a test design for VO2max.

Let's first look at the external load and how we can set this in order to measure VO2max.

First, the external load has to be above VO2max.

If I have a VO2max of 65, somewhere in this range and I'm running at 2.5 meters per second, I will not be able to measure VO2max unless I have a very, very terrible running economy.

On the other hand, if I set the external load to high, fatigue may occur before VO2max is reached because I have this 2.5 to 3 minutes, somewhere in this range time interval, that the VO2 kinetics need in order to answer to the external load.

If I were to go out and do a 100 meter or 200 meter sprint, at the end, I will be totally fatigued.

But the time is not enough to reach VO2max if I want to measure this at the level of the mouth.

Let's look at an example that I have described here below.

If we want or if we are expecting VO2max roughly around 60 foreign athletes, and I'm assuming a normal running economy which would be in the range of 12.5, I could go ahead and calculate 60 divided by 12.5.

I would assume in terms of VO2 demand that I at least need a velocity of 4.8 meters per second.

And the reason, again for this is that the VO2 itself is limited by the work rate, whereas VO2max is limited by the oxidative pathway, the amount of mitochondria that I have, the amount of hemoglobin that I have, and so on, so all these kinds of physiological things.

But I cannot sit here in my chair, think about going to VO2max, and expect that I will measure VO2max at the level of my mouth.

Therefore, I need to make sure that the work rate is high enough in order to even elicit VO2max.

And for this, it's important to have this in the back of your head.

VO2 is limited by the work rate, whereas VO2max on the other hand is limited by the oxidative pathway.
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Let's bring this idea of VO2 being limited by the work rate and VO2 max being limited by the oxidative system.

Let's bring this idea back to our model here.

On the X axis we can see the concentration of ADP which goes hand and hand with a decreasing PCr concentration and on the Y axis we can see the activation of the oxidative and the glycolitic system.

For us let's now focus on the oxidative system which we can see here in blue.

In the red box I have marked you an external load that would be too low to reach VO2 max meaning physiologically the dephosphorylation of the PCr system is not high enough and therefore the ADP concentration is not high enough to saturate the metabolic pathway that works with oxygen so the oxidative phosphorylation.

Therefore if we were to increase the external load to a value that is high enough to actually reach VO2 max which we can see in the green box here.

The system is more dephosphorylated we have more ADP and because of this then we can measure VO2 max.

So this would be the metabolic reason why VO2 is limited by the work rate or if we think about this physiologically the dephosphorylation of the PCr system and then the ADP concentration which is the drive of oxidative phosphorylation.

VO2 max on the other hand is limited by the metabolic pathway itself and the physiological determinants of this so mitochondrhyngry density or to transport capacity and so on so these kind of things and how we can bridge these things from a testing perspective to a metabolic perspective at the level of the cell here.
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Shifting from the parameter of external load to duration, we have to keep in mind this inertia of the oxidative system.

Typically in order to reach, steady state let's say we need roughly four minutes to have a little bit of buffer between and what we expect here, right?

So we have this two and a half to three minutes.

Let's use a little bit of buffer.

So let's say the test has to be at least four minutes in order to reach VO2 max.

Typically what we see in practice is that eight to twelve minute durations are used for ramp life protocols if we want to measure VO2 max.

The reasoning behind this is that the duration has to be longer of the time for the VO2 inertia.

Doesn't make sense to have like a 20 second 30 second sprint test to measure VO2 max because the inertia of the VO2 systems to high.

On the other hand it has to be shorter than the depletion of substrate affects view the VO2 response.

If I were to do a two hour test to make an extreme example then glycogen depletion would definitely play a role and then because of depleting the depleting glycogen system there's a feedback to the calcium release and because of this at the contraction and will not be able to and bring or to actually bring VO2 up to the maximum values.

So these are the two things that we have to keep in mind in terms of duration.
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One strategy that we could now pursue in order to measure VO2max is what we can see on this slide here.

Every blue dot is measured VO2 value for let's say five minute stage.

So five minute stage at the external load that we can see on the corresponding x-axis was performed and then the VO2 was measured and then the last 30 seconds one minute of the stage was used and put here in this graph.

And then after we've performed the first test we can go ahead increase the external workload a little bit and do the same thing again measure the VO2 and put this in this graph here and we can see with increasing external speed how the VO2 reacts and we can see at the end this what is called the leveling off meaning that with increasing external load the VO2 response does not increase any further and this would be decoupling of increasing external load and VO2 response and this would be the major determinant of reaching VO2 max the plateau that we see here.
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The advantage of this process would be that we are very likely to get or to measure a true VO2 max and to actually identify a true plateau in the VO2 external load relationship, which would give us lots of confidence that we have actually measured VO2 max here.

The disadvantage of this is that this takes lots of time.

So let's say we are only using these 5 or 6 data points that we see here at the back end and if we say that we have a 4 to 5 minute stage every time and we push our athlete to very very high lactate concentrations, we need to make sure that the athlete is sufficiently rested until he goes to the next stage because this is a very demanding test.

This takes a lot of time.

Another criteria we should have in the back of our mind is that if we use this kind of test and we have to push our athlete to such high demands just to get the VO2 max value, we would need to have one or two days of rest before we do such a testing maybe afterwards, some kind of rest.

So it will put a huge hole in our training plan because we need to block a couple of days to actually measure VO2 max here to see if our training plan or intervention that we have done beforehand actually works and if we have an adaptation in terms of VO2 max.

And because of this, short or protocols were developed that we see on the next slide.
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Here we can see a classical ramp protocol that is typically used in a sports scientific practice.

I have distinguished here between these two kind of protocols that are slightly different.

So we have this ramp ramp and the step ramp what I call this.

So the step ramp would be an increase of 30 watts every 30 seconds.

So overall the increase is also one watt per second if we were to take the average of this and then it would be similar to what we see in the ramp here where there is one watt increase every one second.

And on the left hand side we can see how this is graphically visualized that we have warm up phase in the beginning for four minutes and then afterwards we go into this ramp protocol or the step ramp protocol and this would only be terminated if fatigue is reached at the end and based on this we have a protocol that we only have to do once.

We only have to push our air fee to exhaustion once and based on the VO2 data that we would gather here we try to calculate or we try to analyze the fire and measure the VO2 max.
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The problem with this approach is that it's sometimes hard to see if the true VO2max was actually measured.

In this graph here we can see the work rate for such a test on the x-axis and on the y-axis the measured VO2 in little per minute.

If we were to have scenario C or B we could ask ourselves have we actually measured VO2max.

So in scenario A it would be clear we have this leveling off so we would assume okay VO2max was actually reached during this test because we have an increase in work rate but we have a leveling off in the VO2 response which is the main criteria for VO2max.

But for scenario C or B it's really tough to say if VO2max was actually measured and because of this there were other criteria than the leveling off VO2 develop to assess if a true VO2max was reached.

So we have this first order criteria which is the leveling off of the VO2 work rate relationship and this is the most important one.

Secondly there were so-called secondary criteria developed which would be for example that the heart rate is 5 to 10 beats below the prediction.

So prediction would be for example for man 220 minus the age of a woman 20-30 minus the age.

Also they are looking at blood lactate so you could measure the blood lactate concentration after the end of this test and if this is above 8mm of the liter then the athlete would be exhausted and the highest VO2 value here would be considered VO2max or potentially you could also look at the respiratory exchange ratio and for this you will find different values so for some RER of higher than 1 is already sufficient for others 1.1 and even for others 1.15.

So based on the different numbers that we see here in the secondary criteria we can already understand that this is kind of vague and it's not a good criteria to assess if VO2max was reached or not.

Because for example if you look at the maximum heart rate prediction and when we do this with these formulas I've mentioned on a group level these formulas are quite accurate but if you look for somebody or if you look for an individual there is quite some variance with respect to the maximum heart rate that you can potentially see in this person and the formula.

So maybe you have somebody who is 40 years old man then 220 minus 40 the prediction would be 180 beats per minute but I can tell you this from practice on an individual level you can see somebody who is already exhausted after 150 and you could see other athletes that go up to even 195 beats per minute so there's quite some individual variance and from my perspective this makes this criteria in applicable.

The same thing goes for the blood lactate concentration.

So the highest blood lactate concentration somebody can sustain if we think about this physiologically again on the one hand must have to do with the glycolytic power.

So if we have somebody with a higher glycolytic power likely you will also go to higher lactate concentration.

If another thing that comes into play here is the buffering capacity.

If we have somebody who has for example supplemented sodium bicarbonate then maybe his buffering capacity is increased and because of this he can go to higher blood lactate concentration where somebody else is finished at lower blood lactate concentration.

The same thing is also influenced influences the respiratory exchange ratio.

So if somebody can go to higher lactate values because of this the bicarbonate system has to buffer more hydrogen ions and because of this more carbon dioxide will be produced by this athlete.

So we would see or we would expect a higher RER in somebody who has supplemented sodium bicarbonate or naturally has a higher buffering capacity than somebody else.

So these secondary parameters are not directly linked to VO2 but of VO2 max but can be manipulated by other metabolic parameters and therefore I would recommend using these parameters when you want to assess the VO2 max from such a ramp ramp or step ramp protocol.
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So what can we do now in cases C and B?

If we are unsure, if we have actually attained a true VO2 max or not, what we can do is we can perform a so-called verification test, meaning we are taking the last or the final stage that was reached during this step ramp and increase the work rate a little bit and perform another 4 minute, 5 minute, 6 minute, bout of exercise and see if the VO2 that we measure during this stage is higher than the VO2 that we have measured in the in the end of this ramp test or ramp ramp step ramp test protocol.

If we then see a plateau in VO2 here then we can be sure that we have actually measured the correct VO2 max.

The question for this obviously is what constitutes a plateau and there we can find different values in the literature.

For example here Taylor and colleagues they say that an increase less than 2.1 millilitre per minute per kilogram constitutes a plateau.

Another way to look at this would be to consider the error of VO2 that is given by the manufacturers which are roughly these 3% that we see here.

So if the VO2 that we measure is more than 3% below the expected VO2 then we could say that there is a plateau within our work rate VO2 relationship.

To make an example here again to calculate the VO2 that we expect or the metabolic demand here we could say that for 300 watts multiplied with the 12.5 so the volume of oxygen that we need per watt we would expect 3750 millilitre per minute or to have another example down here with this 330 watts times 12.5 we would expect roughly 4125 millilitre per minute.

Overall I must say I'm not the highest fan of this because there are also other factors that drive VO2 for example there is this base-to-metabolic rate that maybe puts VO2 to a little bit

higher values also we have to keep in mind that maybe some of the energy that is included in the VO2 response here actually comes from a glycolic system to the production of lactate so it's really tough to get to exact values here it's more like a ballpark estimate that we would expect.

The important thing I think is to perform verification tests when you're unsure and then to look individually if you would consider this being a plateau or not and develop your kind of criteria and district about these kind of things because a literature you will find different kind of criteria.
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Let's look at some actual data that we have gathered during a project couple of years ago.

Here you can see the VO2 response of an athlete performing a typical ramp test and we can see here that after the initial phase which was the ROM up, up to let's say 100 seconds roughly, the VO2 starts to increase in a more or less linear fashion until 352 seconds somewhere in this range and then at the end we see some more leveling off.

However, the data that we see here is unprocessed so this is the raw data that was measured by the metabolic card.

So for every breath this athlete has taken, we can see one data point so the VO2 will later per minute as a function of time.

The upper dashed line in gray that we can see in this graph constitute the highest value that was measured.

So the highest VO2 measured from this athlete.

Now we can see that at the same time we have a VO2 value that is much lower.

So just a couple of seconds after that when we go to the lower gray dashed line there's a VO2 value that is much lower.

So the question for us now is is this highest value that we see here actually the true VO2 max or maybe is this an outlier?

And because of this we need to filter our data to actually find a VO2 value that we can work with at the end.
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Here I have applied different rolling averages to the same data that we have seen on the slide before.

In black you can see a 5-brother average, in yellow, a 10-brother average, in orange 20 and in purple a 30-brother average.

On the top the horizontal dashed line for each color represent the highest value that we would get if we were to perform this filter on the entire data set.

And because of this we can see that there is quite some variance between, for example, the highest data point if we were not to process our data and would just look at every breath individually, which is here the gray dashed line.

So very high VO2 value, whereas when we go to the lowest one the 30-brother average, we see a lower value that is just a little bit above 5 liters per minute.

Whereas this highest value in the gray dashed line of no filter, this would be roughly 5.75.8 somewhere in this regard a little per minute.

So roughly 10-15% difference or maybe 17% difference even between a 30-brother average and as one breath average.

So basically no average at all.

And the other average averages, so 5, 10 and 20 lie between these two ranges.

So the question for us is now where we go ahead and use a rolling mean average, what is the perfect one?

What kind do we actually want to use because this highly affects the VO2 max that we will measure at the end?

On the one hand, we could say if we are just using the same method every time we will get a consistent outcome.

And yes, while this is true, I also have an example where not only the reliability, if you want to call it like this in this part here, is important but also that you get valid estimate.

Because if you are thinking about for example calculating substrate utilization from VO2 data, not from this data here because you can't do it in such a test.

But if you were to do it from a graded exercise test that has longer stages, it's not only important to get consistent values, but it's also important to get true values.

And that because if you are doing a race pacing strategy and you want to have your athlete push the power that is corresponding to let's say 60 grams of carbohydrate oxidation per hour or whatever value you want to use there, it's important to actually hit this point as precisely as possible.

And here it will not help you if you are off let's say 10% every time then maybe you can see a trend in the data that the athlete has improved, but if you want to specifically use this for pacing, it's important to also get a precise value and not only one that you can compare over time.

So there are these two kind of things we need to think about when performing filtering techniques that on the one hand we want to have consistent outcomes, but on the other hand, we also want to be as precise as possible.

On the next slide, there's no voice over you will just see multiple examples of spirometric data and how I filter them and you can just click through the slides and see some actual data.

Also for each slide, you should ask yourself just from visual inspection if you can see a plateau in this data, so leveling off or if you cannot see leveling off in this data and you would rather perform a verification test in order to see if the VO2 could potentially increase of higher workloads.
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In 2023 there was a very nice publication by an alternate colleague, so this is the Cologne Group that worked on this, that actually focused on this specific question and the specific issue and they looked at different filters that were applied to same data sets, so they had really very, very large data set here and they looked at how different filtering techniques actually affect the VO2max that we get from the same data.

From top to bottom, we can see that different filters were applied, so a 750 in breadth moving average, then a digital filter was applied, we can see a 30 second moving in binned average and then lastly a 30 second binned average.

And we can see for the same data set here for 7 breadth moving average, we would get 65 ml per kilogram whereas for the 60 second binned average we roughly get this should be around 61.5 or 62 somewhere in this range, so we quite get some error for this with regards to different filtering techniques, but again just shows you also what I have showed you on the slide before and if you want to have some further information I recommend you to go and read this article because it's actually published in sports medicine, so very high-impact journal.

And so if you want to have some further information regarding this, especially how you can apply the filtering techniques, maybe read into what a digital filter actually is, I highly recommend this publication here from this group.
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Let's take a couple of slides to focus on indirect colorometry, meaning how can we calculate the substrate utilization, the data that we have gathered with the spirometric device.

For this we have to look at the metabolic pathways again and how carbohydrates and how fats are differently metabolized in the body.

For glucose and glycogen we can see that we need six parts of oxygen which convert to six parts of carbohydrates, meaning that the ratio between oxygen that is used and carbon dioxide that is produced is one.

So therefore if 100 percent carbohydrate utilization were to measure we would need to measure as much oxygen consumption as carbon dioxide production.

For fats however the story is a little bit different meaning that we need 23 parts of oxygen but on the other hand we only produce 16 parts of carbon dioxide meaning the respiratory quotient is roughly 0.7.

It depends a little bit on what kind of fatty acid you use for this year the example is pymitic acid but so you also have stereate which is a different fatty acid and the respiratory quotient differ slightly but they are all between 0.96 and 0.2 at 0.7 to my knowledge.

So what does it mean now if we were to measure a respiratory quotient of 0.85?

So 0.85 would mean that we would have 50 percent of carbohydrate utilization and 50 percent of fatty acid utilization.

So we have this one which comes from the respiratory quotient from glucose times 0.5 because we are 50 percent here plus 0.7 which is the respiratory quotient for the fats here also multiplied by 0.5 because we have 50 percent fatty acid oxidation and this is the makeup of a respiratory quotient of 0.85.

Unfortunately the story is not as simple as I've put it here as we can see on the next slide.
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The problem is that this relationship between the respiratory quotient and here the carbohydrate oxidation, as linear as we can see it here, is true in theory but the problem is that we do not measure the O2 consumption and the carbon dioxide production, especially the carbon dioxide production, at the level of the muscle.

If we were to do this perfectly at the level of the muscle then this relationship would persist and the linear relationship between respiratory quotient and carbohydrate oxidation can also be used in practice but the problem is that we are not measuring at the level of the muscle but we are actually measuring the gas exchange at the level of the mouth.

So therefore the kinetics that we are measuring here are representative for the whole organisms so for the entire human being and not just for the muscle and therefore we need to keep in mind that there are other processes that might produce CO2 that obscure the relationship between respiratory quotient and carbohydrate oxidation.
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The most important process that comes to mind here is the buffering of hydrogen ions.

Especially when lactate is produced, hydrogen ions accumulate and are brought together with the lactate out of the muscle cell and into the blood.

But the blood has a very narrow range in pH in which it is kept and this pH range is stabilized by the so-called bicarbonate buffering system.

Meaning that when H plus ions or hydrogen ions come from the muscle cell into the blood, they react together to form on the right hand side here CO2 and H2O.

This CO_2 is then brought back to the lungs where it diffuses and then through the breathing leaves the body.

Therefore, we have the production of CO_2 that is not directly related to the oxidative metabolism of carbohydrates and fats.

And this then obscurates the relationship between VO_2 and VCO_2, so the IER and the substrate utilization.

Therefore, we need to correct for this process with some kind of equations.
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There's a highly cited publication from 2004 by Jorgen Drup and Wallace, so Ask Jorgen Drup, the guy from the Netherlands who has lots of publications and lots of papers about nutrition, especially during exercise.

They calculated or they came up with these equations that you can potentially use to calculate the carbohydrate fat oxidation as a function of the VO_2 and the VCO_2 that you have measured.

Because of this process that you have more accumulation of lactate for higher intensities, they have come up with these two kinds of equations that can be used for low intensity exercise, so roughly 40 to 50 percent of VO_2 marks, which are the upper equations that we can see here, and on the bottom the proposed equations for moderate or high intensity, which roughly are valid for the ranges of 50 to 75 percent of VO_2 marks.

I will upload a file for you on Moodle from a step test, some graded exercise test, and your job will be to first filter the data, and then secondly, use the equations that we have here to calculate the carbohydrate oxidation in gram per minute, as well as the fat oxidation in gram per minute or gram per hour, whatever unit you want to choose.

Also, you should prepare to couple of slides, or however you want to present it, and be ready to present the findings that you have from such a test.
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