Reliability of common exercise performance measures

PERFORM

Final report minor thesis

Supervisors:

Marco Mensink

Report writer:

Francesco Di Martino Comaschi

900429181080

Course code:

HNE 85324

WAGENINGEN UR
For quality of life


WAGENINGEN UR
For quality of life


WAGENINGEN UR
For quality of life

table of content

List of abbreviation

1 MR	One Repetition Max
BMI	Body Mass Index
CV	Coefficient of variation
O_2	Oxygen
PCr	Phosphocreatine
rpm	Pedal revolutions per minute
VO_2 max	Maximal oxygen consumption
Wmax	Maximal exercise capacity

1. Introduction

Many diet strategies and nutritional-supplements claim to increase physical performances of athletes (Saris,2003). Moreover, the use of supplements and diet strategies is continually rising and developing, also among non-professional athletes. However, measuring exercise performance is difficult, but needed to clarify if a particular supplement, or diet, increases effectively physical performances. Different aspects of physical performances are recreated in laboratory, to test the eventually beneficial effects of these products. Force/power, aerobic or anaerobic capacity are the most studied aspects of physical performance. All these aspects use different energy sources which depends on the duration and intensity of physical performance; these energy sources are Phosphocreatine (PCr) breakdown, anaerobic glycolysis (lactic acid system) and oxidation of carbohydrates and fats (aerobic system) (figure1).

1.1. Aspects of physical performances

1.1.1. Strength

For strength, it is considered the torque production in a mono-articular movement from the activation of a single group of muscles (arm flexion, knee extension) or several muscle groups in a whole body exercise task (e.g. weight lifting, sprinting, jumping). The energy source of this kind of exercise is the PCr breakdown, which is present in muscle fibres and its breakdown in phosphor and creatine release a great amount of free energy in a rapid time but for a limited duration (3-4 seconds) (Saris,2003, Spriet,1995). Example of athletes that use this kind of energy source are weightlifters (figure1) .

1.1.2. Anaerobic capacity

Anaerobic capacity refers to the total work output (Joule) for a period of time between a couple of seconds to two minutes. The main energy sources is anaerobic glycolysis of glucose with lactate as final product. Anaerobic glycolysis releases energy at high intensity but for a short time as for PCr.in general we use this source of energy when the exercise continues from 5 to 10 seconds (Saris,2003; Spriet,1995).The most relevant example of athletes, who use anaerobic glycolysis as main energy source, are the 100 meter sprinters (figure1).

1.1.3. Aerobic capacity

Aerobic capacity, or endurance, is typically defined as resistance to fatigue during prolonged exercise. Typically endurance exercise is therefore 30 minutes or longer. The main energy source is the oxidation of carbohydrates and fats which need oxygen (O_2) (Saris,2003). Aerobic metabolism releases energy slower and in a less intensive way, but for a longer period of time compared to PCr and anaerobic glycolysis system. The aerobic system became the main energy source for prolonged and submaximal exercise (more than 30 minutes, figure 1);the athletes, who aerobic energy system are cyclists and long distance runners (figure1).

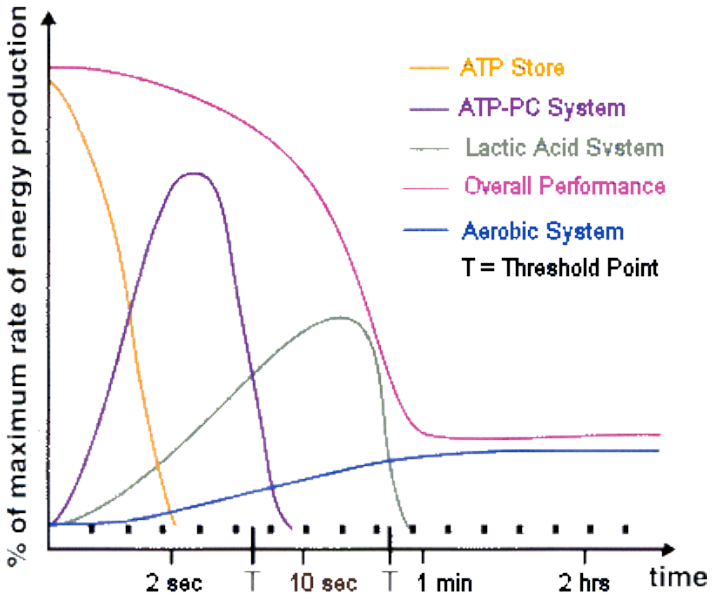


Figure1: Energy metabolism related to the timing and the intensity. For high intensity and short duration(3-4 seconds) exercises (as weight lifting) the main energy source is the PCr breakdown. When duration of the exercise is prolonged from few second to two minutes the anaerobic system is the most used. While for prolonged submaximal exercises (more than 30 minutes) the main energy sources is the oxidation of glucose or fats.Figure from www.healthylivingheavylifting.com)

1.2. Physical performances measures

Three important factors, to consider during the testing of physical performance, are the validity, reliability and sensibility (Hopkins,2001; Currell,2008).

1.2.1. Validity

is the similarity of the test with the real sport; in other words a valid trial is when the excises, asked to perform, reflects the field situations (Currell,2008). To study the effect of a supplement or of a diet on physical performance is important having a valid test because the eventual beneficial effect of a treatment (as a supplement ingestion or a particular diet) can be seen in the real sport.

1.2.2. Reliability

(or reproducibility) is considered as the variation of the measurements of the same subject at the same experimental conditions, but at different occasions. In other words, it detects differences in the outcomes of an experiment done multiple times within the same subject without changing any variables. Generally, reliability is indicated through the coefficient of variation (CV) (figure 2) which is an equivalent of the standard error expressed in percentage. A low CV means and high reliability that is the desirable condition of any experiment (Hopkins,2001; Currell,2008).

$$\text{Coefficient of Variation} = \frac{\sigma}{\mu} \text{ x } 100$$

Figure2:Coefficent of Variation(CV) formula.

1.2.3. Sensibility

Sensibility is the ability to detect the eventually outcome changes attributable to a particular treatment (in this case a diet strategies or the use of a supplements). A sensible test is able to identify little differences during the test. For the studies with elite athletes, where the performances change in a small rate, it is very important to have sensible test otherwise small improvements, due to a treatment, cannot be detected (Currell,2008).

2. Aim

As explained previously, exercise performances (as strength or endurance) can be difficult to measure because there are different factors which may influence these measures. Many of these factors can be controlled in laboratory settings but still some of them can increase the variability of physical tests (not only between, but also within individuals). Moreover, other factors as the study population and the equipment of the laboratory have an important role in physical performance tests.

The aims of my master thesis (minor) are:

Individuate all the factors that may influence the reliability of physical test (see section 3) and write a list of instruction to control these factors (appendix I).

Prepare the study design to evaluate the reliability of two aerobic capacity tests (10km time trial and 15 minutes trial) and one strength test (one repetition max). Moreover see if these test are suitable for trained and untrained subjects (see section 4).

Calibrate the ergometer (used for the aerobic capacity tests) to the proper linear mode for cyclist and untrained athletes (see section 5).

Each aim of the project has dedicate a section in the report. In section 3 all factors, which may influence the reliability of physical test, are explained, in section 4 the study design is described and in section 5 is about the linear mode, of the ergometer, chosen for the study. In section 6 some suggestions for future researches are present.

3. Factors influence reliability

Exercise performances measures, as strength anaerobic and aerobic capacity, must be reliable and sensible. However, several factors influence the reliability and the sensitivity of these tests. Principally the main factors that influence the reliability are the type of test chosen, participant selections, test execution and diet of participants.

3.1. Type of test

Physical performances can be analysed by several type of tests; the choice of which kind of test to use depends by which aspect of performance you want to study. The most studied aspect of physical tests are strength are aerobic and anaerobic capacity.

3.1.1. Strength test

Strength can be measured principally with iso-inertial test or isokinetic test; the first test analyses the explosive power of a single movement as a jump, throwing or lift a weigh(Saris,2003). While in the isokinetic test, it is measured the peak power or the mean power of a repetition series of a single movement as arm flexion, or knee extension (Saris,2003). For this kind of trial an ergometer is needed to block the rest of the body, allowing the movement of the part interested.

Both of the tests have positive and negative factors. For example the article review of Hopkins suggests isokinetic test has an higher coefficient of variation (CV) which means a lower reliability (Hopkins,2001). However, the study of Emery said, CV can be lowered if the participants does not move from the ergometer between the trials (Emery, 1999). Iso-inertial test seems to be more reliable but their results may be influenced by the learning skills of the subjects. in other words the performances may not be determined only by the treatment, but also by other factors as co-ordination or training status that may reduce the impact of the treatment and act as confounders.

3.1.2. Aerobic and anaerobic capacity

Aerobic and anaerobic performance can be measured trough different tests that differ for their methodology. Each test has some strengths and some weaknesses. In base of the goal of the researcher one test is more or less suitable for the experiments (figure 3).

Constant-work test

It is also known as time trial, where the subjects must perform a fixed work output or tread a set distance as fast as possible (Hopkins,2001). The main outcome, of this kind of test, is the time taken to finish the work out. The methods used to complete the set workload or distance are cycling, running (in the field or treadmill) or rowing. This kind of test has many advantages: as their validity (reflects the reality) and their reliability considered quite high (Saris,2003;Currel,2008). However, metabolic measurements (as blood samples) are not possible during the trial because they may influence the performance (Currell,2008). Thus for the experiments which need these kind of measures time trial is not a suitable test. Moreover another weakness is that exist many versions of time trial and sometime could be difficult the comparison of different results (Saris,2003; Jeukendrup,2004).

Constant-duration tests

Constant-duration test is the opposite of time trial, in this case subjects must complete the maximum of the work, or tread the maximal distance they can, in asset of time (Hopkins,2001). Constant-duration test presents the same strong points and weaknesses of

time trial, as high reliability and validity but the impossibility of metabolic measurements during the trial (Saris,2003). A version of constant-duration tests is the submaximal test where for a set time subject must do a work at a lower intensity; outcomes as maximal oxygen consumption (VO_2max) are estimated trough some formulas. These test are valid, reliable and metabolic measures during the trial are possible; Moreover submaximal test seem to be suitable for less trained people (Noonan,2000). On the other hand, protocols about this kind of exercise still are not totally developed; furthermore there are doubts about their sensibility which seems to be lower than maximal exercises (Noonan,2000).

Exhaustion test

In the exhaustion trial, subjects must maintain work output until they are able (end of the test). Two version of this experiment exist: constant-power and incremental tests, in the first case the work output is kept constant while in the second case intensity of the exercise gradually increases (Hopkins,2001). There are doubts about the validity and the reliability of these tests; indeed exhaustion trials does not represent the real sport (Saris,2003). Moreover the study of Jeukendrup demonstrated constant-power test has a much higher CV compared with time trial and constant-duration (Jeukendrup,1996). However incremental tests seem to have an higher reliability compared to the constant-power (Hopkins,2001). On the other hand, studies suggest exhaustion trials have an higher sensibility and metabolic measurements are possible (Saris,2003). Incremental tests are often used in the pre-test to estimate the maximal workload (Wmax) and VO_2max of athletes (Jeukendrup,2004;Laursen,2002).

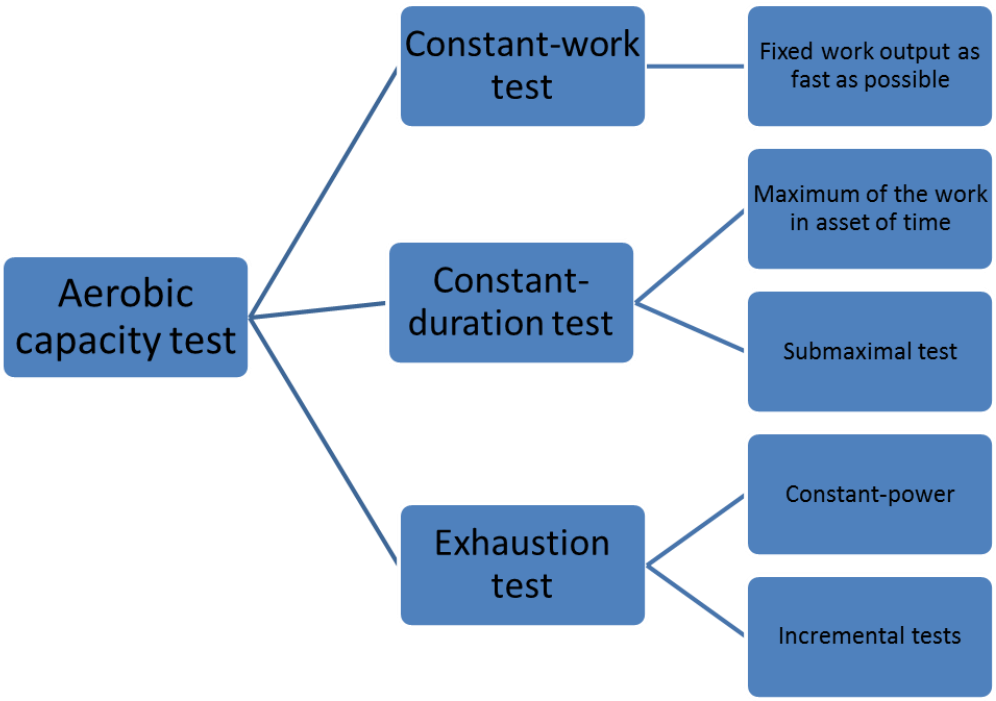


Figure 3 Aerobic capacity tests: *They are divided in Constant-work test, Constant-duration test and exhaustion test.*

3.2. Participants selection

Selection criteria of the participants may significant influence the reliability of the tests (Hoffman,2006); for this reason different variables must be taken in account.

3.2.1. Athletic Status

The review of Hopkins, suggests well trained subjects are the most suitable subjects for studying the effect of supplements, or diet strategy, on physical performances (Hopkins,2001). The main reason is due the low variation (low CV) of the performances of trained athletes. Other two factors to consider are the motivations and familiarization which are higher for athletes compared to non-athletes. On the other hand, the use of professional athletes in some studies is not always advised because some time the physical performances can be influenced by technical skills which could make harder to detect the effect of particular treatment (Saris,2003). For example if in a study are used professional athletes where the results of their performances change minimally, the effect of treatment (as the administration of a supplement or a diet) will be very small and eventually difficult to detect (Hopkins,2001).

3.2.2. Gender

No significant differences in reliability are found between male and female athletes, but these differences between the genders are more significant in non-athletes (Hopkins,2001). Untrained females have a lower reliability compared to untrained males. Moreover, the effect of menstrual cycle on physical performances seems to affect more non-athletes than professional athletes which seems to be less significant (Beidleman,1999; Lebrun,1993).

3.3. Test execution

During the execution of the trails there are many factors to consider which could influence the reliability and the sensitivity.

3.3.1. Mode of execution

The mode of execution refers principally to the aerobic and anaerobic capacity tests. Generally trials are carried out running, cycling, swimming and rowing; no significant differences in reliability and sensibility are found between the modes of exercise (Hopkins,2001; Currel 2008). Also no significant differences are found between test in the field and in the laboratory (treadmill or ergometer) about the validity and reliability (Nummela,2007). On the other hand, for “general” athletes the best execution mode is cycling and running; swimming and rowing mode is only for few class of athletes and could be influenced more by technical skills.

3.3.2. Familiarization

During the trial measures, often subjects must work in unusual situations (as ergometer with VO_2max mask) lowering the performance of tests in particular for not professional athletes and untrained people (Laursen, 2002). However, if the trial is repeated more time the subjects are used to laboratory situations and in the last trials an increase of physical performances may be present (due to the adaptation) reducing the reliability of the test. For this reason, before the starting of the official test, subjects should have at least one “familiarization” trial where they can practice and the measures must not be taken in account for the study.

3.3.3. Duration of the test

The duration of a protocol seems to be correlate with the CV of the results, in other words longer is the trial higher is the CV (Currel, 2008). In particular exhaustion test seems to be the most affected by this factor and in part it could explain why these kind of test have a lower reliability. Generally, constant power test has a higher CV compared to the incremental power due to a longer duration (generally more than one hour). For this reason, Billat suggests to choose the incremental power test (Billat,1994). In particular, if the subjects are non-athletes the duration of the test may influence even more the CV of the test (Hopkins,2001) The reliability of time trial and constant-duration tests seems to be less affected by the duration of the exercise (Currell,2008).

3.3.4. Inter-trial time

The inter-trial time is the interval period between two tests of the same subject, and this period may influence the reliability (Hopkins,2001). If the trial is repeated more than two times this interval period should be keep constant. About the length of the inter-trial time is still not clear because there are no specific studies about that. However the review of Hopkins suggests a really short period (as a couple of days), between two trials, may increase the CV, due to fatigue effects of the previous test (Hopkins,2001). On the other hand, prolonged periods (two weeks or more) between the trials may also influence negatively the reliability because in that period it may happen injuries, illness, loss of motivations or changes in the training status. Even specific studies are not published, an inter-trial period of about one week seems to be the proper one (Hopkins,2001).

3.3.5. Training

Another factor that could influence the reliability is the training during the period of the study in particular the day before the trial. In many researches subjects are asked to maintain a similar training routine or even trainings are scheduled for the duration of all the study (Hoffman,2006, Angus,2000) Also, strenuous physical exercise, or competitions, the days before the trial are not allowed due to possible fatigue effects which may impair the performances (Hoffman,2006; Angus,2000). The off-season, without competitions or specific trainings, is the best timing for the trials.

3.3.6. Verbal encouragement

Verbal encouragements are often used in physical test to help the participants. This situation could respect the reality because during a race or a soccer matches athletes are subjected to encouragements by supporters, even in non-professional competition. On the other hand, verbal encouragement could impair the reliability of the test and represent a possible confounder (Currell,2008). Currell, supported by the study of Gibson, suggests verbal motivations positively influence physical performances of repeated maximal force exercise (Currell,2008; Gibson,2004). Moreover, external encouragements may change between the trials or the reaction of the subjects may differs, increasing the CV of the test (Currell,2008). However, in the study of Hulleman, with cyclists which performed a time trial, did not found any effects of external encouragements in performances (Hulleman,2007). Verbal encouragements should be standardized or reduced at the minimum to avoid any risk.

3.3.7. Music

Also music is often used during physical tests and they may present the same problems of verbal encouragement being a possible confounder and impairing the reliability of the test (Currell,2008). In a study where cyclists performed a 10-km time trial with or without high

tempo dance music (trance), it was found when the subjects listened music the speed significant increased (Atkinson,2004). Reliability may be negatively be influenced by music because the effects could be determined by the preferences of the subjects which is personal. On the other hand, the study of Hagen suggests to use a self-selected music playlist which should not influence the physical performance of athletes (Hagen,2013).

However, trials with music may have a lower validity because the most of the sport competitions are without music.

3.3.8. Feedback

Another factor, which may influence performances is giving feedbacks to the subjects (as distance tread, work load done or timing) after the completion of a trials (Nikolopouios,2001). In particular the reliability may be negatively affected; normally, if subjects know their results during or after the trails, in the next one they would try to beat the previous and consequently the CV will be much higher. Studies demonstrated the perception of the events, thus the Rate of Perceived Exertion (RPE), seems to be the most changed when athletes received a feedback during the trials (Nikolopouios,2001; Nethery,2002).

3.3.9. Measurements

Physiological measurements as blood sampling may interfere with physical performance and reduce the validity of the test (Currell,2008). For this reason, in many types of test (as time trials and constant-duration tests) these kind of measurements are not possible. Exhaustion test and submaximal trials are the most suitable for physiological measurements. For this reason physiological measurements should be done only when they are necessary. The only situation, where the these measurements seem to interfere less, are during submaximal tests (Noonan,2000).

3.4. Diet

As explained previously, nutritional strategies or the use of supplements play an important role in sport competitions (Jeukendrup, 2004). For this reason the diets (as high-carbohydrates) and supplements effects (as creatine administration), are studied in laboratory. However, some nutritional habits of the subjects could influence the reliability of the tests or be a possible confounder.

Between the trials, subject should do not vary their diet otherwise they could increase the CV of the outcomes. Also, studies suggest to standardized the meals before the test (at least 24h before) (Angus,2000; Coyle,1983). In the study of Angus, about the effect of carbohydrates beverages ingestion during time-trial performances, the participant were supplied with a food parcel with a standard energy and substrate composition 24h before the trial. In the study of Coyle the diet was keep in constantan composition and in the amount of energy in a 48h period. High reliability found these two studies demonstrate have a standardized diet before the trials it's very important. Moreover, the studies of Angus and Coyle suggest the food intake prior to the test must be keep quantitatively and qualitatively constant; in other words subjects should consume the same amount of energy(kJ or Kcal) and with a similar macronutrient composition (carbohydrates, fats and proteins) (Angus,2000; Coyle,1983).

The consume of tobacco, alcohol and caffeine, before the trial, should not be allowed because they could influence the performances (Angus,2000). A standard diet, before the test, for all projects is not possible, but it will depend by the treatment that the subjects will

received during the study. Before decide which foods are allowed and not, eventually interferences or overlap with the treatment must be taken in account. For example in the study of Bel, about the effects of caffeine pills on anaerobic capacity, the subjects was asked to do not drink coffee or beverage with caffeine the days before the test (Bel,2001).

Moreover, subjects with pathology linked to foods or particular diet restrictions must be considered in the participant selection of the study. Two examples are the studies of Becque and Vandenberghe, where the effect of creatine supplementation was studied (Becquw,2000; Vandenberghe,1997). In these studies vegetarian athletes were not included because the effect of creatine supplementation on vegetarians significant differs from non-vegetarians (Burke,2003). Thus, in the exclusion criteria it is important to take in account if eventually food intolerances or particular diet restrictions may influence the treatment.

3.5. Conclusion

All the factors mentioned (table1) may be controlled in laboratory but other ones which are intrinsic to the subjects cannot be controlled in laboratory and may cause an increase the reliability. In appendix I is present a list of instructions to reduce, as possible, the variability of physical performance measures.

Table1: factors which influence the reliability of physical test. Instructions to control these factors are present in Appendix I

Type of test	Strength
	Anaerobic capacity
	Aerobic capacity
Participants selection	Athletic status
	Gender
Test execution	Mode of execution
	Familiarization
	Duration of the test
	Inter-trial time
	Training
	Verbal encouragement
	Music
	Feedback
	Measurements
Diet	Variation during the study
	24 hours before the trials
	Alcohol, tobacco and caffeine
	Influences with the treatments
	Food intolerances or diet restrictions

4. Study design

The aim of the research is to establish the reliability of different exercise performance tests. Strength/power and aerobic capacity are the performance aspects chosen in this study. The aerobic tests will be repeated three times by the same individual at three different occasions, with control of external factors, while strength tests will be repeated four times at the same conditions. For the strength measurements, one-repetition-maximum (1RM) will be considered, while for the anaerobic capacity a 10 km time trial and 15 minutes constant-duration tests will be chosen (Table 2).

Table2: Physical performances and tests.

Strength/Power	One-repetition-maximum
Aerobic capacity	Time trial
	Constant-duration tests

4.1. Aerobic capacity study design

The primary object of the study is to establish the reliability of two aerobic test: 10km time trial and 15 minutes constant duration test and see eventually difference in Cv. For each trial six cyclists (with 4-10 hours of training per week) will be chosen. Moreover, to establish if these kind of protocols are suitable for non-athletes, another group, composed by six subjects with a weekly routine lower than two hours, will perform a 10km Time trial and their reliability will be compared with the cyclists, who perform the same trial (Figure 4).

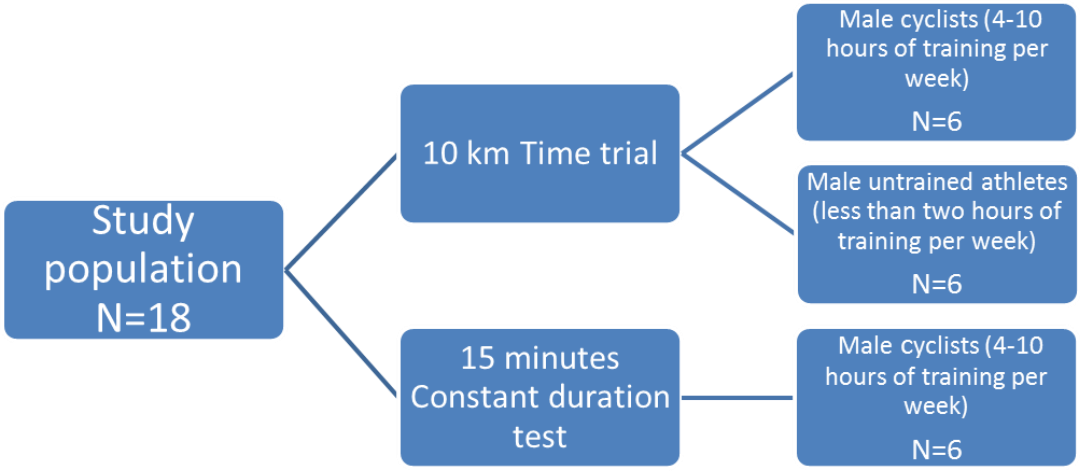


Figure 4: Study design of aerobic capacity tests. Three equal groups composed by six subjects will be formed. In two of them the participants will perform 10km time trial where one group will be composed by competitive cyclist while the second one will be formed by sedentary subjects. The third group will be composed competitive cyclist, which will perform 15 minutes contestant duration test.

4.2. Study design of strength test

The aim of the research is to study the reliability of 1RM test; more over if factors as training status may influence the reproducibly of these tests. First of all, two equal groups will be formed: one composed by six weight lifters (training routine composed by at least four times per week) and the second by six non-athletes (less than two hours of training per week) (Figure 5).

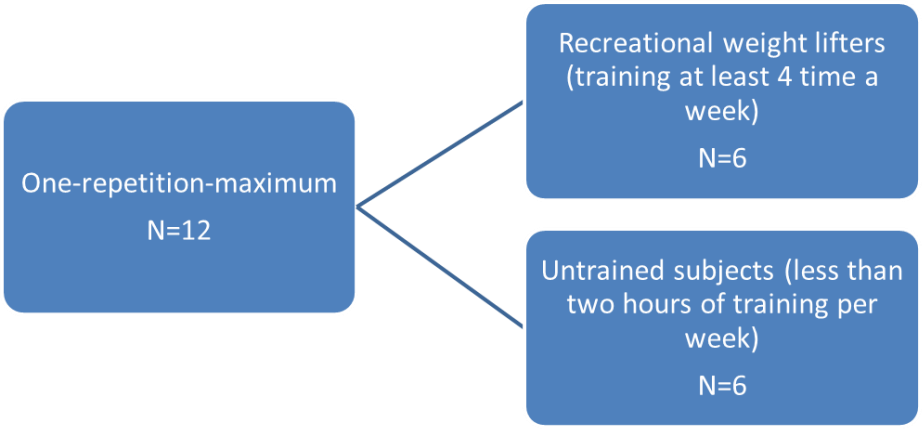


Figure 5: Study design strength tests. Six weight lifters and six recreational athletes will perform 1RM. Eventually differences, in reliability between groups will be take in account.

4.3. Time-frame

After an information meeting and a screening, subject will participate in pre-trail measures, including, height, weight, body composition and, for endurance athletes, VO_2max. After a learning (or familiarization) trial, for the aerobic each test will be repeated three times with an interval of a week between trials (Figure 6). For strength measures, instead of VO_2 test a resistance submaximal trial will be performed in the first week; subsequently 1MR test will be repeated four times, always with an interval of a week between trials (Figure 7). For each subject, the trial will be repeated at the same hour and day of the week.

Figure 6: Time frame aerobic tests. In week one all the measures will be made (body composition and WO2max) while in week two subjects will familiar with tests through a learning trial where it will be explained the correct procedure of the test. The outcomes of the learning trial will be not used in the statistical analysis. Subsequently the learning trials each test will be repeated three times with an interval of one week between the trials.

Figure 7: Time frame strength test. In week one all the pre-trail measures, including, height, weight, body composition and submaximal test will be performed at the same day. Subsequently the submaximal trials each test will be repeated four times with an interval of one week between the trials.

4.4. STUDY POPULATION

4.4.1. Population

Thirty volunteers, from 18 to 35 years old, will be recruited from Wageningen University. As previously explained two types of study design are present: one about aerobic capacity tests and one about strength tests. For the aerobic capacity test, the study population will be composed by 12 male cyclists (from four to ten hours of training) and six healthy male subjects with a training routine not higher than two hours per week. For the strength tests six weight lifters (training four times per week) and six healthy male subjects (with a training routine not higher than two hours per week) will be recruited.

4.4.2. Inclusion criteria

In order to be eligible to participate in this study, a subject must meet all of the following criteria:

- Generally healthy
- Age between 18 and 35 years old
- Male
- not use any drugs or medicine that may interfere with exercise capacity, e.g. all substances on the international doping list.
- fit in one of the following groups if:
    - Are a cyclist and you have performed endurance-like exercise or training on a regular basis of 4-10 hours per week in the past year.
    - Are a recreational weight lifter and you have performed resistance-like exercise or training on a regular basis with a minimum of 3 sessions per week in the past year.
    - Are not a cyclist and you have exercised a maximum of 1 times a week average in the past year and you have a BMI < 28.
    - Are not a weight lifter and you have exercised a maximum of 1 times a week average in the past year BMI < 28.

4.4.3. Exclusion criteria

A potential subject who meets any of the following criteria will be excluded from participation in this study:

- Subjects with injuries or active diseases which may impair performance
- Participant in sport competitions between the trials.
- Not able or willing to comply with the study procedures

4.5. Study parameters/endpoints

4.5.1. Main study parameter/endpoint

The objective of the research is to study the reproducibility of different exercise performance tests. Thus, the main parameter will be the coefficient of variation (CV) of each test.
For the 1 RM the main outcome is the variation of absolute (Kg) and relative weight (Kg/Kg BW) lifted.
For the endurances test, the total work performed (kJ) and the time (s) to complete the target amount of work will be measured respectively for 15 minutes constant the duration test and the 10km time trial.

4.5.2. Other study parameters

Other parameters will be considered before the starting of the test are height (cm), weight (Kg), body composition (%) and, for endurance athletes only, aerobic capacity (VO2max, L/min).

4.6. Study procedures

4.6.1. Information meeting

About 2 weeks prior to the beginning of the study, an information meeting will be organized by the research team to inform persons that are interested in taking part in this study. During this meeting also appointments can be made for screening and preliminary testing. Participants will also receive an informed consent form, which they can either take home to sign if they decide to join the study or, if they had made up their mind already, can sign immediately (appendix II).

4.6.2. Screening

Once the informed consent is signed, the following data will be collected:

- Anthropometric variables for BMI calculation: body weight and height
- A general screening questionnaire (appendix III)

This is a screening on paper and will be performed prior to the pre-trial measurements in order to recruit subjects who fulfil all criteria.

4.6.3. Pre-trial measurements

One week prior to the first experimental period subjects are invited for preliminary testing. At this point several anthropometric variables will be measured:

- Body weight
- Length
- Body composition: Skin fold

Maximal aerobic capacity test (W_{max} and $VO2_{max}$) – Endurance athletes only
Next to this, maximal aerobic capacity testing will be performed on an electronically braked cycle ergometer (Lode Excalibur) and by using indirect calorimetry (Oxycon Pro).
At this first visit participants will perform maximal aerobic capacity test (Wmax and VO2max) to determine the aerobic capacity. After a short warming up of five minutes, subjects will start with cycling at 150W or 100W at the preferred pedal frequency, indicated as rotations per minute (RPM). Every minute the power will increase with 10W. Participants will have to cycle until he is no longer able to maintain the workload. Oxygen consumption is measured throughout the exercise test, breath by breath, using indirect calorimetry (appendix IV). Peak oxygen consumption (VO_{2max}) is calculated as the highest average VO_2 during any 30 seconds in any given period during exercise. Subjects are allowed to drink water ad libitum. The test will be considered valid when at least two out of three criteria are met: levelling of VO_2 with increasing workload; heart rate within 10 beats of the predicted maximum (220-age) and a respiratory exchange ratio (RER) of >1.05. When the test is valid, the participant can be included in the study.

Submaximal test - Resistance group only
The subjects will start with a warming up of five minutes on a cycle ergometer. Thereafter, an appropriate weight will be chosen (which the subject can lift for a maximum 10 times) will be chosen. The subject will perform as many repetitions as possible.
The predicted one repetition maximum will be calculated using the following formula: 1-RM=load (Mayhew,1995). This estimated weight will be used to determine the initial load for the 1RM tests in the following visits

4.6.4. Dietary record diary

Participants will have to fill a diary (appendix V) of the food, beverage intake and eventually disease symptoms in the day of the test and the previous one (appendix X). After the first test subjects will have a copy of the diary, whose will help them to keep constant the food and beverage intake in the following trials.

4.7. Exercise performance tests

4.7.1. 15 minutes Constant-duration test

The test will be started with a warm up of five minutes at 60% Wmax. Thereafter a 15-min time trial will follow in which subjects have to perform as much work as possible. During the warm-up the electromagnetically braked ergometer (Lode Excalibur) is in the hyperbolic mode, so that the work rate is independent of pedalling rate. During the 15-min time trial the ergometer will be in the linear mode so that with increasing pedalling rate the work rate increases, according to the formula.

W = L x rpm2;

where W = workload, L = linear factor, rpm = pedal revolutions per minute.

The linear mode for the subjects will be set at the preferred rpm (about 80/85 rpm) at 85% Wmax. In this way the subject can pace himself and try to maintain a high pedalling rate over 15 minutes to maximize power output.

4.7.2. 10 km Time trial

To make this trial comparable with the constant duration test, the distance that the subjects have to complete as fast as possible should have similar timing of 15 minutes constant duration test.
After a warm-up period of 5 minutes at 60% Wmax, subjects will start the trial and they will bike as quickly they can on the ergometer. The software linked to the ergometer can estimate the distance done in base of the linear mode chosen. When the subjects will reach 10 km the test will finish. The linear mode will be the same used in the 15 minutes Constant-duration test. A display will inform the subject about the distance made and the cadence The time taken to complete the test will be measured (Jeukendrup,1996; Pottier,2010).

4.7.3. One-repetition maximum (1-RM)

The subjects will start with a warming up of five minutes on a cycle ergometer. After the warming up the subject will perform eight repetitions at 50% of the estimated weight on the leg press. After the warm up set the subjects will rest for one minute. Thereafter, the subjects will perform 3 repetitions at 75% of the estimated weight. This will be followed by a series of attempts with a gradual increase of the of weight until maximum is reached. The subjects will rest for 3 minutes between the attempts. After a failed lift, one more attempt is made with a load calculated as the sum of the heaviest load lifted plus the half of the difference between

that load and the load of the failed lift. The 1RM is attained in 3-5 attempts to circumvent exhaustion. (Hoffman, 2006; Becque 2000).

4.8. STATISTICAL ANALYSIS

Physical test will be repeated three or four times and all the parameters, for each trial, will be assessed trough ANOVA for repeated measures. The parameters will be studied are peak (kJ) and relative peak power(kJ/Kg), absolute (Kg) and relative weight lifted (Kg/KgBW), time to complete the target amount of work (s) or completed work load (J). The individual coefficients of variation (CV) of each trial will be measured. The CV of the members of each group will be averaged. CV differences between the trials will be analysed through Mann-Whitney test and comparison will be made through unpaired t-test.

5. Linear mode

5.1. Introduction

To measure the workload or distance travelled in the aerobic capacity tests, the ergometer must be set in liner mode so the work rate can be measured in base of the pedalling rate of the subjects. Generally linear mode is indicated with the following formula:

$$P\ [W] = \alpha^* (n\ [min\text{-}1])^2 * (s)$$

The P [W] is the workload during the test, α is the liner factor of the ergometer during the test, n [min-1] is the rpm recorded during the test and (s) is the duration of the test (in second). The rpm depends on the performance of the subject; while the liner factor and the duration of the test are fixed values (in case of constant duration test). The liner factor it may be considered as the resistance of the ergometer (same principle of the gear bike) thus higher is α, higher is the workload at constant rpm. However an high linear mode implies an higher effort for the participant to maintain the same rpm. The linear factor is indicated according with the following formula:

$$\alpha = \%Wmax / (n\ [min\text{-}1])^2$$

Where %Wmax is the rate of the Maximal exercise capacity of a subject performing the test, and n [min-1] is the rpm which subjects have to maintain to work at the established Wmax%. An example of liner mode used in a previous study is : “linear mode set at 90 RPM at output 70% Wmax” (Jeukendrup,1996) .It means the linear factor of the ergometer is set in a way when the subject is pedalling at 90 RPM his output is 70% of his Wmax. Generally, the linear mode is the same for all the subjects who participate to the same study while the linear factor (α) changes for each subject because depend on individual Wmax, which is previously estimated.

5.2. Set the liner mode

As already mentioned, before the starting a time trial or a constant duration test, it is necessary to set the proper liner mode of the ergometer which is the most comfortable for the participants to complete the exercise. The duration of the tests and the intensity of the exercise influence the proper linear mode to use. If the linear mode of the ergometer is too high the test could lose validity and reliability because the trial, for the participants, may result too difficult to perform. Even a low linear mode is not advised because it could underestimate the outcome of the trial.

For example, in studies where competitive cyclists performed one-hour time trial the liner mode of the ergometer was set at 90 RPM at output 70% Wmax (Jeukendrup,1996; Desbrow,2004; Ivy,2009; Pottier,2010). In another study where 10km (about 20 minutes of trial) time trial were performed still by cyclist and the linear mode chosen was 85±7 rpm at 85% Wmax of the subjects) (Nyakayiru,2016, in press publication).

In the studies mentioned the participants were cyclist and but the same liner mode can be used for less trained people. Indeed, the linear factor, for less trained athletes, will be lower compared than cyclist because at the same liner mode (for example biking at the 70% of Wmax) the cyclists have an higher Wmax and consequently an higher α (according with the formula α = %Wmax / rpm2). The only doubt refers about some tests could not be suitable

for subjects with a low training status for example one hour time trials could be result too hard to perform for a sedentary person independently by his/her linear factor. The aim of this project was to develop a physical test protocol suitable also for less training people, thus for this reason short duration tests as 15 minute constant duration and 10km time trial (about 15/20 minutes of test) were chosen, instead of one hour time trial.

5.3. Linear mode tested

Before the start of the test measures (15 minutes constant duration test and 10km time trial) it is important to set the proper liner mode for the participants. To have a valid and reliable test is important the linear factor is not too high (subject could not complete the trial) or too low (the subject could not reach his maximal capacity). In this project cyclists and untrained subject will perform four time (one familiarization plus three measures) the 10km time trial.

According with the study of the study of Nyakayiru, where also10km time trial was performed by cyclists, the linear mode chosen was the 80%/85% of the subject Wmax at the most comfortable rpm which allows to finish the trial. This linear mode was test among three subjects with a different training status: a sedentary person, a recreational football player and a cyclist. All subject performed at least on 10km time trial and 15 minutes constant duration test and the aim of these tests was to check if the subjects were able to complete the trial at the linear mode at 80%/85% of their Wmax . Each subject has a preferred rpm due to his/her training status; for example the rpm of the cyclist was much higher of the untrained subject (table 3). Moreover, a 15 minutes test was performed a liner mode at 90% of Wmax (only with the cyclist), but the test resulted too hard to perform thus it was decided to continue with the linear mode at 80% of Wmax.

Table3: Wmax of the subjects and their favourite rpm.

subject	Wmax	rpm
untrained	200	65
football player	270	70/75
cyclist	370	85

5.4. Conclusion

All subjects could finish the tests without been exhausted; thus the liner mode chosen for the study will be 80% of the subject Wmax. The facts all subjects could finished the test without any problem is very important because a possible exhaustion of the subjects could decrease the reliability (Hopkins,2001; Saris,2003) of the trials or cause a drop out during the study (due to a loss of motivations).

6. Suggestion for future researches

Several factors may influence the validity and the reliability of performance tests but still not all of them are clear. In this project the two factor studied were the typology of test (constant duration test vs time trial) and training status of subject (trained athletes vs sedentary people); however there are other factors that influence reliability that could influence reliability.

As described in section 3 and in the article review of Hopkins, gender is a factor who may influence the reliability of the test, in particular in the case of sedentary subjects where untrained females are considered less reliable than untrained males (Hopkins,2001). On the other hand, this difference of gender seems less evident for athletes (Hopkins,2001). However specific studies who tested directly the differences in reliability between male and female athletes were not found. The most studies, where physical performance are measured, use only male athletes and sometime recruitment of participants may be not easy. If no difference in reliability are not found between the genders the recruitment of female athletes will be possible allowing an higher availability of participants for future researches.

In the present this project, only the reliability of One repetition max is studied, but it is not the only strength test available; there are also isokinetic tests which have some advantages and some disadvantages compared 1 MR (already mentioned in section 3). Develop a protocol even for isokinetic tests and see if it suitable for trained and untrained subject could be useful, for the department of Human Nutrition, as an alternative of 1 MR. Moreover a limitation of this study is the strength was measured only in the low part of the body (leg press test) and to have a whole measure of athletes resistance also the upper part of the body strength should be measured.

In the case of 15 minute constant duration test and 10km time trial do not result suitable for the untrained people, due to an high CV or for the difficulties of the subject to complete the trials, an alternative could be submaximal aerobic test. As already mentioned in section 3, in the submaximal tests subjects must perform an amount of work at a lower intensity (with fixed duration) compared to the trials of this project. Generally the outcomes of these kind of test, as maximal oxygen consumption (VO2max), are estimated trough some formulas (Noonan,2000). Moreover an advantage of this kind of test is measurements (as blood lactate or glycaemic index) are possible during the test. On the other hand, protocols about this kind of exercise still are not developed as for the maximal tests, thus future researches are needed.

One limitation of this project is that anaerobic tests were not taken in account. At beginning of this study also 30 second Wingate was included and it should have been performed before 1MR; however, the ergometer was already used for the aerobic tests, thus it was not possible to perform the 30 second Wingate. Several studies already demonstrated 30 second Wingate is a valid, reliable and diffused anaerobic test (Weinstein,1998; Sands,2004); thus other protocols for this kind of test could be developed maybe without the use of the ergometer as field tests or with treadmills.

7. References

Angus, D. J., Hargreaves, M., Dancey, J., & Febbraio, M. A. (2000). Effect of carbohydrate or carbohydrate plus medium-chain triglyceride ingestion on cycling time trial performance. Journal of Applied Physiology, 88(1), 113-119.

Atkinson, G., Wilson, D., & Eubank, M. (2004). Effects of music on work-rate distribution during a cycling time trial. International Journal of Sports Medicine, 25(08), 611-615.

Becque, M. D., Lochmann, J. D., & Melrose, D. R. (2000). Effects of oral creatine supplementation on muscular strength and body compositioin. Medicine and Science in Sports and Exercise, 32(3), 654-658.

Beidleman, B. A., Rock, P. B., Muza, S. R., Fulco, C. S., Forte, V. A., & Cymerman, A. (1999). Exercise V E and physical performance at altitude are not affected by menstrual cycle phase. Journal of Applied Physiology, 86(5), 1519-1526.

Bell, D. G., Jacobs, I. R. A., & Ellerington, K. R. I. S. T. I. N. A. (2001). Effect of caffeine and ephedrine ingestion on anaerobic exercise performance. Medicine and science in sports and exercise, 33(8), 1399-1403.

Billat, V., Renoux, J. C., Pinoteau, J, & Koralsztein, J. P. (1994). Reproducibility of running time to exhaustion at VO~ 2~ m~ a~ x in subelite runners. Medicine and science in sports and exercise, 26(2), 254-257

Burke, D. G., Chilibeck, P. D., Parise, G. I. A. N. N. I., Candow, D. G., Mahoney, D. O. U. G. L. A. S., & Tarnopolsky, M. (2003). Effect of creatine and weight training on muscle creatine and performance in vegetarians. Medicine and science in sports and exercise, 35(11), 1946-1955.

Coyle, E. F., Hagberg, J. M., Hurley, B. F., Martin, W. H., Ehsani, A. A., & Holloszy, J. O. (1983). Carbohydrate feeding during prolonged strenuous exercise can delay fatigue. Journal of Applied Physiology, 55(1), 230-235.

Currell, K., & Jeukendrup, A. E. (2008). Validity, reliability and sensitivity of measures of sporting performance. Sports medicine, 38(4), 297-316.

Desbrow, B., Anderson, S., Barrett, J., Rao, E., & Hargreave, M. (2004). Carbohydrate-electrolyte feedings and 1h time trial cycling performance. International journal of sport nutrition and exercise metabolism, 14, 541-549.

Emery, C. A., Maitland, M. E., & Meeuwisse, W. H. (1999). Test-Retest Reliability of Isokinetic Hip Adductor and Flexor Muscle Strength. Clinical Journal of Sport Medicine, 9(2), 79-85.

Gibson, A. S. C., & Noakes, T. D. (2004). Evidence for complex system integration and dynamic neural regulation of skeletal muscle recruitment during exercise in humans. British journal of sports medicine, 38(6), 797-806.

Hagen, J., Foster, C., Rodríguez-Marroyo, J., De Koning, J. J., Mikat, R. P., Hendrix, C. R., & Porcari, J. P. (2013). The effect of music on 10-km cycle time-trial performance. Int J Sports Physiol Perform, 8, 104-106.

Hoffman, J., Ratamess, N. A., Kang, J., Mangine, G., Faigenbaum, A., & Stout, J. (2006). Effect of creatine and ß-alanine supplementation on performance and endocrine responses in strength/power athletes. Int J Sport Nutr Exerc Metab, 16, 430-446.

Hopkins, W. G., Schabort, E. J., & Hawley, J. A. (2001). Reliability of power in physical performance tests. Sports medicine, 31(3), 211-234

Hulleman, M., De Koning, J. J., Hettinga, F. J., & Foster, C. (2007). The effect of extrinsic motivation on cycle time trial performance. Medicine and science in sports and exercise, 39(4), 709.

Ivy, J. L., Kammer, L., Ding, Z., Wang, B., Bernard, J. R., Liao, Y. H., & Hwang, J. (2009). Improved cycling time-trial performance after ingestion of a caffeine energy drink. International journal of sport nutrition.

Laursen, P. B., Shing, C. M., Peake, J. M., Coombes, J. S., & Jenkins, D. G. (2002). Interval training program optimization in highly trained endurance cyclists. Medicine and Science in Sports and Exercise, 34(11), 1801-1807.

Lebrun, C. M. (1993). Effect of the different phases of the menstrual cycle and oral contraceptives on athletic performance. Sports Medicine, 16(6), 400-430.

Noonan, V., & Dean, E. (2000). Submaximal exercise testing: clinical application and interpretation. Physical therapy, 80(8), 782-807.

Nethery, V. M. (2002). Competition between internal and external sources of information during exercise: influence on RPE and the impact of the exercise load. Journal of Sports Medicine and Physical Fitness, 42(2), 172.

Nikolopouios V, Arkinstall MJ, Hawley JA. Pacing stråtegy insimulãted cycle time{rials is based on perceived,rather thân actual distañce. J Sci Med Sport 2001;

Nyakayiru, J., Jonvik, K. L., Pinckaers, P. J., Senden, J., Van Loon, L. J., & Verdijk, L. B. (2016). No Effect of Acute and 6-Day Nitrate Supplementation on VO2 and Time-Trial Performance in Highly-Trained Cyclists. International Journal of Sport Nutrition and Exercise Metabolism, 1-25.

Nummela, A., Hämäläinen, I., & Rusko, H. (2007). Comparison of maximal anaerobic running tests on a treadmill and track. Journal of sports sciences, 25(1), 87-96.

Jeukendrup, A. S. K. E. R., Saris, W. H. M., Brouns, F. R. E. D., & Kester, A. D. (1996). A new validated endurance performance test. Medicine and Science in Sports and Exercise, 28(2), 266-270.

Jeukendrup, A. E. (2004). Carbohydrate intake during exercise and performance. Nutrition, 20(7), 669-677.

Pottier, A., Bouckaert, J., Gilis, W., Roels, T., & Derave, W. (2010). Mouth rinse but not ingestion of a carbohydrate solution improves 1-h cycle time trial performance. Scandinavian journal of medicine & science in sports, 20(1), 105-111.

Sands, W. A., McNeal, J. R., Ochi, M. T., Urbanek, T. L., Jemni, M., & Stone, M. H. (2004). Comparison of the Wingate and Bosco anaerobic tests. The Journal of Strength & Conditioning Research, 18(4), 810-815.

Saris, W. H., Antoine, J. M., Brouns, F., Fogelholm, M., Gleeson, M., Hespel, P., ... & Stich, V. (2003). PASSCLAIM–Physical performance and fitness. European journal of nutrition, 42(1), i50-i95.

Spriet, L. L., Hargreaves, M., Kjaer, M., Bonen, A., Horowitz, J. F., & Turcotte, L. P. (1995). Exercise metabolism. Exercise metabolism.

Vandenberghe, K., Goris, M., Van Hecke, P., Van Leemputte, M., Vangerven, L., & Hespel, P. (1997). Long-term creatine intake is beneficial to muscle performance during resistance training. Journal of Applied Physiology, 83(6), 2055-2063.

Weinstein, Y. I. T. Z. H. A. K., Bediz, C., Dotan, R. A. F. F. Y., & Falk, B. A. R. E. K. E. T. (1998). Reliability of peak-lactate, heart rate, and plasma volume following the Wingate test. Medicine and Science in Sports and Exercise, 30, 1456-1460.

www.healthylivingheavylifting.com

Appendix I

Recommendations to increase the reliability

Time trials and constant-duration tests are the most reliable and valid tests of physical performances.

Exhaustion incremental tests could be used in the pre-trial measurements of Wmax and VO2max.

In the study include only athletes and if it is possible at same training level (similar maximal aerobic capacity) and similar sports (to reduce factors as training status and technical skills).

No difference between male and female athletes.

Mode of exercise does not influence test but I would avoid rowing and swimming (influenced by technical skills and very specific for few classes of athletes). The best could be cycling.

The duration of the test should not be longer than one hour for trained athletes, for untrained subjects should be less (about 20 minutes).

Before start the test, participants should perform at least one trial to get familiar with the test and receive eventually instruction to make in the proper way the test.

Encouragements should be standardized and music should be avoided.

Subjects should work at the test only with staff and not in presence with other participants (subjects could compete between each other).

Inter-trial time should not be too short but also not too long. If it is possible about one week.

During the period of the study, athletes should not change their training routine. If it is possible the study should be done during the off-season or in absence of competitions.

Environmental situation, as room temperature and humidity, must be keep constant in each trials.

Subject should wear the same clothes and same shoes during each trials.

All trials of a single subject should be performed at the same time of the day.

Feedbacks should not be given after a single trial because may influence the next performances.

Measures of blood or lactate during the exercise should be done only if they are really necessary.

Subject should not have intensive trainings the days before the test.

Subjects should be advised about the meals the day before the trials, or received food parcels, trials to avoid variation.

Diet should be quantitatively (calories) and qualitatively (macronutrients) constant.

Meals before the test should not influence the treatment.

No tobacco, alcohol and caffeine, before the trial.

Eventually food intolerances and particular diet restrictions should be considered in the inclusion and exclusion criteria of the study.

Appendix II

Information Brochure

PERFORM

Master thesis project: Reliability of common performance measures (PERFORM)

Introduction

Dear Sir/Madam,

You are asked to take part in this project.
Participation is voluntary. Participation requires your written consent. Before you decide whether you want to participate in this study, you will be given an explanation about what the study involves. Please read this information carefully and ask the investigator for an explanation if you have any questions. And you may also discuss it with your partner, friends or family.

1. General information

This project is carried out at Wageningen University as part of a Master thesis of 4 Nutrition and Health Master students.

2. Purpose of the study

The purpose of this study is to investigate the variability in strength and endurance exercise performance over time, both in trained and untrained individuals.

3. Background of the study

Exercise performances (as strength or endurance) can be difficult to measure reliably because there are different factors which may influence these measures, such as temperature, hydration and food intake. Many of these factors can be controlled in laboratory settings but still there remains variability in the results of physical tests. The aim of this master thesis project is to develop physical tests which can be used to measure strength performance (one test) or endurance performance (two tests). These three physical test protocols will be tested both in trained and untrained participants. Every participant will perform one of the tests several times, so we can compare the variation in the results of that test over time for each participant.

4. What participation involves

our participation will last about 5 weeks.
We will first evaluate whether you may participate. The investigator will ask you about your health and your training status.

You are eligible to participate in this research when:

- You are male and aged 18 - 35 years.

- You are in good general health and do not have any injuries that may interfere with your performance (this will be checked by filling in a questionnaire).
- You do not use any drugs or medicine that may interfere with exercise capacity, e.g. all substances on the international doping list.
- You fit in one of the following groups, you:
    - Are a *cyclist* and you have performed endurance-like exercise or training on a regular basis of 4-10 hours per week in the past year.
    - Are a *recreational weight lifte*r and you have performed resistance-like exercise or training on a regular basis with a minimum of three sessions per week in the past year.
    - Are *not a cyclist* and you have exercised a maximum of 1 times a week in the past year and you have a BMI < 28
    - Have not performed any resistance-like exercise or training in the past year *and* you have exercised a maximum of 1 times a week in the past year *and* you have a BMI < 28

Visits and tests

You will visit the research facility 5 times over a period of 5 weeks. The first and the second visit will take 30-45 minutes and the other visits will take approximately 1 hour. The study will be performed at the university (the endurance/cycling part) and the Hospital Gelderse Vallei in Ede (the resistance/leg press part).

5. What is expected of you

In order to carry out the study properly, it is important that you follow the study instructions.

The study instructions require that you:

- Do the exercises as directed.
- Do not participate in another medical study involving exercise.
- Keep appointments for visits.
- Avoid large changes in your habitual training loads and habitual diet
- Do not train progressively during the 5 weeks of the project
- Avoid strenuous exercise 48 hours preceding the test days
- Refrain from alcoholic and drug (cannabis etc.) consumptions 24 hours preceding the test days

It is important that you contact the investigator if you:

- Start using other medicines. Also if these are homeopathic or natural remedies, vitamins and/or over-the-counter medicines.
- Are admitted to hospital or are going there for treatment.
- Suddenly develop any health problems.
- No longer want to participate in the study.
- If your contact details change.

6. Activities

Based on the filled in questionnaire during the information session, all participants will be assigned to a group.

Group 1: trained cyclists – constant duration tests

Group 2: trained cyclists – 10km tests

Group 3: untrained participants – 10km tests
Group 4: recreational weight lifters – leg press tests
Group 5: untrained participants – leg press tests

The following will take place:

Week 1 (screening):

- We will do a physical examination (weight, length and body composition via skinfold measure)
- You will perform a maximal endurance capacity test (Wmax and VO2max) to determine your aerobic capacity (group 1, 2 and 3)
- You will perform a submaximal test on the leg press to estimate the maximal weight you can lift in one repetition (group 4 and 5)

Week 2, 3, 4 and 5:

- You will perform a constant duration test: cycle for 15 minutes as fast as you can (group 1)
- You will perform a time trial: cycle 10 km as fast as you can (group 2 and 3)
- You will perform a one repetition maximum test (1RM) on a leg press to determine the maximal weight you can lift in one repetition. You will start with a warm-up that will be followed by a series of attempts with a gradual increase of the weight until you reach the maximum. (group 4 and 5)

7. Possible side effects/complications (and other)/undesirable effects/discomforts

During the study you will perform maximal tests, therefore your legs may feel a bit of tired and heavy after the test and you can experience muscle soreness in the 48 hours after the tests. This is a completely normal response to resistance or endurance training.

8. Possible advantages and disadvantages

Possible benefit of participating:

- The tests give you an insight in your current physical strength or endurance capacity, and body composition. You may adjust your training schedule with this information

Disadvantage of participation in the study may be:

- possible muscle soreness in the 48 hours after the test

9. If you do not want to participate or you want to stop participating in the study

It is up to you to decide whether or not to participate in the study. Participation is voluntary.
If you do participate in the study, you can always change your mind and decide to stop, at any time during the study. You do not have to say why you are quitting, but you do need to tell the investigator immediately. The data collected until that time will still be used for the study.
If there is any new information about the study that is important for you, the investigator will let you know. You will then be asked whether you still want to continue your participation.

10. End of the study

Your participation in the study stops when

- you have completed all the visits according to the schedule as described under point 4
- you choose to stop
- the investigator considers it is best for you to stop

The study is concluded once all the participants have completed the study.
After processing the data, the investigator will inform you about the most important results of the study. This will happen about 3 months after your participation.

11. Usage and storage of your data

All your data will remain confidential. Some people may access your personal data. This is to check whether the study has been conducted in a good and reliable manner. General information about this can be found in the general brochure on medical research.
People who may access your data are: the study team, the safety committee supervising the study, and the Healthcare Inspectorate. They will keep your data a secret. If you sign the consent form, you consent to your medical and personal data being collected, stored and accessed.

12. Compensation for participation

You will be compensated for participation in this study: € 12 per test day, with an additional € 15 for completion of the project (so, € 75in total). No other (travel) costs will be reimbursed. This reimbursement should be communicated to the Tax Authorities as income. If you stop before the study is over, you will receive a smaller amount.

13. Any questions?

If you have any questions, please contact the project team.

14. Signing the consent form

When you have had sufficient time for reflection, you will be asked to decide on participation in this study. If you give permission, we will ask you to confirm this in writing on the appended consent form. By your written permission you indicate that you have understood the information and consent to participation in the study. You will get a copy or a second copy of this consent form.

Thank you for your attention.

Contact details

Research team

Francesco Di Martino Comaschi (strength)

Renee de Meijer (strength)

Lieke Martens, BSc (endurance)

Pieter ten Kortenaar, BSc (endurance)

Dr. Marco Mensink (project leader)

Contact

Division of Human Nutrition, Research team PERFORM study

perform.study@wur.nl

Tel. 0317-482646

Location information meeting and cycle tests

Helix (building 124)

Stippeneng 4

6708 WE Wageningen

Location leg press tests

Hospital Gelderse Vallei

Willy Brandtlaan 10

Postbus 9025

6710 HN Ede

- I have read the participant information brochure, and have been allowed to ask additional questions. My questions have been answered thoroughly, and I have had enough time to decide whether I want to participate or not.
- I am aware the participation is voluntary. I know that I can decide to end/not participate (anymore) without the need of providing a reason for withdrawal.
- I know that some of the investigators will be able to see my data.
- I allow for the use of my data for the aims as described in the information brochure.
- I want to participate in this research

Name participant:

Signature: Date: ___ /___ /_____

Herewith, I declare that I have informed this participant about the aforementioned research.

I will timely inform the participant if any information that could influence the participant's decision to participate is brought to light during the research.

Name researcher (or his/hers representative):

Signature: Date: ___ / ___ / _____

Appendix III

Questions to assess eligibility for participation

General information

1. I have signed the informed consent for this study Yes/No

Please do not answer any further questions if you didn't sign the informed consent (yet)!

2. Name: ……
3. Date of birth: ___ /___ /_____ (mm/dd/yyyy)
4. What is your gender? M/F
5. What is your age? …… years
6. Adress: ……
7. PC + City: ……
8. Phone number(s): ……
9. Email address: ……
10. Are you currently involved in any other scientific research? Yes/No

General health and habits

11. Do you smoke? Yes/No
12. Are you a blood donor? Yes/No
13. Do you have diabetes? Yes/No
14. Do you have any cardiovascular disease or hypertension? Yes/No
15. Are you using any drugs that are on the international WADA doping list, or drugs that may interfere with your performance, such as beta-blockers? Yes/No

Physical

16. What is your weight? ……kg
17. What is your length? …….m
18. Can you estimate how many weekly hours of endurance training have you performed over the past year? …….. hours
19. Can you estimate how many weekly sessions of strength training have you performed over the past year? ……. sessions
20. Have you been participating in any race or event involving bodily strength or endurance in the past year? Yes/No
21. Do you plan on performing much more or much less endurance or strength training in the upcoming two months than usual?
22. Do you have any injuries that hinder you in performing exercise at the moment? Yes/No
23. Are you able to perform 5 times in our laboratory in 5 consecutive weeks? (the last three weeks on the same (part of the) day?) Yes/No
24. In the period 7 November to 13 December, I will be able to participate on (encircle if applicable):

five consecutive **MONDAYS**, in the MORNING/AFTERNOON/BOTH
five consecutive **TUESDAYS**, in the MORNING/AFTERNOON/BOTH
five consecutive **WEDNESDAYS**, in the MORNING/AFTERNOON/BOTH
five consecutive **THURSDAYS**, in the MORNING/AFTERNOON/BOTH
five consecutive **FRIDAYS**, in the MORNING/AFTERNOON/BOTH

Appendix IV

Oxycon

- Switch on the system by the green button
- Turn on the computer
- Log in with password Master
- Wait 11 minutes (time is recorded on screen)

- Press Ambient conditions → Wait till values appear
- F12 exit → save YES

- Press Auto calvol oxycon
- Take care that there are no objects (like clothes) in front of the V-tube
- F1 Start
- F12 exit → save YES

- Press Gas analyser calibration
- Open the gas bottle (against the clock)
- Settings → entry of cal.gas concentr. O_2 :15.99 $C0_2$:5.00 → Ok
- F1 start → Wait till values appear (PRC gas values close to set values)
- F12 exit → save YES
- **Close the gas bottle !!**

Oxycon ready to use

- Patient data
  - F1 Current patient
  - F2 New patient → Fill in data patient (LBW not necessary)
    → extended patient data not necessary
  - F3 Search patient
- Determine weight participant (only short)
- Determine length participant (without shoes)
- F12 exit → data saved automatically

Ready to test participant

- Take care that the twin tube is in a horizontal setting
- Do not forget the nose clip
- Put on the mask

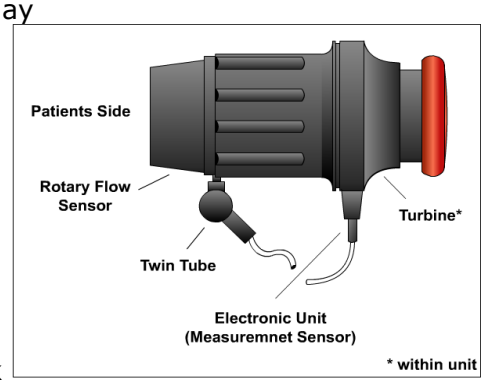
Breath by Breath ergospirometry

- Press Breath by Breath ergospirometry
- Test settings ; bicycle
- Deadspace: 110 CombiToxMask
- Method: Mean
- Breaths: 8
- Mark; skip min. and max values

- F1 start
  - 1. Background testing → **Wait till ok!**
  - 2. Rest
  - 3. Reference
  - 4. Test
  - 5. Recovery
  - 6. Stop
- F12 exit → Save measurement YES

Ergometer

- Turn on ergometer with button in front of bike
- Display box → system parameters → modus installed → hyperbolic
- Manueel → Press up and down buttons to set Power in Watt
- Make the heart rate monitor wet and put around one's middle
- Set seat position of participant
- Cycle for 45 minutes
- Display box → Esc
- Systeem parameter → Modus instellen → Linear → Opslaan JA
- Sys
- Manueel → Set Alfa with up and down arrows
- Cover display



- Show clock
- Cycle 15 minutes

- Switch off ergometer with button in front of bike

Hygiene

- Remove measurement sensor
- Remove sample tube (Twin tube)
- Put rotary flow sensor into a cleaning bath with the Descogen solution. Use a cloth damped with Descogen solution to disinfect the measurement sensor
- After at least 15 minutes remove and rinse parts thoroughly with distilled water
- Dry parts thoroughly after disinfection
- Reassemble Triple V in reverse order

Masker

- Put the breathing mask in a cleaning bath with the Descogen solution
- Rinse them with distilled water
- Dry them thoroughly after disinfection

Nose-clip

- For hygienic reasons the foam pads should be replaced after every use

Appendix V

INTRODUCTION

The purpose of this diary is to keep track of your food and beverage intake and disease symptoms the day before the test days and on the test days.
Therefore you will start this diary the day before the first test day until the first test, you will start again the day before the second test day until the second test etc.
In order to keep these circumstances as fixed as possible we ask you to use your intake before and during the first test day as a guideline for the remaining test days. So please try to keep your food and fluid intake as identical as possible on the day before the test days and during the test days itself.

On the following pages you find guidelines for recording food intake and disease symptoms. It is important that you read these guidelines carefully before you start recording.

For questions about this project you can contact:
Lieke Martens
Pieter ten Kortenaar
Francesco Di Martino Comaschi
Renée de Meijer

E-mail: perform.study@wur.nl
Tel: 0317-482646

GUIDELINES NUTRITION AND PHYSICAL ACTIVITY

- We ask you to refrain from alcoholic consumptions and soft drugs (e.g cannabis) the day before the test and the test day.
- We ask you to keep your diet constant during the period of this project.
- We ask you to eat and drink approximately the same on the day before and during all the test days.
- We ask you to refrain from strenuous exercise the day before the test days, but to maintain your habitual training load throughout the week.
- We ask you to keep your transport to our research facilities the same on every test day. (Please don't take the bus on the first day and go by bike on the second day).

Disease symptoms and possible details/remarks
Disease symptoms (mark the appropriate box): • No symptoms • Cold symptoms: Runny nose, cough, sore throat, sneezing, runny nose • Flu symptoms: Fever, headache, weakness, fatigue, chest pain • Feeling sick, vomiting, diarrhoea • Sore muscles, joint aches • Allergies: itching eyes, runny nose • Other:
Details/ remarks

Food intake
Breakfast:
Snack:
Lunch:
Snack:
Dinner:
Snack:

Disease symptoms and possible details/remarks
Disease symptoms (mark the appropriate box): • No symptoms • Cold symptoms: Runny nose, cough, sore throat, sneezing, runny nose • Flu symptoms: Fever, headache, weakness, fatigue, chest pain • Feeling sick, vomiting, diarrhoea • Sore muscles, joint aches • Allergies: itching eyes, runny nose • Other:
Details/ remarks