Examination of four different instruments for measuring the blood lactate concentration

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ABSTRACT

There is incomplete information on the performance of different instruments used to measure the blood lactate concentration. We have therefore examined instruments from Yellow Springs Instruments (YSI 23L and YSI 1500), and three cheaper and simpler instruments: Dr. Lange’s LP8+, Lactate Pro from Arkray in the KDK corporation, and Accusport from Boehringer Mannheim. First a number of blood samples were analyzed by standard enzymatic photofluorometry (our reference method) and in addition by one or more of the instruments given above. Second, measurements done by two or more «identical» instruments were compared. Third, since Lactate Pro and Accusport are small (≏100 g, pocket-size) battery-driven instruments that might be used for outdoor testing, the performance of these instruments was examined at simulated altitudes (O₂ pressure of <10 kPa) and also at temperatures below −20 °C while screening the instruments as much as possible from the cold. Most of the different instruments showed systematically too high or too low values (10–25 % deviation). The observed differences between instruments may affect the «blood lactate threshold» by 2–5 %. We found different readings between “equal” YSI 1500 instruments, while we saw no such difference when comparing the other instruments of the same type. Lactate Pro gave reliable results both at −21 ± 1 °C and at simulated altitude. Accusport gave reliable results in the cold but 1.85 ± 0.08 mmol L⁻¹ (mean ± SD) too high readings at the simulated altitude. Of the three simpler instruments examined the Lactate Pro was at least as good as the YSI instruments and superior to the other two tested.

KEYWORDS: Bicycling; Blood; Exercise; Lactate; Lactate threshold; Plasma; Training; Testing.
INTRODUCTION

The blood lactate concentration is often measured in relation to training and testing of athletes (1, 2) and also for patients (3). This was traditionally done by time-consuming methods in the laboratory, but faster and simpler methods have been developed as a consequence of technologic development. Today there are several different instruments that can measure the lactate concentration in a small blood sample in a few minutes or less. We have examined properties and qualities of different instruments.

All measurements are subject to some random error even if minimized as much as possible. Systematic errors may also exist. One aim has been to compare the imprecision of different instruments and also to look for possible systematic errors. We have in addition looked for possible differences between instruments of the same type by measuring the same blood sample on two or more similar instruments. Two of the instruments examined, the Lactate Pro™ from Arkray in the KDK corporation and the Accusport® from Boehringer Mannheim, are battery-driven pocket-size instruments that might be suitable for outdoor testing too. Thus, the performance of these instruments was examined at simulated altitude and in the cold. In addition to the instruments mentioned above we have also examined instruments from Yellow Springs Instruments (YSI 23L and YSI 1500) and the LP8+ from Dr. Lange. We have as our reference method used a standard enzymatic photofluorometric method.

SUBJECTS, PROCEDURES AND METHODS

Subjects

Healthy young to middle-aged trained men and women have served as subjects in these experiments. All subjects were told that they served as volunteers in our experiments. The subjects were also told that they were free to leave the experiments at any stage and that they could do so without giving any reason.
Experiments

Most of the exercises have been carried out as bicycling on ergometers, but blood samples have also been taken during treadmill running, rollerskiing, bicycling outdoors, and as canoeing or rowing on ergometers. In some experiments described in more detail elsewhere (2) blood was drawn from catheters in the femoral artery and vein. Otherwise capillary blood has been taken from a finger after the hand has been warmed for at least 30 s in temperated water. This procedure increases the perfusion of the hand and thus makes it easier to get enough blood for several samples in sequence. Sweat contains lactate, and washing in water will also dissolve and thus remove lactate on the skin that otherwise could contaminate the blood. The skin was punctured by a lancet, the first drop of blood was wiped off, and thereafter blood was taken as explained in further detail below.

Blood samples were in some experiments taken in connection with intense exercise to exhaustion, in other experiments as part of testing subjects and finding their «blood lactate threshold» and maximal O₂ uptake, and finally in some experiments with strenuous exercise with a significant anaerobic energy release but where the exercise was stopped before exhaustion. We have altogether taken around 800 blood or plasma samples that have been measured by at least two different methods or instruments, and altogether nearly 3000 single measurements of the lactate concentration in blood or plasma samples have been carried out.

Methods

Enzymatic photofluorometry

A volume of 25 or 50 µl of whole blood or plasma was taken by Acupette capillary tubes (P4518-50, Dade Diagnostics inc., Puerto Rico, USA; according to the manufacturer the accuracy of the tubes’ volume is better than 0.5 % and the coefficient of variation is better than 1 %). The fluid was transferred to a tube containing 500 µl of 0.4 mol L⁻¹ perchloric acid (PCA) that lyzes the red blood cells and thus frees lactate inside the cells. These tubes were stored frozen at –20 °C until later analyses of the lactate concentration by a method according
to Passoneau and Lowry (4). That method uses the increase of NADH-concentration in the LDH-reaction (lactate dehydrogenase, EC 1.1.1.27; from beef hearts, L2625 type III from Sigma, St. Louis, MO, USA). Pyruvate produced was further processes by the glutamate-pyruvate transaminase reaction (l-alanine:2-oxoglutarate aminotransferase, EC 2.6.1.2; from swine heart, Boehringer Mannheim GmbH, Mannheim, Germany), thus allowing almost complete processing of all lactate in the sample. The fluorescence was read off in an RF-5000 spectrofluorophotometer (Shimadzu, Kyoto, Japan) at 460 nm (652 THz) after 45 min incubation at 23.5 °C.

For each analysis a new second-order standard curve that covered the whole range of fluorescence values was made. The error of regression (scatter around the regression line, the statistical error in reading off from the standard curve) was 0.1–0.2 mmol lactate L⁻¹ blood (or plasma). A standard solution of 1.00 mol lactate L⁻¹ (L-lactat(e) standard 125 440, batch 66762401 from Boehringer Mannheim) was used for making the samples for the standard curve. The method’s total imprecision is 0.1–0.3 mmol L⁻¹, depending on the lactate concentration in the measured sample. The volume fraction of water in whole blood was taken as 84 % and that of plasma as 94 % (5).

Since we found systematic differences between the results given by the tested methods and our reference method, the standard solution from Boehringer Mannheim was calibrated independently by an enzymatic reaction similar to that described above (4). The increase in the NADH-concentration was read off in a Shimadzu MPS-2000 spectrophotometer at 340 nm (882 THz) in quarts cuvettes (type 18-B, Starna, Essex, UK) where the light traversed 10.00 mm. The lactate concentration was taken from the measured increase in the NADH-concentration using a coefficient of extinction of NADH of 6270 m⁻¹ mol⁻¹. These measurements showed that the lactate concentration of the standard solution did not differ from the reported value, and the imprecision (SE) in this calibration was 1 %. That imprecision was included when we examined whether there was a systematic difference between a tested instrument and the reference method. Otherwise the possible error in the reference method is considerably less than the systematic deviations given later and is thus regarded as without significance for this study.
YSI instruments

The lactate concentration in samples of 25 µl was measured on an YSI 23L (Yellow Springs Instruments, Yellow Springs, OH, USA) on nonhemolyzed blood and on plasma samples. Further blood analyses were done on five YSI 1500 instruments on either hemolyzed or unhemolyzed 25 µl blood samples. All YSI instruments were calibrated by 5 mmol lactate L⁻¹ solutions (YSI 2327). As an additional control a standard solution of 15 mmol lactate L⁻¹ was used (YSI 2328); on some occasion we also used a 30 mmol L⁻¹ solution (YSI 1530). We have in accordance with the instruments’ manuals required that readings with these solutions should give 5.0 ± 0.1 mmol L⁻¹, 15.0 ± 0.7 mmol L⁻¹, and 30.0 ± 1.5 mmol L⁻¹, respectively, before any analyses were carried out.

YSI instruments assume that a known and constant amount of blood is used for each measurement, and the instruments are equipped with two types of pipettes, a pipette with needle (YSI 1501 syringepet) and a pipette using capillary tubes (YSI 1502 pipette). We used in agreement with the manufacturer’s instructions the same pipette for both the calibrations and the subsequent measurements. When analyses were done on more than one YSI instrument per day, the same syringes and calibration solutions were used for all instruments, and all instruments were in addition handled by the same operator.

In blood the lactate is found in both plasma and the red blood cells; and in most cases ≈75% of the lactate in blood is found in the plasma compartment (1, 2). According to the manuals of the YSI instruments only lactate in plasma is measured unless the blood samples are hemolyzed.

Dr. Lange miniphotometer LP 8+

Blood was taken by 10 µl end-to-end capillaries and placed in a reagent solution hemolyzing the blood, lactate was processes in a reaction producing quinonimin in proportion to the amount of lactate in the sample, and the concentration of quinonimin was read off in an LP8+ apparatus (Dr. Bruno Lange GmbH, Berlin, Germany) at 540 nm (576 THz) after a 3 min reaction time.
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Lactate Pro
The lactate concentration has been measured in blood and plasma samples on altogether seven different LT-1710 Lactate Pro™ analyzers (Arkray factory inc., KDK corporation, Shiga, Japan). This instrument measures lactate on a tiny drop of 5 µl of fluid. The lactate concentration is read off after 60 s. We have for our measurements used strips with production numbers L8D04A (F-4) and L8L07B (F-7).

Accusport
The blood lactate concentration has been measured on two different Accusport® portable lactate analyzers (type 1488767, Boehringer Mannheim). A drop of no less than 15 µl of blood is applied to the strip (6), and 60 s later the lactate concentration is read off. We have in accordance with the manufacturer’s instructions checked the «window» on the under side of each strip after the analyses and examined the coloring; if uneven, the analysis was rejected. We have also checked that enough blood was added to each strip: When blood is added, some penetrates the surface and thus reaches chemicals that start the reactions processing the lactate. We required that there should be liquid blood on the top of the strip’s pad after each analysis. Otherwise the result was rejected. We have thus used 25–50 µl of blood for each analysis. Blood was added to the strip by letting it drip from a finger; in accordance with the instrument’s instructions we never let the finger touch the strip’s pad.

This instrument only measures the lactate in plasma, but built-in equations shall according to the manufacturer calculate the concentration in whole blood from the measured value in plasma (7). We have for our measurements used strips from Boehringer Mannheim with code number 512.

In general, for all instruments all measurements were carried out in agreement with the instructions for each instrument. All measurements were done by experienced test leaders who each had done at least a thousand analyses of the blood lactate concentration.
Measurements at simulated altitude

Control solution from Boehringer Mannheim (article no. 1447335) with a reported lactate concentration of 3.6 mmol L\(^{-1}\) (low; BM-control-Lactate 1) and 9 mmol L\(^{-1}\) (high; BM-control-Lactate 2) were measured in a chamber where the O\(_2\) pressure was less than 10 kPa, corresponding to an altitude of more than 6 km above the sea level. In practice N\(_2\) gas was led into a bag serving as the chamber. The O\(_2\) pressure in the bag was measured by a Metamax 1 O\(_2\) analyzer (Cortex Biophysic GmbH, Leipzig, Germany), and no analysis was done unless the O\(_2\) pressure was less than 10 kPa. Each standard solution was measured at least ten times by each of the instruments Lactate Pro and Accusport. As a control the same control solutions were measured at least ten times at normal laboratory conditions, that is at a temperature of 22 ± 1 °C and at an air pressure of ≈100 kPa, corresponding to an O\(_2\) pressure of ≈21 kPa.

Measurements in the cold

Each of the control solutions from Boehringer Mannheim (3.6 and 9 mmol L\(^{-1}\) nominal lactate concentration) was measured at least ten times on each of the Lactate Pro and the Accusport in a freezing storage room, that is at a temperature of –21 ± 1 °C. The test leader was well clothed carrying a thick sweater and coat. To shield the instruments as much as possible from the cold the test leader wore a bag under the sweater and thus close to the body. While the strip was inserted in the instrument and the control solution was applied to the strip, actions that took 20–30 s, the instrument and the strip were exposed to the cold. As soon as the analysis started the instrument was placed in the bag and thus was no longer exposed to the cold. The instrument and the strips lay in the bag between each analysis too. The temperature in the bag was measured by a digital thermometer. It appeared that repeated opening and closing of the bag lowered the temperature, and if it dropped to 5 °C or lower, the experiments were stopped.

Statistics

The values are given as means ± SD unless otherwise stated explicitly. Different instruments or methods have been compared by linear regression, using the approach geometric mean that gives equal weight to errors in both series of measurements (8, 9); standard linear regression assumes that all errors or imprecision is caused by measurements of the ordinate (Y). For the
regression analyses carried out here the approach geometric mean gives the same results as a more demanding, robust, and distribution-free approach (10), and the method by Bland and Altman (11) also gives corresponding results for our data. If two instruments or methods differ systematically, the slope will differ from 1.00, and the data points will not be randomly distributed around the line of identity. Thus, possible differences have been tested by examining the residual and by standard t-tests on the slope. As stated above we have also paid attention to the fact that our reference method may have a systematic error of $\approx 1\%$ (SEM). The error of regression (scatter around the regression line) is used as a measure of the random errors. Different errors of regression have been tested by Fisher-tests (12).

Errors may occur in all measurements. When measurements with two methods or instruments turn out as expected, the values will fall close to a line. If there is something wrong with one of the measurements, the corresponding point will deviate from the linear relationship. A simple scatterplot alone cannot decide whether the deviation seen is due to an error in the abscissa (X) or in the ordinate (Y). For a number of the samples analyzed here each sample has been measured by at least three different instruments or methods, here called X, Y, and Z. If a point appears as an outlier in the XY-plot and the YZ-plot but not in the XZ-plot, it is likely that the deviation is caused by an error in the Y-measurement. We have used this principle to unravel possible errors of measurements and outliers.

RESULTS

Blood samples were taken during different experiments, and the blood lactate concentration was measured by at least one of the instruments to be tested (Y) and in addition by enzymatic photofluorometry (X) serving as our reference method (Fig. 1).

Nonhemolyzed blood samples measured on the YSI 23L showed in average a 22\% lower value than the reference method ($P < 0.001$; Fig. 1A). Since the YSI 23L does not hemolyze the blood, it does not record lactate in the red blood cells, and this may have caused the systematic deviation. Hemolyzed blood samples were measured on two different YSI 1500, here referred to as the YSI 1500\textsubscript{1} and the YSI 1500\textsubscript{2}. The values from the
YSI 1500₁ were in average 20% higher than those of the reference method (P < 0.001). For the YSI 1500₂ the values were in average 5% higher than those of the reference method; these latter values did not deviate significantly from the line of identity (Fig. 1B, C).

**Figure 1.** The blood lactate concentration measured by enzymatic photofluorometry (X) and by six different instruments (Y). The samples measured by the YSI 23L (panel A) were on nonhemiolyzed blood, and these measurements do not record lactate inside the red blood cells. YSI 1500₁ and YSI 1500₂ (panels B and C) refer to measurements on two different YSI 1500 instruments. LP 8+ (panel D) is a lactate analyzer from Dr. Lange, Lactate Pro (panel E) is from Arkray in the KDK Corporation, while Accusport (panel F) is from Boehringer Mannheim. Data on the regressions are given for each set of data, and the regression lines are shown as solid lines. S_y|x is the error of regression and is expressed in mmol L⁻¹. S_b is the error of the slope. The thin dashed lines show the lines of identity for each set of data. ▼ refer to accepted measurements, while ▼ refer to outliers that were formally rejected because of an error of measurement by the instrument examined; these values were therefore not used to calculate the regression lines.
Figure 2. The blood lactate concentration measured by enzymatic photofluorometry (X) and by six different instruments (Y). The data are the same as in figure 1, but only samples with a blood lactate concentration less than 6 mmol L\(^{-1}\) as measured by enzymatic photofluorometry are included. The samples measured by the YSI 23L (panel A) were on nonhemolyzed blood, and these measurements do not record lactate inside the red blood cells. YSI 1500\(_1\) and YSI 1500\(_2\) (panels B and C) refer to measurements on two different YSI 1500 instruments. LP 8\(+\) (panel D) is a lactate analyzer from Dr. Lange, Lactate Pro (panel E) is from Arkray in the KDK Corporation, while Accusport (panel F) is from Boehringer Mannheim. Data on the regressions are given for each set of data, and the regression lines are shown as solid lines. \(S_y\) is the error of regression and is expressed in mmol L\(^{-1}\). \(S_b\) is the error of the slope. The thin dashed lines show the lines of identity for each set of data. \(\nabla\) refer to accepted measurements, while \(\downarrow\) refer to outliers that were formally rejected because of an error of measurement by the instrument examined; these values where therefore not used to calculate the regression lines.

Blood samples were also measured on three different simpler instruments. The values from the LP8\(+\) were in average 26 \% higher than those of the reference method (P < 0.001; Fig. 1D). These measurements showed also a larger error of regression than the other measurements (P < 0.05). The relationship to the reference method was convex, and a second-order curve fit was better than a linear one (P < 0.001). Values from measurements on the Lactate Pro were in average 12 \% higher than those from the reference method (P < 0.001; Fig. 1E). For all of the instruments mentioned so far all the regression lines found go close to the origo.
This means that possible systematic errors in the measurements were small for low blood lactate concentrations and rose roughly proportional to the lactate concentration. The Accusport showed two kinds of deviations. First the Y-intercept was close to 1.0 mmol L\(^{-1}\) (P < 0.001 versus an intercept of 0.0). In line with this the instrument reported blood lactate concentration around 2 mmol L\(^{-1}\) even for blood samples taken at rest, that is in samples with a true concentration less than 1 mmol L\(^{-1}\). The slope found for this instrument was 0.81, which is considerably less than 1.00 (P < 0.001). Thus, this instrument showed correct values for blood samples with a lactate concentration of \(\approx 5\) mmol L\(^{-1}\). For samples with less lactate the reported values were systematically too high, while for samples with higher concentration the reported values were too low.

When the so-called «blood lactate threshold» is sought in sports testing, one is particularly interested in the instruments’ performance at low to moderate blood lactate concentrations. The same is true for most clinical testing too. Therefore blood samples with a lactate concentration above 6 mmol L\(^{-1}\) as measured by enzymatic photofluorometry were let out, and the remaining data from figure 1 were reanalyzed (Fig. 2). For the YSI 23L the relationship for this reduced data set is largely as for the full set (Fig. 2A). Thus, also within this range of values the YSI 23L showed too low values. The error of regression was roughly half of that found for the full data set. Hemolyzed blood samples measured on the YSI 1500\(_1\) showed in average 13 % too high values (P < 0.001), while for YSI 1500\(_2\) there was no sign of a systematic deviation (Fig. 2B,C).

The values from LP8+ were in average 25 % too high (P < 0.001; Fig. 2D). The error of regression found for this instrument was around twice as large as that found for the other instruments (P < 0.001). The Lactate Pro showed no systematic deviations from the reference method for samples with a blood lactate concentration less than 6 mmol L\(^{-1}\) (Fig. 2E). For the instruments mentioned so far the systematic errors were less both in absolute and relative terms for samples with less than 6 mmol lactate L\(^{-1}\) blood than for samples covering a larger span. The values from measurements by the Accusport were systematically higher than those from the reference method (P < 0.001; Fig. 2F).
The LP8+ showed a larger variability than the other instruments (see figures 1 and 2). We did therefore three parallel measurements on a number of blood samples with this instrument. The median of each measurement was related to measurements with the reference method (Fig. 3A). That approach did not give considerable better values than single measurements as judged from the errors of regression in figures 1D and 3A.

Figure 3. A, the blood lactate concentration measured on LP8+ versus corresponding measurements by enzymatic photofluorometry. The values given for the LP8+ is the median of three parallel measurements. B, the lactate concentration measured on nonhemolyzed blood samples on the YSI 1500 apparatus versus measurements by the Lactate Pro. † show accepted measurements. ‡ show measurements that were rejected because of an incorrect measurement by the instrument examined and therefore not used in calculating the regression parameters. C, the lactate concentration measured on nonhemolyzed blood samples on the YSI 1500 (†), YSI 1500 (‡) and YSI 1500 (§) instruments versus measurements by the Lactate Pro. Regression data are given for each set of data, and the regression lines are shown as solid lines. SY|x is the error of regression and is expressed in mmol lactate L⁻¹ blood. Sb is the error of the slope. The thin dashed lines are the lines of identity. The thin long-dashed line in C is a copy of the regression line in B.

We did further comparisons of measurements on unhemolyzed blood samples measured by the YSI 1500 and by the Lactate Pro. The values found by the YSI 1500 were in average 67 % of those found by the Lactate Pro (Fig. 3B). Correcting for a 12 % bias in the values from the Lactate Pro (see Fig. 1E) suggests that the YSI 1500 on nonhemolyzed blood gives values 25 % less than the true value of whole blood. For a further evaluation of the YSI 1500 blood samples were measured by three other instruments, here called YSI 1500, YSI 1500, and YSI 1500 and by the Lactate Pro (Fig. 3C). The values given by these three YSI 1500s
did not differ systematically, but for a given value as measured by the Lactate Pro these three instruments gave a 13 % higher value than the instrument we called YSI 1500².

The instruction manual gives no information on how the Lactate Pro reflects lactate in the red blood cells and how it responds to plasma. Therefore plasma from blood samples taken after intense bicycling were measured by enzymatic photofluorometry and by the Lactate Pro. For samples with a lactate concentration less than 10 mmol L⁻¹ the Lactate Pro responded as for blood samples, that is the values found by the Lactate Pro were up to 10 % higher than those of the reference method. For samples with a higher plasma lactate concentration the curve leveled off, meaning that the Lactate Pro showed too low values (not shown).

**Figure 4.** The lactate concentration of two control solutions from Boehringer Mannheim with a nominal lactate concentration of 3.6 and 9 mmol L⁻¹ as measured by the Lactate Pro and the Accusport in control experiments at room temperature and normal air and O₂ pressures, at room temperature but with a O₂ pressure below 10 kPa, and at normal air and O₂ pressures but at −21 ± 1 °C.

**Measurements at simulated altitude**

The lactate concentration in two control solutions were measured in a chamber with an O₂ pressure less than 10 kPa, corresponding to that found at an altitude of more than 6 km above the sea level. The values were compared with corresponding measurements at normal O₂-
pressure and temperature (Fig. 4). The Lactate Pro showed no effect of a reduced O₂-pressure. The values found by the Accusport at reduced O₂-pressure were 1.85 ± 0.08 mmol L⁻¹ (mean ± SD) higher than those found in the control experiments (P < 0.001).

Measurements in the cold
The lactate concentration of the control solutions was measured at –21 ± 1 °C (Fig. 4). The instruments were exposed to the cold while a strip was inserted and the control solution was applied to the strips. Otherwise the instruments were well shielded from the cold. The mean values did not differ from those of the control experiments (P > 0.5 for each instrument), but the random variations were larger (P < 0.01).

DISCUSSION

The main results in this study are first that for all the instruments examined the reported blood lactate concentration rose roughly linearly by the value given by the reference method. Most of the instruments showed either systematically too high or too low values, and the deviations were mainly in the range 10–25 %. Apparently equal YSI 1500 instruments showed different values on the same blood samples. Lactate Pro gave reliable results both at simulated altitude and in the cold experiments. Accusport gave too high readings at simulated altitude, while the values in the cold were reliable. The LP8+ was the most biased instrument and showed in addition the largest random variations.

Evaluation of the method enzymatic photofluorometry used here
We measured the lactate concentration in blood and plasma by a method used for more than 30 yr in numerous studies. It is common knowledge that the fluorescence from a sample with a known lactate concentration varies from day to day due to interassay-variations, and consequently a new standard curve is made for each day of analysis.

While standard curves are usually fitted by linear regression, we used second-order regression. First, a slight curved relationship was readily visible; samples with a high lactate
concentration showed a smaller fluorescence than a linear extrapolation dictates. Second, NADH is produced in proportion to the amount of lactate in the sample. There may be a $\approx 5\%$ quenching of the signal in a cuvette with a NADH-concentration of 10 $\mu$mol L$^{-1}$ (4), and since the effect is proportional to the concentration, a parabolic relationship is expected. Finally, in samples with a high lactate concentration 1–2 $\%$ of the lactate in the sample may not be processes (4). This latter effect, although quantitatively less important, will add further curvature to the standard curve. There is no reason to assume that our use of second-order curve fits have caused the possible nonlinear trends in figure 1 since the second-order component of our standard curves was small and in addition tended to reduce rather than increase nonlinearities.

We used a commercial 1.00 mol lactate L$^{-1}$ stock solution to establish the standard curve, and separate laboratory controls showed that the concentration of this stock solution was accurate within the precision of 2 $\%$ (95 $\%$ confidence interval) of the control analyses. A possible bias less than 2 $\%$ or random variations less than 0.3 mmol L$^{-1}$ of our reference method has no influence on the conclusions drawn in this study.

Do differences between instruments matter?

When the same sample was measured on two different instruments or by two different methods, we found typically differences of 10–25 $\%$. To examine how this may affect the so-called blood lactate threshold, that entity, here taken as the exercise intensity giving a blood lactate concentration of 3.00 mmol L$^{-1}$, was established for one subject during bicycling at 1.5 Hz and found to be 3.42 W kg$^{-1}$ for that subject (Fig. 5). To each measured lactate concentration we added (and subtracted) 10 and 25 $\%$. That caused the 3.00 mmol L$^{-1}$ «threshold» to appear at 2 and 5 $\%$ lower (higher) powers, respectively. Systematic differences of that magnitude may be important when judging possible changes for athletes. Thus, for precise testing of possible changes by time the same instrument or instruments known to have similar properties should be used.
Figure 5. Measurements of the blood lactate concentration by stepwise increments of the power during bicycling (thin and thick solid line). Each step of constant power lasted 5 min, and a 1 min rest separated each step to allow blood sampling and setting a higher power. To each measured value first 10% (thin dashed-dotted lines) and 25% was added and subtracted (thin dashed lines). The dotted line shows the level of 3.00 mmol L⁻¹, and the power corresponding to that value is here taken as the «lactate threshold».

There are other definitions of the lactate threshold. For blood lactate concentrations higher than 3 mmol L⁻¹ the curve is steeper, and if for example the «4 mmol L⁻¹ threshold» is used, systematic differences between instruments have less effect. For lower lactate concentration the curve is less steep, and small variations in the measured value will have a larger effect on the reported threshold. For long-distance running, bicycling, and skiing the intensity corresponding to a blood lactate concentration of 2 mmol L⁻¹ may be of interest. We found that the Accusport showed that value even on samples taken at rest. This instrument should therefore perhaps not be used for that kind of testing. It has also been proposed that an intensity giving a lactate concentration of rest value +1.5 mmol L⁻¹ should be sought (13). That definition may be less sensitive to variations between instruments.

Effect of simulated altitude
Control solutions with given lactate concentrations were measured on the Lactate Pro and the Accusport when the O₂ pressure was less than 10 kPa, corresponding to that found at altitudes above 6 km. The readings by the Lactate Pro were not affected by the reduced O₂-pressure, suggesting that this instrument may be well suited for testing at altitude. The Accusport reported too high values in our measurements. Our measurements do not allow us to conclude how this instrument will behave at the O₂ pressures of altitudes of 1500–3000 m that are more typical for athletes’ training.

**Measurements in the cold**

According to the manufacturers both the Lactate Pro and the Accusport may be unreliable for testing at temperatures below +10 °C. Our data suggest that if the instruments and the strips are shielded from the cold except for the 20–30 s when the strips are inserted in the instrument and blood or test solutions are applied to the strips, the reported values are not affected by surrounding temperatures down to –20 °C. Thus, both of these pocket-size instruments may be as suited for outdoor testing as for use in the laboratory. Admittedly, the variations in the measurements were larger in the cold. That was probably caused by technical problems since it was more difficult to add a drop of the test solutions from the nipple of the flasks than to add a drop of blood from a fingertip to the strips, particularly for the Lactate Pro in the cold.

**Evaluation of the different instruments**

**Common comments**

The error of regression is a simple and usually the best measure of how well the model fit the data. In figure 1 the random variations rise by the lactate concentration, and the reported error of regression in that figure is a mean of the error at low and high concentrations. In figure 2 where only samples with a concentration less than 6 mmol L⁻¹ are included, the error of regression is less, and within the range of data in that figure the random variations seem independent on the concentration.
Except for the data on the YSI 23L (figures 1A and 2A) parallel measurements «on the same blood sample» were done by taking two or more samples from the same finger in sequence, that is 10–30 s apart, and in random order. The reported error of regression depends on the imprecision in the measurements by enzymatic photofluorometry (X), in the instrument examined (Y), and in possible variations between blood samples taken some seconds apart. The error of regression was least for the YSI 23L. It is likely that the slightly larger errors found for the YSI 1500s, the Lactate Pro, and the Accusport are a consequence of small variations between blood samples rather than less precise measurements by these instruments. Statistical considerations suggest that the random error of measurements with each of these three instruments is roughly half of the reported errors of regression in figures 1 and 2.

**YSI instruments**

We used one YSI 23L and five YSI 1500s. On nonhemolyzed blood samples the YSIs showed values 20–25 % less than the reference method. This finding agrees with data of Foxdal et al. (14) and Lormes et al. (15). It is clearly stated in the YSI instruments’ user manual that if unhemolyzed blood is measured, lactate in the red blood cells is not recorded. It is well known that after a few minutes of equilibration around 25 % of the lactate in blood is found in the red blood cells (f. ex. 1, 2). Thus, one must expect too low apparent blood lactate concentrations when unhemolyzed blood samples are measured.

We did parallel measurements on two different YSI 1500 instruments. Both had fresh membranes, were calibrated using the same solutions and by the same pipette and handled by the same test leader. Nevertheless, these instruments showed a 12 % systematic difference on the same samples. Three other YSI 1500s were examined and tested versus the Lactate Pro. That latter instrument showed a systematic but reproducible bias versus the reference method and was therefore regarded as suited as a control when comparing different YSI 1500s. These three YSI 1500s showed larger lactate concentration in unhemolyzed blood than the YSI 1500 did as judged from the comparisons with the measurements by the Lactate Pro. We
have no explanation for why different YSI instruments gave different results in our experiments. As stated in the method section, the instruments were operated by experienced test leaders and according to the instruments’ instruction manual.

Dr. Lange’s LP8+
This instrument showed values 25–30% too high, and the error of regression was nearly twice as large for this instrument as for the others. Using the median of three parallel measurements did not reduce the random error much. This instrument costs about the same as Lactate Pro and Accusport and only 10–15% of that of an YSI 1500. Apart for the lower price than for an YSI 1500 we see no advantage with this instrument compared with the others. This instrument performed less well than the others, and we felt that this instrument was more difficult to use than the others both in terms of handgrips (caps repeatedly off and on), to obtain precise measurements (fill the capillaries completely without spilling blood on the outside), and in terms of not mixing samples in larger series.

Lactate Pro
This instrument showed little bias when blood samples with low to moderate lactate concentrations were measured, while we got values 12% too high on samples with a blood lactate concentration above 10 mmol L\(^{-1}\). We saw no systematic difference when the same blood sample was analyzed on different instruments of this type. The random variations in measurements with the Lactate Pro were similar to those of the YSI instruments and the Accusport. Lactate Pro gave reliable results at simulated altitude and in the cold. Shimojo et al (16) have also examined this instrument. They found no bias at high lactate concentration, but their random variation was larger than ours.

There is no information on how this instrument accounts for lactate in the red blood cells. If the instrument measures lactate in plasma only and adds an assumed value for the lactate in the red blood cells, one would expect too high reading when fluids without red blood
cells are measured. Measurements of the lactate concentration in plasma samples and in control solutions (not shown) with Lactate Pro gave reliable results for solutions with a lactate concentration less than 10 mmol L\(^{-1}\). Thus, this instrument must lyze the red blood cells and thus measures the cell lactate. For nonblood fluids with a lactate concentration above 10 mmol L\(^{-1}\) the Lactate Pro gave too low values in our measurements. This instrument may therefore not be suited when fluids other than blood are analyzed and the lactate concentration is high.

**Accusport**

This instrument gave correct values only for blood samples with a lactate concentration of \(\approx 5 \text{ mmol L}\(^{-1}\)\). Even for blood samples taken at rest and with a known concentration less than 1 mmol L\(^{-1}\) the Accusport reported values around 2 mmol L\(^{-1}\). When this instrument reported a value of 3.0 mmol L\(^{-1}\), the true value was \(\approx 2.5 \text{ mmol L}\(^{-1}\) in our experiments, and that error may be important when testing athletes. Our data suggest that there may be an inbuilt error in this instrument. Roßkopf et al. (17) examined the Accusport versus a reference method. They did not find the bias we saw, but their random errors were considerably larger than ours.

Two different Accusport-instruments gave similar results when the same blood samples were measured. The instrument did not work adequately at simulated altitudes above 6 km, but it worked well in our cold experiments.

The Accusport was more difficult to use than the Lactate Pro, and we got a number of incorrect measurements with this instrument. Some errors could be excluded on formal reasons (uneven coloring seen through the «window» on the underside of the strip; too little blood applied), but we also experienced errors that could not be detected without comparisons with other analyses. Several measurements where lost because we accidentally hit the on/off-button when the analysis was started by closing the cover. We had technical problems with both instruments used. One reported «low battery» and dropped out even when new batteries
with a proper voltage were used. The other had problems reading the code on the strips used even when the instrument’s window was properly cleaned. Switching batteries and strips between the two instruments showed that the problems resided in the instruments and not in the strips or batteries.

Summary and conclusions
Different instruments gave different values when the lactate concentration of a blood sample was measured. The differences were usually in the range 10–25 %, and a bias that large may have some effect when athletes are tested. Of the four instruments examined the Lactate Pro was best. The Lactate Pro cost only ≈15 % of an YSI 1500 and is much easier to use. Different YSI instruments gave different values. The LP8+ was inferior to the others. Both Lactate Pro and Accusport may be used for outdoor testing in the cold. Lactate Pro may also be used for testing at altitude.

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