Examination of the Metamax I and II oxygen analysers during exercise studies in the laboratory

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Running title: Examination of Metamax O_2 analysers.

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ABSTRACT

The performance of the Metamax I and the Metamax II portable analysers for measuring the O$_2$ uptake has been examined during exercise. Healthy subjects ran on the treadmill or bicycled on ergometers while the O$_2$ uptake was measured by the Metamaxes and in addition by the Douglas bag technique or the Vmax 29. In the first series of experiments the O$_2$ uptake was first measured by one instrument and thereafter by a second. In later experiments two or more breathing valves were connected in series, thus enabling us to measure the O$_2$ uptake simultaneously by more than one instrument. The O$_2$ uptake measured by the Metamaxes rose linearly by the value given by the control methods. However, there were variations of $\approx 5\%$ because the relationships differed between the subjects. When the data from each subject were examined separately, the error of regression was 0.5–1 μmol s^{-1} kg^{-1} (2–3%), and the error of regression when relating the O$_2$ uptake to the exercise intensity was similar to that found when using the Douglas bag technique alone. In most cases the lung ventilation reported by the Metamaxes was a few percent less than that given by the control methods while the fractional extraction of O$_2$ was higher for the Metamaxes. The respiratory exchange ratio (R-value) reported by the Metamaxes agreed well with those of the control methods only in the range 0.9–1.0; for this parameter the Metamaxes do not seem to be reliable for exercise testing. The O$_2$ uptake and the R-value were also calculated from the raw data reported by the Metamaxes. The calculated values differed somewhat from those given by the instruments, and the calculated values agreed better with those obtained by the Douglas bag technique than those reported by the instrument did. This study suggests that the O$_2$ uptake reported by the Metamaxes is precisely measured within subjects but that there are some systematic errors and in addition variations between subjects.

INTRODUCTION

Exercising muscles release energy by breaking down ATP, and except for very short bursts of intense exercise, aerobic processes dominate the regeneration of ATP [1, 2]. The O_2 consumption, which is increased during exercise, is often used as an indirect measure of the aerobic ATP-turnover rate. The body's O_2 uptake has traditionally been measured by the Douglas bag technique were typically $\approx$100 L of expired air is collected in a bag while the sampling time is recorded. The volume of the expired air is later measured in a spirometer while the temperature and pressure are recorded, a small sample of the air is analysed separately for its fractions of O_2 and CO_2, and the O_2 uptake is calculated from these measurements, using either assumed or measured concentrations of O_2 and CO_2 of the inspired air. This is a precise method, but it is quite time consuming and thus gives a limited time resolution, and it is in addition largely restricted to experiments in the laboratory.

While the fractions of O_2 and CO_2 have traditionally been measured by for example the Scholander technique [3], electronic gas sensors measuring the fractions or partial pressures of O_2 and CO_2 have been available for more than 20 yr. The gas volume or flow can also be measured electronically, and as the electronic sensors have become gradually smaller, portable instruments that allow the O_2 uptake to be measured outside the laboratory have now become available. We have in this study examined the Metamax I and the Metamax II portable analysers and compared data from these instruments with those obtained by the Douglas bag method and by the Vmax 29, the latter being a commercial fully automatic laboratory instrument.

METHODS

Subjects

Healthy men and women 17–46 yr old served as subjects in this study. All were physically active and some were top athletes. The subjects were familiar with exercise testing and the equipment used before any measurements were done. They were informed that they as volunteers could leave the study at any stage without giving a reason for doing so.

Experiments

Alternating measurements by two instruments

Series 1. Nine top bicyclists cycled for 2 h at a power of $\approx$2.9 W kg^{-1} body mass that required an O_2 uptake of $\approx$30 μmol s^{-1} kg^{-1} (40 ml kg^{-1} min^{-1}) and that gave a blood lactate concentration of $\approx$2 mmol L^{-1} and a heart rate of $\approx$155 bpm. The O_2 uptake was measured after 15, 45, 75, and 105 min of bicycling first for 5 min in 2–3 separate Douglas bags and then immediately after by the Metamax I analyser, see [4] for further details.

Series 2. Six well-trained subjects ran at stepwise increasing treadmill speeds for 6 min at each speed. The speeds were chosen to tax 50–90% of the subjects' maximal O_2 uptake. The O_2 uptake was measured at 2–4 min and 4–6 min of running at each speed by the Vmax 29 (Sensormedics, Yorba Linda, CA, USA) and by the Metamax II analysers at each speed. The order of the two instruments used was alternated between each step within each subject, and the instrument to be used first on the first step was randomised between the subjects.

Simultaneous measurements by more than one instrument

In these first two series of experiments the O_2 uptake was measured by only one instrument at a time. The experimental conditions were kept as constant as possible until the measurements had been repeated using the second instrument. However, this second measurement was not done on the same expired air as the first one. Moreover, the breathing resistance of our Douglas bag system was considerably larger than that of a Metamax, and it could be that a different resistance influenced the breathing pattern. To allow more than one instruments do simultaneous measurements on the expired air, we made adapters connecting the breathing valves of two or more instruments in series. More specifically, the breathing valve used to collect expired air in Douglas bags was connected to the outlet of the breathing valve or volume transducer of the Metamax. Thus, when expired air left the volume transducer of the Metamax, it was led further to a Douglas bag for separate analysis rather than being released to the surrounding air. We also made adapters that allowed us to connect the breathing valve of two Metamaxes in series and to connect the breathing valve of the Vmax to that of the Metamax. Since the instruments were connected in series, the dead spaces differed, and that introduced some systematic errors between the parallel measurements that was removed mathematically as explained below.

Series 3. Six top junior cross-country skiers ran on stepwise increasing treadmill speeds for 5 min at each speed while the O$_2$ uptake was measured simultaneously by the Metamax II and by the Vmax 29. The speeds were chosen to tax 60–100% of the subjects' maximal O$_2$ uptake; the last run for each subject was a standard test for establishing the maximal O$_2$ uptake.

Series 4. Twelve physically active students ran on stepwise increasing treadmill speeds for 5 min at each speed while the O$_2$ uptake was measured simultaneously by the Metamax II and by the Douglas bag technique. Six of these subjects also repeated the experiments on the bicycle ergometer on a separate day. The speeds or powers used were chosen to tax 50–100% of the subjects' maximal O$_2$ uptake; the last run for each subject was a standard test for establishing the maximal O$_2$ uptake.

Series 5. Six moderately trained subjects bicycled at stepwise increasing powers for 5 min at each step while the O$_2$ uptake was measured simultaneously by the Metamax I, the Metamax II, and by the Douglas bag technique. The powers used were chosen to tax 30–90% of the subjects' maximal O$_2$ uptake. For one of the subjects the data on the fractions of O$_2$ and CO$_2$ in the expired air analysed by the Douglas bag technique were lost because of a leaky tube, and for this subject only the data on the lung ventilation are given.

Equipment and analyses

Douglas bag method. The sampling time of expired air collected in the Douglas bags was recorded by stop watches connected to the switch used to start and stop the collection. The fractions of O$_2$ and CO$_2$ in both the inspired and the expired air were measured by an S 3A/I analyser with an N-22M zirconium oxide-type O$_2$ sensor and a CD-3A analyser with a P-61B infrared-type CO$_2$ sensor, respectively (Applied Electrochemistry, Pittsburgh, PA, USA). The volume of the expired air was measured by an S430-A ventilation measure system with a K520–C521 flow transducer (Applied Electrochemistry) while the air temperature was measured simultaneously by a digital thermometer. The air pressure was recorded to the nearest hectopascal by a portable mercury barometer calibrated against high-precision instruments at the Norwegian Institute of Meteorology.

According to Applied Electrochemistry the instrument's zirconium oxide-type O_2 sensor gives a response that is proportional to the logarithm of the fraction or pressure (activity) of O_2 from the ppm-range to 100% O_2. Our separate control experiments using the Scholander technique have verified this for the range 0.1–21% O_2 (not shown). Thus, a possible error in the assumed fraction of O_2 in the room air will affect the readings of the expired and inspired gases proportionally. The errors introduced will thus largely cancel on the difference between the readings of the inspired and the expired air. Moreover, while the fraction of O_2 in outdoor air or the air in a well-ventilated room is accurately know, we have experienced that the fraction of O_2 in purchased calibration gases as examined by the Scholander technique may differ significantly from that reported by the supplier (not shown). The O_2 analyser was therefore calibrated against room air (one-point calibration) since that gives the most reliable values according to our experience.

The CO_2 sensor was undertaken a two-point calibration against room air and against a gas of known fraction of 5–6% CO_2 in N_2 delivered by a commercial supplier. The commercial gases used (AGA, Oslo, Norway) were regularly checked separately by the Scholander technique, but over an 18 yr period we have never received calibration gases where the measured fraction of CO_2 differed significantly from that reported by the supplier. The S430-A ventilation measure system was calibrated using at 7 L calibration syringe (series 4900, Hans Rudolph, Kansas city, MO, USA).

Metamaxes. We used one Metamax I (serial number MMX 43 139 801, Cortex Biophysic, Leipzig, Germany) and two instruments of the type Metamax II (serial number MII 53 229 901 and serial number MII 63 229 901) in our experiments. The instruments record and display the data in 10-s intervals but after a built-in averaging (personal communication with Ralph Henkel, Cortex Biophysic). The Metamax I weighs 1.8 kg. The base unit of the Metamax II weighs only 0.8 kg, and even with batteries the total weight of the instruments is only 1.3 kg. The latter instrument is supplied with a harness, and it can sample and store data for up to 8 h for later downloading to a PC. These properties together with its low weight make the Metamax II potentially suitable for field testing too. However, testing in the field is outside the scope of this study.

The instruments have built-in sensors for O_2, CO_2, a barometer and a thermometer, and it measures the flow of the breathed air by a turbine flow meter attached to the breathing mask or mouthpiece. The expired air sampled is either dried (by $CaCl_2$) or its humidity is equilibrated with that of the surrounding air before the fractions of O_2 and CO_2 is measured; we used only the latter option in our studies. According to the manufacturer the accuracy of the flow meter is better than 1.5%, that of the barometer better than 20 hPa (2%), that of the thermometer better than 0.5 °C, that of the infrared CO_2 sensor better than 0.1 volume percent, and that of the zirconium oxide O_2 sensor better than 0.1 volume percent.

The instruments were used according to the instructions in the manuals. In particular, the instruments were calibrated against a commercial gas of known concentrations of O_2 and CO_2 in the morning before each experiment started. The fractions of CO_2 and O_2 in the gas used to calibrate the two Metamaxes at Sogndal University College were 6.02% and 15.02% according to the manufacturer (AGA). The last Metamax II, the one at Nord-Trøndelag University College, was calibrated by a commercial gas from Sensormedics (Yorba Linda, CA, USA) with nominal values of 4% CO_2 and 16% O_2 (reorder no. 673 666). The instruments were further calibrated against room air, and the concentration of O_2 and CO_2 of room air was read and the flow transducer was calibrated using a 3 L high-precision calibration syringe (Calibration syringe D, Sensormedics) before each experiment on a new subject, see [5] for further details. During the experiments the data collected were immediately transferred to and stored in a PC rather than being stored in the instrument. The barometers were controlled once a year by comparing the reading of the instrument's barometer with the air pressure given by a local meteorologic station or airport after proper correction for differences in altitude between the two sites.

Vmax 29. The measurements by the Vmax 29 (Sensormedics) were carried out at a commercial exercise testing institute (Nord-Trøndelag Regional Centre for Top Sports, Steinkjer, Norway). The O_2- and the CO_2-sensors were calibrated in the morning before the testing by gases of 16% O_2, and 4% CO_2, and thereafter 26% O_2 in N_2, respectively. The Vmax registers the concentrations of O_2 and CO_2 in the ambient air continuously during the tests. The volume transducer of the Vmax was calibrated every second hour with a similar 3 L calibration syringe as that used for the Metamaxes. The Vmax measured the relevant parameters every 30 s.

Examination of the performance of Metamax' built-in barometers. The ambient air pressure was measured to the nearest hectopascal (millibar) by the mercury barometer described above. The Metamaxes record the pressure of the ambient air by built-in barometers, and we have used the reported value rounded to the nearest hectopascal. The pairs of barometric readings have been compared throughout the studies.

Examination of the calibration gases used. The calibration gases used were examined separately by the Scholander technique [3]. In addition, as a separate control the analysers from Applied Electrochemistry were first calibrated as described above. The calibration gases purchased for the Metamaxes were thereafter run through the analysers from Applied Electrochemistry, and the fractions of O_2 and CO_2 in the calibration gas were read off.

Calculations

Calculations of the O_2 uptake from measured parameters. The volume of expired gas sampled in a Douglas bag (V_s), its temperature (T), the collection time (t), and the air pressure (p_B) were measured. The volume was first corrected to standard ambient temperature ($T_0 =$ 25 °C = 298,15 K) and pressure ($p_0 =$ 1000 hPa [6]), dry air (SATPD), using the equation of state and assuming that the expired gas was saturated with vapour at the recorded temperature:

(1) $$V_{e,\text{SATPD}} = V_s\,(p_B - p_{\text{sat}}\text{H}_2\text{O}(T))\,/\,p_0 \cdot (T_0/\text{K})\,/\,(T/\text{K})$$

That entity was then divided by the sampling time, thus giving the flow of expired gas averaged over the sampling period. The saturation pressure of vapour ($p_{\text{sat}}\text{H}_2\text{O}$) for the temperature in question was taken from the following quadratic approximation

(2) $$p_{\text{sat}}\text{H}_2\text{O}(T)\,/\,\text{hPa} = 0.04\,(T/°\text{C})^2 - 0.08\,T/°\text{C} + 9$$

that gives $p_s\text{H}_2\text{O}$ exactly for $T = 10$ °C, $T = 20$ °C and $T = 30$ °C according to data of Tennet [7] and that approximates the saturation pressure well for temperatures between 5 and 35 °C (not shown). For experiments at temperatures below 5 °C, for example for skiing, we recommend using a vapour pressure of 6 hPa (the saturation pressure at 0 °C; no such experiments are given here).

The fractions of O_2 ($x\text{O}_2$) and CO_2 ($x\text{CO}_2$) in the inspired (index i) and expired air (index e) were measured. The fraction of "nitrogen" (that is other components than O_2 and CO_2

in the air, mainly N_2 but also small amounts of Ar and other gases), here symbolically called xN_2, was taken as

(3) $xN_2 = 1 - xO_2 - xCO_2$

While the volume V_e of expired gas was measured, the volume of the inspired air was calculated assuming that the amount of "nitrogen" was constant:

(4) $V_i \, x_iN_2 = V_e \, x_eN_2$ ➔ $V_i = V_e \, x_eN_2 / x_iN_2 = k_{i,e} \, V_e$

where

(5) $k_{i,e} = x_eN_2 / x_iN_2 = (1 - x_eO_2 - x_eCO_2)(1 - x_iO_2 - x_iCO_2)^{-1}$

The fractions of O_2 and CO_2 in both the inspired and the expired air were measured, and the uptake of O_2 and release of CO_2 per volume of expired air were taken as

(6a) $\Delta cO_2 = (k_{i,e} \, x_iO_2 - x_eO_2) / V_mO_2$

(6b) $\Delta cCO_2 = (x_eCO_2 - k_{i,e} \, x_iCO_2) / V_mCO_2$

using molar volumes of O_2 and CO_2 of

(7a) $V_mO_2 = 24.765 \, L_{SATPD} \, mol^{-1}$

(7b) $V_mCO_2 = 24.622 \, L_{SATPD} \, mol^{-1}$

[8]. The O_2 uptake and the respiratory exchange ratio (R-value) were taken as

(8) $nO_2 = \Delta cO_2 \, V_e \, t^{-1}$

(9) $R = \Delta cCO_2 / \Delta cO_2$

The reported lung ventilation was taken from the expired volumes expressed at body temperature (37 °C), ambient pressure and saturated with vapour ($p_{sat}H_2O = 63$ hPa at 37 °C; BTPS), using the equation of state, and for convenience also ΔcO_2 and ΔcCO_2 were expressed per L_{BTPS}.

The Metamaxes report their measured parameters ($x_{i,e}O_2$, $x_{i,e}CO_2$, and V_e) in addition to the O_2 uptake and the respiratory exchange ratio. Using the reported parameters we also cal-

culated the O_2 uptake and the R-value using the same principles as given above for the Douglas bag technique, thus allowing a comparison of the values reported by the Metamaxes to that the equations above give for the reported raw values.

Corrections for different dead spaces. Connecting instruments in series increases the dead space for the instruments closest to the mouth, and that will again increase the O_2 uptake reported by the instruments correspondingly. More specifically, the air inspired during the first part of an inspiration will be from the dead space with a composition like that of the expired air. The fraction of O_2 of inspired air averaged over the whole inspiration is therefore less than that of room air, while the fraction of inspired CO_2 is raised correspondingly. To correct for this error the extra dead space added by the adapters and breathing valves were measured. The tidal volumes for each experiment was taken as the ratio between the lung ventilation and the breathing frequency reported by the Metamaxes, and the ratio, x, of the extra dead space to the tidal volume was calculated. The O_2 uptake reported by each instrument was reduced by multiplying by the factor $(1 - x)$, and this corrected O_2 uptake is reported as the instrument's value for that recording.

Statistics. The data were examined by scatter plots, regression analyses, plots of residuals, and by looking for systematic deviations from the line of identity. We also looked for systematic differences between the subjects. The random variation was taken as the error of regression (scatter around the regression line, $S_{Y|x}$). The data are given as means ± SD or the error of regression. Linear regression were calculated as the geometric mean, thus taking into consideration that errors in both sets of measurements (here: instruments or methods) affect the regression parameters [9].

RESULTS

Alternating measurements by a Metamax and a control method

Comparisons of the Metamax I to the Douglas bag technique (series 1). During the 2 h bicycling there was no systematic difference between the parallel measurements of the O_2 uptake by the Douglas bag technique and by the Metamax I (P = 0.13), but the error of regression (random variation) for the pooled data was 1.6 µmol O_2 s^{-1} kg^{-1} (≈5%; **Fig. 1A**). For two subjects the measurements by the Metamax were systematically higher than those by the

Douglas bag technique, while for another subject the opposite was found. Consequently the error within each subject was 0.8 ± 0.3 μmol s^{-1} kg^{-1} or around half the error of regression of the pooled data.

There were no systematic differences between the lung ventilation (Fig. 1 B) or the O_2 extraction (not shown) measured by the two systems, but the relative errors of regression of 6–10% were larger than that for the measured O_2-uptake. One reason may be that a higher (lower) ventilation during the measurements by the Metamax was at least partly compensated by a lower (higher) O_2-extraction (not shown). The Metamax I reported a higher excretion of CO_2 to the air breathed (mean difference $\pm$ SD = 0.09 ± 0.08 mmol L$_{BTPS}$$^{-1}$, +6%, P 0.001; not shown). Consequently also the reported respiratory gas exchange ratio (R-value) was 0.033 ± 0.048 (+4%) higher for the Metamax I (P < 0.001; Fig. 1C). The difference between the pairs of R-value differed systematically between the subjects (not shown).

Comparisons of the Metamax II to the Vmax (series 2). Trained subjects ran on stepwise increasing treadmill speeds while the O_2 uptake was measured by the Vmax and the Metamax II. The O_2 uptake measured by the Metamax II was 2.8 ± 2.0 μmol s^{-1} kg^{-1} (mean $\pm$ standard error of regression) higher than that reported by the Vmax for the data from all subjects pooled (not shown). The relationship differed systematically between the subjects. Thus, when each subject was examined separately, the error of regression was 0.8 ± 0.6 μmol s^{-1} kg^{-1} ($\approx$40% of that for all data pooled). Moreover, for each instrument the reported O_2 uptake rose linearly by the treadmill speed with little random variation when the data for each subject were examined separately (Metamax II: $S_{Y|x} = 0.6 \pm 0.4$ μmol s^{-1} kg^{-1}; Vmax: $S_{Y|x} = 0.9 \pm 0.6$ μmol s^{-1} kg^{-1}).

The lung ventilation reported by the Metamax II was 5% less than that given by the Vmax, while the Metamax II reported an extraction of O_2 from and excretion of CO_2 to the breathed air $\approx$14% higher than that given by the Vmax (not shown). The reported respiratory exchange ratios, which were largely in the range 0.88–1.00, did not differ systematically between the two instruments, and the mean absolute difference in the R-values was 0.03.

In the experiments described above the O_2 uptake was measured under similar conditions but not on the same expired air by the different instruments. It could be that differences in the breathing resistance between the instruments affected the ventilation or that there have

been some variations within a few minutes in the lung ventilation and the O_2 extraction even during exercise at a constant power.
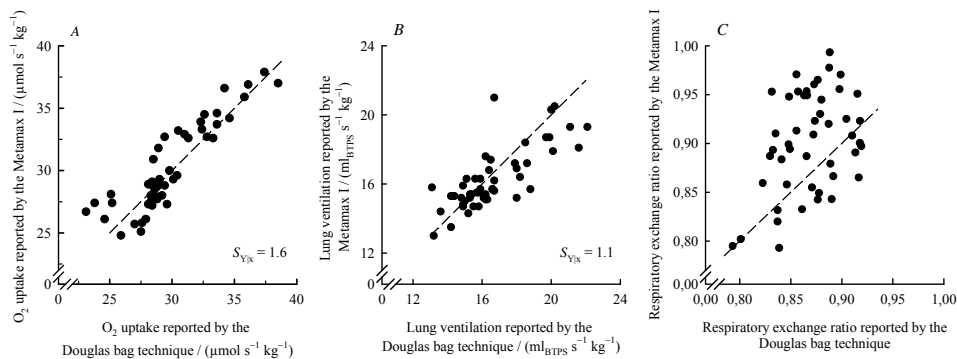

Figure 1. Data reported by the Metamax I versus those of parallel measurements by the Douglas bag technique in series 1. *A*, the O_2 uptake, *B*, the lung ventilation, and *C*, the respiratory exchange ratio. The dashed line is the line of identity, and $S_{Y|x}$ is the error of regression.

Simultaneous measurements by the Metamaxes and control methods

To eliminate possible variations between parallel measurements with different instruments, the flow transducers or breathing valves of two or more instruments were connected in series. Since that approach led to different dead spaces for the different instruments, an effect that will influence the O_2 uptake reported by the instrument, the effect of increased dead space was removed as explained in the methods.
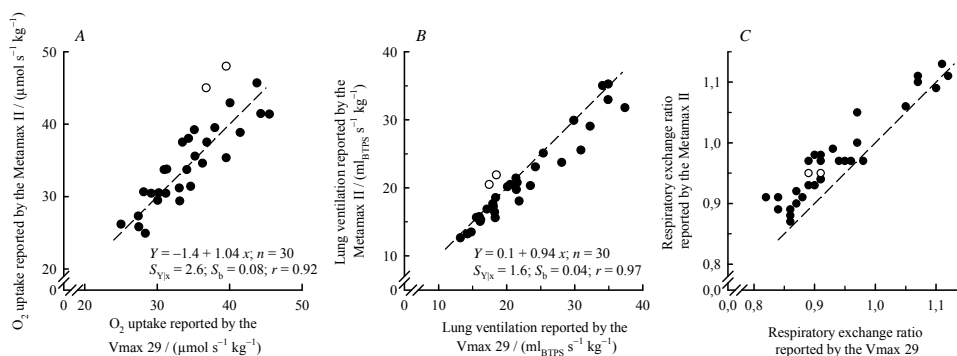

Figure 2. Data reported by a Metamax II versus those of parallel measurements by the Vmax in series 3. *A*, the O_2 uptake, *B*, the lung ventilation, and *C*, the respiratory exchange ratio. The dashed line is the line of identity. Two values were regarded as outliers and thus not included in the regression lines (open symbols).

Comparisons of the Metamax II to the Vmax (series 3). The O_2 uptake did not differ systematically between the two instruments when all measurements were pooled (**Fig. 2A**), but

there was a considerable variation since for two of the subjects all measurements by the Metamax II were above the line of identity while for two other subjects all values were below that line (not shown). Consequently, the error of regression for each subject was 0.8 ± 0.3 μmol s^{-1} kg^{-1}, which is $\approx$30% of the value of 2.6 μmol s^{-1} kg^{-1} when the data from all of the subjects were pooled. The lung ventilation reported by the Metamax II was in average 6% less than that given by the Vmax (Fig. 2B). There were systematic differences between subjects for the lung ventilation too (not shown). The respiratory exchange ratio reported by the Metamax II was 0.038 ± 0.027 higher than that given by the Vmax 29 (P < 0.001; Fig. 2C), and that difference was largely the same for all of the subjects.
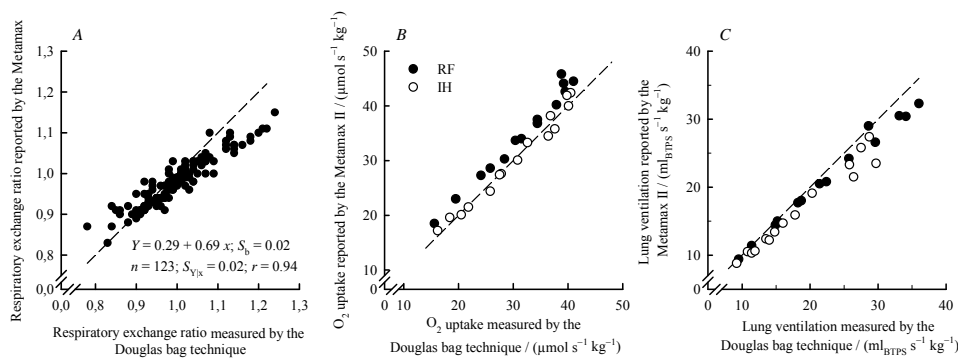

Figure 3. Data reported by a Metamax II versus those of parallel measurements by the Douglas bag technique in series 4. *A*, the respiratory exchange ratio for all subjects pooled, *B*, the O₂ uptake, and *C*, the lung ventilation for subjects RF and IH. The data from these two subjects were chosen to show systematic differences between subjects. The error of regression appeared to be larger for these subjects than for most others. The dashed line is the line of identity.

Comparisons of the Metamax II to the Douglas bag technique (series 4). The O₂ uptake given by the Metamax II was in average 4% higher than that of the Douglas bag technique (P = 0.03; not shown). For lung ventilations up to $\approx$25 ml$_{BTPS}$ s^{-1} kg^{-1} ($\approx$110 L min^{-1}) the value reported by the Metamax II was $\approx$3% less than that given by the Douglas bag method, while for higher ventilations the discrepancy was $\approx$10%. The O₂ extraction from the air breathed was in average 7% higher as reported by the Metamax II than that given by the Douglas bag method, and the excretion of CO_2 was 0.09 ± 0.06 mmol L$_{BTPS}$$^{-1}$ ($\approx$6%) higher as reported by the Metamax II than by the Douglas bag method. For high R-values the respiratory exchange ratio reported by the Metamax deviated clearly from that measured by the Douglas bag technique (**Fig. 3A**).

A further examination of these data showed considerable variations between the subjects, and the error of regression of 0.9 ± 0.4 µmol s^{-1} kg^{-1} when the data for each subject was examined separately, was half of that when the data from all of the subjects were pooled. Two extremes are chosen to illustrate the variations between the subjects (Fig. 3B). For subject IH the data on the O₂ uptake fall around the line of identity, while for the subject RF the data are 3.3 ± 1.3 µmol s^{-1} kg^{-1} above that line ($\approx$10%; P < 0.001). For both subjects the data on the lung ventilation are a little below the line of identity (Fig. 3C).

Comparisons of the Metamax I and the Metamax II to the Douglas bag technique (series 5). The O₂ uptake reported by the two Metamaxes was 9% (Mmx I) and 13% (Mmx II) higher than that given by the Douglas bag technique (**Fig. 4**). The errors of regression of the pooled data were 1.1 (Mmx I) and 1.4 µmol s^{-1} kg^{-1} body mass (Mmx II; $\approx$5%), twice the corresponding errors when the data for each subject were examined separately. The O₂ uptake given by the Metamax II was systematically higher than that given by the Metamax I (P < 0.001).
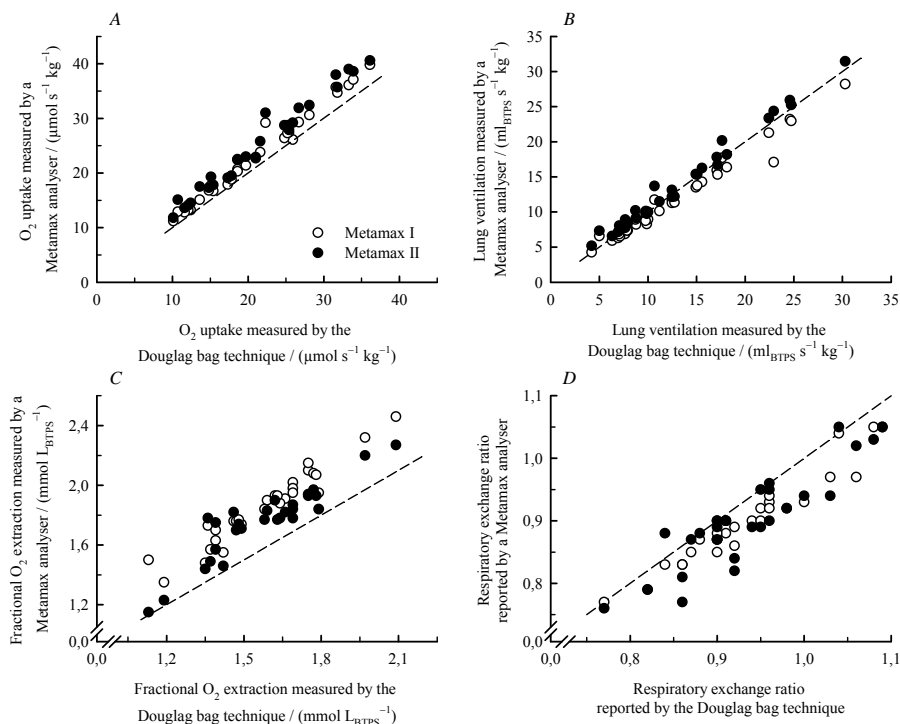

Figure 4. Data reported by the Metamax I (open symbols) and a Metamax II (filled symbols) versus those of parallel measurements by the Douglas bag technique in series 5. *A*, the O₂ uptake, *B*, the lung ventilation, *C*, the fractional O₂ extraction, and *D*, the respiratory exchange ratio. The dashed line is the line of identity.

The lung ventilation reported by the Metamax I was $\approx 7\%$ less than that given by the Douglas bag technique (P < 0.001), while the values for the Metamax II were $\approx 1\%$ higher than those of the Douglas bag technique. The errors of regression were 0.6 (Mmx I) and 0.8 ml_{BTPS} s^{-1} kg^{-1} (Mmx II) or 5–6%. For these experiments there were no sign of a levelling off at high ventilations for the Metamaxes, not even for a subject breathing 30 ml_{BTPS} s^{-1} kg^{-1} (147 L_{BTPS} min^{-1}). Both Metamaxes reported a higher extraction of O_2 (Fig. 4 C) and excretion of CO_2 (not shown) per volume of expired air than the Douglas bag technique did, and the difference was larger for the Metamax I than for the Metamax II. The random variation in these differences was 0.06–0.11 mmol L_{BTPS}^{-1} or $\approx 5\%$.

The respiratory exchange ratio reported by the Metamaxes was in average 0.02–0.03 less than that given by the Douglas bag technique (P < 0.01; Fig. 4D). The reported R-value did not differ significantly between the two Metamaxes.
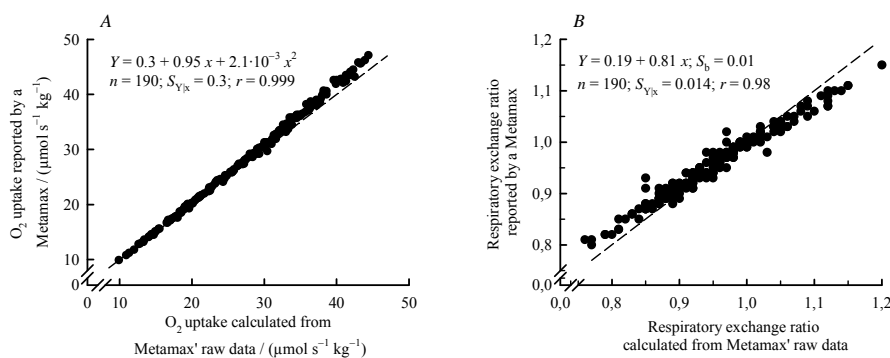

Figure 5. Data reported by the Metamaxes versus those calculated from the instruments' reported raw data. *A*, the O_2 uptake, *B*, the respiratory exchange ratio. The dashed line is the line of identity. The data are from 17 subjects in series 4 and 5.

Comparison of the O_2 uptake reported by the Metamaxes to that calculated from the raw data. The O_2 uptake reported by the instruments was higher than that calculated from the raw data (**Fig. 5A**). For O_2 uptakes less than ≈ 20 μmol s^{-1} kg^{-1} the difference was ≈ 0.2 μmol s^{-1} kg^{-1} (1% of the O_2 uptake), for values ≈ 30 μmol s^{-1} kg^{-1} the difference was ≈ 1 μmol s^{-1} kg^{-1}, while for O_2 uptakes above ≈ 40 μmol s^{-1} kg^{-1} the reported value was ≈ 2 μmol s^{-1} kg^{-1} higher than that calculated from the reported raw data ($\geq 5\%$). The observed difference thus rose roughly quadratically by the O_2 uptake. This relationship did not differ between the three different Metamaxes examined (not shown). The respiratory exchange ratio reported by the

instruments differed systematically from that calculated from the reported raw data (Fig. 5B), and the calculated R-values agreed better with those given by the Douglas bag technique than those reported by the instruments did (not shown).

Examination of Metamaxes' built-in barometers. In most cases the barometric pressure read by the Metamaxes agreed with that of a mercury barometer. Apart from one series of experiments we never saw a difference larger than 1 hPa. In series 5 the reported barometric pressure of the Metamax I used was 1–3 hPa less than that of the mercury barometer, while the values of the Metamax II used were 1–4 hPa higher than that of the mercury barometer.

Control of the calibration gases used. The gas used to calibrate the CO_2-analyser from Applied Electrochemistry used for our Douglas bag technique was first examined twice, more than one year apart, by the supplier using high-precision gas chromatography. The fraction of CO_2 was reported to be 5.10% and 5.13% respectively. That gas was also measured by the Scholander technique [3], and the fraction of CO_2 was found to be 5.108 ± 0.009%.

The fraction of CO_2 in the gas used to calibrate the two Metamaxes at Sogndal University College was 5.947 ± 0.005% according to our analyses by the Scholander technique while the nominal value given by the supplier was 6.02%. The corresponding values for O_2 were 14.933 ± 0.022% (Scholander technique) and 15.02% (nominal value). The last Metamax II (the one at Nord-Trøndelag University College) was calibrated by a commercial gas from Sensormedics with nominal values of 4% CO_2 and 16% O_2 (reorder no. 673 666). The analyses by the Scholander technique gave 3.903 ± 0.025% CO_2 and 16.097 ± 0.030% O_2 for this gas.

DISCUSSION

The O_2 uptake given by the Metamaxes rose linearly by that given by the control methods. For each subject the O_2 uptake also rose linearly by the power and with little random variation. The O_2 uptake given by the Metamaxes was in most cases larger than that given by the control method, and the relationships differed systematically between the subjects. The respiratory exchange ratio reported by the Metamaxes did not agree well with that of the

Douglas bag technique. The O_2 uptake and the respiratory exchange ratio reported by the instruments differed from those calculated from the instruments' raw data.

The O_2 uptake reported by the Metamaxes rose linearly by the value of the control methods. When the data from several subjects were pooled, there was a random variation of ≈5%. This appeared largely to be caused by systematic differences between the subjects since the residual variation (error of regression, scatter around the regression line) was less than 1 μmol s^{-1} kg^{-1} or ≈2% when each subject was examined separately. Moreover, the O_2 uptake reported by the Metamax rose linearly by the bicycle power or treadmill speed, and the error of regression was ≈0.5 μmol s^{-1} kg^{-1} or ≈2%. That value is similar to what we have found for the Douglas bag technique during exercise studies (unpublished data from [1, 10, 11] see also [12, 13]). A further examination of the Douglas bag technique has shown that the analytical error is less than 0.5% but that the biological variation is 1–3% (Medbø, unpublished results). The data from this study thus suggest that the random error of the Metamax when each subject is examined separately is no larger than the biological variation. Moreover, the precision of the Metamaxes is in this respect as good as that of the Douglas bag technique, at least when all measurements are carried out on the same day.

While the data reported by a Metamax showed little random variation for each subject, there were systematic differences between the subjects. Moreover, for most of the studies the values reported by the Metamaxes were higher than those of the control methods. The lung ventilations reported by the Metamaxes were usually a few percent less than those reported by the control methods, while the extraction of O_2 as reported by the Metamaxes was in most cases systematically larger than those of the control methods. In addition, for moderate to high values of the O_2 uptake the O_2 uptake reported by the Metamaxes were higher than those calculated from its raw data. This latter finding means that the Metamax does not calculate the O_2 uptake only according to equations 1–9 on the reported raw data.

The respiratory exchange ratio as reported by the Metamaxes agreed fairly well with that of the control methods only in the range 0.9–1.0. When the R-value was above 1.0, the Metamaxes underestimated the true value. For R-values ≈0.8 or less our data suggest that the Metamaxes overestimates the true value. Moreover, the R-value calculated from the reported raw data differed from that reported by the instruments, and the calculated value agreed better

with those of the control methods than the instruments' reported values did. Thus, this study suggests that while the O$_2$ uptake reported by Metamax is quite reliable, the R-value is not. Moreover, our data suggest that the R-values of the Metamaxes may be improved if equations 1–9 are used on the reported raw data.

Barometers. The Metamaxes have a built-in barometer, and according to the manufacturer the barometer is accurate within 20 hPa or 2%. Our data suggests that it is far better than that, and if properly calibrated, the accuracy of the barometer seems to be at least one order of magnitude better than what suggested by the manufacturer. An error in the recorded pressure of 1 hPa (0.1%), which in our experiments could be due to round-off errors, will affect the final result by only 0.1%. This is of no importance in physiologic or medical experiments. In our experience a control of the reading of the barometers once a year is enough.

Calibration gases. We examined the calibration gases used for the Metamaxes by the Scholander technique, and the results of those analyses differed systematically from the values provided by the suppliers. Using the gas from Sensormedics as an example, if an instrument is calibrated by this gas, the reported O$_2$ uptake will be 2% too large, and the reported CO$_2$-release will be 2.5% too low. The reported respiratory exchange ratio will be $\approx$4.5% too low. The inaccuracies in the calibration gas used for the other Metamaxes were of similar magnitude but in the opposite direction. Thus, the quality of the calibration gases used affects the results, and better gases and routines may be sought. For example, as explained in the methods, using room air improved our measurements of the fraction of O$_2$ measured by the zirconium oxide-type analyser from Applied Electrochemistry used for our Douglas bag technique. The Metamaxes also measure the fractions of O$_2$ using a zirconium oxide cell. It may be that using one-point calibration against room air rather than a two-point calibration may improve the performance of that analyser too. For calibration of the CO$_2$ analysers a high-precision gas of 5–6% of CO$_2$ rather than a gas of $\approx$4% CO$_2$ of poorer precision should improve the results. However, inaccurate calibration gases can only partly explain the errors we saw in the reported gas exchange ratio.

We have in this study compared the results of the Metamaxes during exercise with those of the Douglas bag technique. The latter method is the gold standard in measurements of the O$_2$ uptake to which other methods should be compared, and that approach has been used in several studies comparing different instruments [14–18]. We also compared the results of the

Metamax to those of the Vmax, a commercial and fully automatic instrument. Others have used a corresponding approach [19–27]. Including that part of our experiments in this study thus makes comparison with those latter studies more easy. In addition, since the Metamaxes showed higher values and systematic differences between subjects whether compared with the Douglas bag technique or the Vmax, it is conceivable that these deviations are due to the Metamaxes and not to the two control methods used. However, it should be noted that we have not compared the performance of the Vmax to the Douglas bag technique directly.

Most others who examined two different instruments for measuring the O_2 uptake, have done so by doing alternating measurements by two instruments under similar conditions [15, 17, 19, 20, 21–23, 25, 27]. We used that approach in the first part of the study. In further experiments we connected the breathing valves of two or more instruments in series. The latter approach, which has been used in four former studies too [14, 16, 18, 26], allows measurements of the O_2 uptake on the same expired air by two or more instruments. However, that approach introduces systematic errors because the dead space is increased. That problem that has not been addressed by other as far as we know. One group who tried that approach, dropped it and used alternating measurements because they observed that the serial connection influenced the readings of the K4 studied [17]. The errors caused by serial connections are readily removed mathematically, and that correction was important. For example, in series 5 there were no difference in the output of the two Metamaxes when the Metamax I was closest to the mouth, while the difference was ≈10% when the Metamax II was closest to the mouth.

Our data suggest that for studies where the conditions can be kept constant for several minutes or when the result can be reproduced precisely from one day to the next, alternating measurements by two different instruments may be an adequate approach. On the other hand, during intense, short-lasting exercise the conditions change continuously, and for such studies only simultaneous measurements by two or more instruments will be adequate. For example, our data suggest that the Metamaxes do not report the respiratory exchange ratio properly. To test that further it may be necessary to carry out studies of high intensity, anaerobic types of exercise with hyperventilation and where it is known that the R-values rises above one during the exercise and in the early recovery. For such studies only the approach of serial connection of two or more breathing valves will be adequate.

Examinations of other portable O$_2$-analysers. There are several other commercial portable instruments for measuring the O$_2$ uptake that have been examined by others [14–17, 19–24, 26]. The Metamaxes in our study seem to perform as good as or better than the other portable analysers examined, perhaps with an exception for the K4 from Cosmed [21]. For example, the random variation in the data from the Metamaxes appears less than that of other instruments. Moreover, most portable analysers seem to have problems with measuring the R-value reliably.

A common conclusion in all of the studies referred to above is that the instruments examined "performed well", but none of the studies give criteria for a good performance or minimum requirements for being acceptable. No one has examined the respiratory exchange ratio over a wide range, nor have they to our knowledge addressed possible individual variation in the O$_2$ uptake or other measures.

Statistical analyses. We have analysed our data statistically by scatter plots and looked for non-linear effects and possible systematic deviations from the line of identity. We have further looked for possible differences between subjects and used the error of regression as a measure of the random variation, and we have quantified these entities. That is in line with recommendations of textbooks of statistics [f. ex. 28–30] and of leading statisticians in biomedicine [31, 32]. Leading experts in biomedical statistics regard use of correlation coefficients and related measures, as used by some (f. ex. [33]), as inadequate statistics [31, 32]. More recently the approach of Bland-Altman for testing two possibly equivalent techniques have been used when a new instrument is compared with an established control or calibration method [15–18, 21, 23, 24, 27]. That approach is far better than using correlation methods, but as clearly pointed out by Bland and Altman, their approach is recommended only if a control method (or calibration method as they call it) is not available [31, 32]. When the performance of a new and possibly uncertain method is to be compared with that of a well established and examined control method (f. ex. the Douglas bag technique), the statistical analyses we used should be chosen. However, although the Bland-Altman approach used by others is somewhat more complicated, it has not led to incorrect conclusions in the studies referred to above.

Conclusions

The random variation in the measurements of the O_2 uptake by the Metamaxes was as good as for the Douglas bag technique when each subjects was examined separately. There were some systematic errors in the O_2 uptake reported by the Metamaxes, and there were some variations between the subjects. The respiratory exchange ratio was not well measured by the instruments.

ACKNOWLEDGEMENTS

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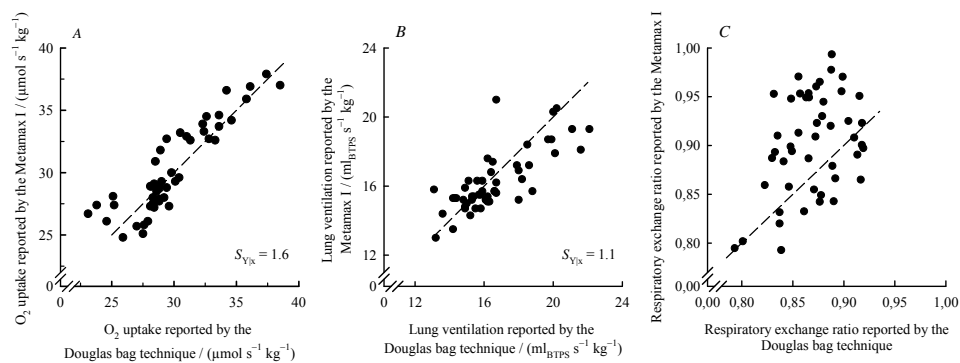
FIGURE LEGENDS

Figure 1

Figure 1. Data reported by the Metamax I versus those of parallel measurements by the Douglas bag technique in series 1. *A*, the O_2 uptake, *B*, the lung ventilation, and *C*, the respiratory exchange ratio. The dashed line is the line of identity, and $S_{Y|x}$ is the error of regression.
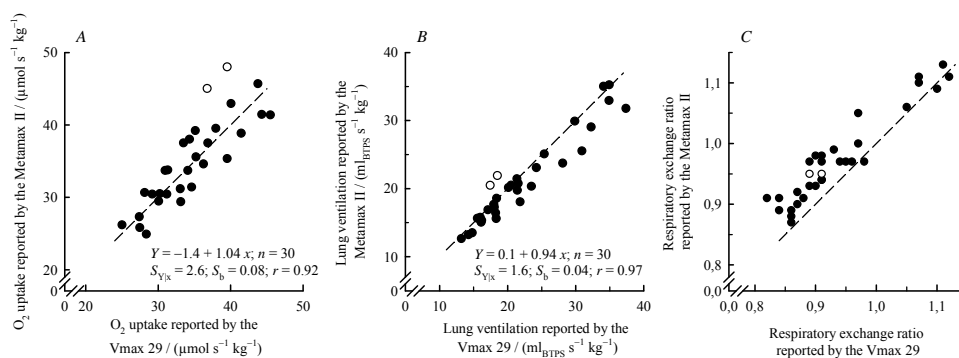
Figure 2

Figure 2. Data reported by a Metamax II versus those of parallel measurements by the Vmax in series 3. *A*, the O_2 uptake, *B*, the lung ventilation, and *C*, the respiratory exchange ratio. The dashed line

is the line of identity. Two values were regarded as outliers and thus not included in the regression lines (open symbols).
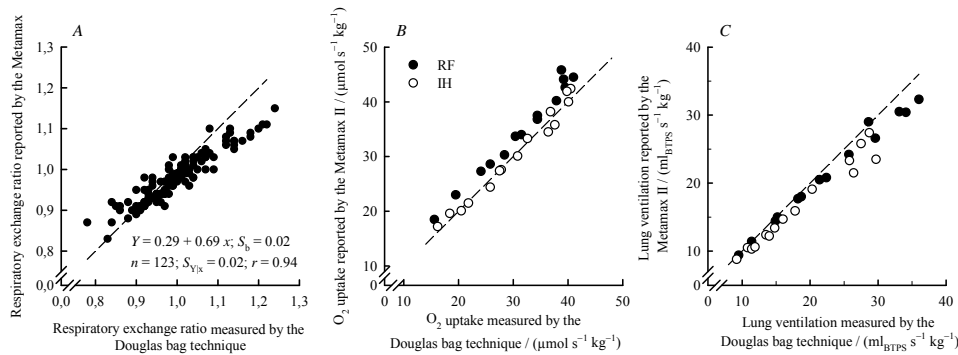
Figure 3

Figure 3. Data reported by a Metamax II versus those of parallel measurements by the Douglas bag technique in series 4. *A*, the respiratory exchange ratio for all subjects pooled, *B*, the O₂ uptake, and *C*, the lung ventilation for subjects RF and IH. The data from these two subjects were chosen to show systematic differences between subjects. The error of regression appeared to be larger for these subjects than for most others. The dashed line is the line of identity.
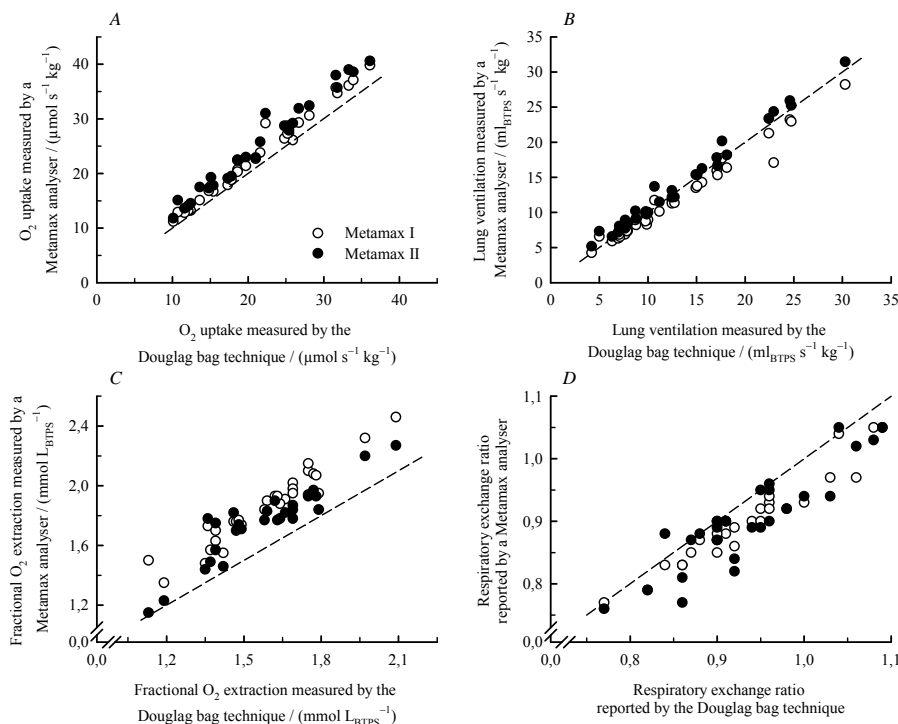

Figure 4. Data reported by the Metamax I (open symbols) and a Metamax II (filled symbols) versus those of parallel measurements by the Douglas bag technique in series 5. *A*, the O₂ uptake, *B*, the lung ventilation, *C*, the fractional O₂ extraction, and *D*, the respiratory exchange ratio. The dashed line is the line of identity.
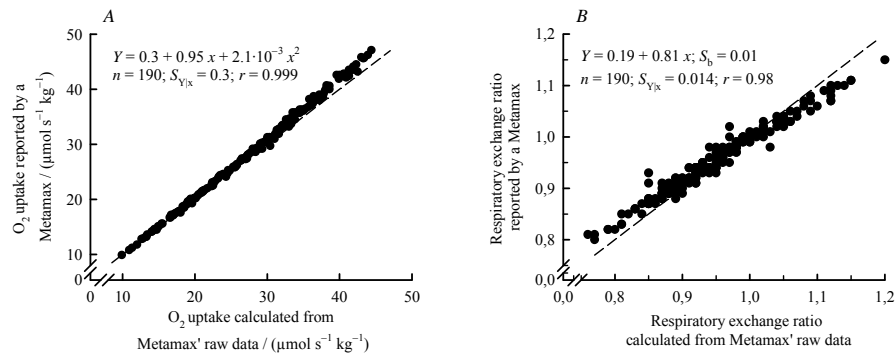
Figure 5

Figure 5. Data reported by the Metamaxes versus those calculated from the instruments' reported raw data. *A*, the O_2 uptake, *B*, the respiratory exchange ratio. The dashed line is the line of identity. The data are from 17 subjects in series 4 and 5.