Determinants of sedentary 24-h energy expenditure: equations for energy prescription and adjustment in a respiratory chamber1–3

Yan Y Lam, Leanne M Redman, Steven R Smith, George A Bray, Frank L Greenway, Darcy Johannsen, and Eric Ravussin

ABSTRACT

Background: Achieving energy balance is critical for the interpretation of results obtained in respiratory chambers. However, 24-h energy expenditure (24EE) predictions based on estimated resting metabolic rate and physical activity level are often inaccurate and imprecise.

Objective: We aimed to develop and validate equations to better achieve energy balance in a respiratory chamber by adding or subtracting food items.

Design: By using a randomized data set with measures of 24EE (n = 241) performed at the Pennington Biomedical Research Center, we developed equations to predict 24EE from anthropometric, demographic, and body composition variables before and at 3 and 7 h into the chamber measurement. The equations were tested on an independent data set (n = 240) and compared with published predictive equations.

Results: By using anthropometric and demographic variables, the equation was as follows: 24EE (kcal/d) = 11.6 [weight (kg)] + 8.03 [height (cm)] – 3.45 [age (y)] + 217 (male) – 52 (African American) – 235. The mean prediction error was –9 ± 155 kcal/d (2046 ± 305 compared with 2055 ± 343 kcal/d for measured 24EE; P = 0.36). The prediction achieved a precision of ±10% of measured 24EE in 83% of the participants. Energy prescription was then refined by equations with the use of energy expenditure values after 3 h, 7 h, or both into the chamber study. These later equations improved the precision (±10% of measured 24EE) to 92% (P = 0.003) and 96% (P < 0.0001) of the participants at 3 and 7 h, respectively. Body composition did not improve 24EE predictions.

Conclusions: We showed the use of a set of equations to prescribe and adjust energy intake to achieve energy balance in respiratory chambers over 24 h. These equations may be used in most respiratory chambers and modified to accommodate exercise or specific feeding protocols. Some of the data used in this study were from trials registered at clinicaltrials.gov as NCT00493701, NCT00099151, NCT00565149, NCT01672632, NCT00945633, NCT00829140, NCT00936130, NCT01275235, NCT01898949, NCT01775163, and NCT00943215. Am J Clin Nutr 2014;99:834–42.

INTRODUCTION

Respiratory chambers, or whole-room indirect calorimeters, were first described >100 y ago (1) and modern versions were described 3 decades ago (2–4). These chambers allow measurement of energy expenditure (EE) in humans and remain one of the most important tools to investigate all of the aspects of energy metabolism in sedentary conditions or with prescription of exercise. The continuous measurement of respiratory gas exchange (ie, oxygen consumption and carbon dioxide production) throughout the study and nitrogen excretion allows investigators to capture dynamic changes in energy metabolism, which provides critical insights into the interindividual variability of energy and macronutrient balances. Respiratory chambers are gaining importance in the clinical research of energy metabolism, as evidenced by the rapid increase in the number of chambers from 2 to 3 in the early 1980s to >30 at the present time worldwide.

One of the most important and yet very challenging aspects of a respiratory chamber study is to provide participants with the right amount of energy to consume over the day. In over- or underfeeding studies, the target energy surplus/deficit is mainly achieved by adjusting food intake. In studies of substrate utilization in which conditions of achieving energy balance (ie, energy intake equal to EE) are key for interpretation of findings, one has to essentially “guess” the energy intake to provide to volunteers that will eventually match the EE being measured. Fluctuations in energy balance modify fuel selection with a preference of carbohydrate over fat oxidation in response to overfeeding, or the contrary in underfeeding (5, 6). Substrate oxidation is calculated from the rates of gaseous exchange and nitrogen excretion with the use of standard equations (7). These calculations, however, become invalid when certain metabolic processes (eg, lipogenesis, gluconeogenesis, ketone body production, and lactate metabolism) are activated as a consequence of significant deviations from energy balance (8). In a respiratory chamber study, energy prescription is based on the calculation of

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2 Supported by NORC center grant P30DK072476; research grants R01 AG030226, R01 DK60412, U01 AG20478, and U01 AG022132 from the NIH; research grants 2005-34323-15741 and 58-6435-5-071 from the USDA; and pharmaceutical companies (Takeda Pharmaceuticals of North America, Arena Pharmaceuticals, Inc, Ethicon Endo Surgery, Beartown Pharma, Inc, and Archer-Daniel–Midland).

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4 Abbreviations used: AEE, activity energy expenditure; DXA, dual-energy X-ray absorptiometry; EE, energy expenditure; FFM, fat-free mass; FM, fat mass; PAL, physical activity level; RMR, resting metabolic rate; 24EE, 24-h energy expenditure.

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daily energy requirement, which in most cases involves an estimation of both the resting metabolic rate (RMR) and physical activity level (PAL) of the participants. The accuracy and precision of the existing predictive equations for RMR vary considerably (9). In addition, the use of conventional activity factors (10) is likely to overestimate (or sometimes underestimate) the energy requirement because physical activity is not only limited in the confined setting of the chamber but also highly variable in people.

The aim of the present study was to develop ways to achieve energy balance more effectively in a respiratory chamber. We generated equations to predict 24-h EE (24EE) before a study and 2) predict 24EE by using the first 3 and 7 h of measurement in the respiratory chamber to allow adjustment of food intake for the remainder of the 24-h study.

SUBJECTS AND METHODS

Subjects

This study was an internal analysis of respiratory chamber studies performed at the Pennington Biomedical Research Center between March 1998 and May 2013. Protocols of all studies conducted during the selected period were obtained from the principal investigators. Studies were included in the current work if they had measurements of 24EE in a respiratory chamber and body composition by dual-energy X-ray absorptiometry (DXA) as part of the baseline testing and there was no mandatory exercise component in the protocol. Data from the eligible studies were obtained directly from the principal investigators. Subjects were excluded if they 1) had diabetes with a fasting blood glucose concentration >126 mg/dL (11), 2) were of an ethnicity other than white or African American, or 3) were not in “reasonable” energy balance [energy intake ± 20% of 24EE (12)] during the chamber stay. All studies were approved by the Institutional Review Board of the Pennington Biomedical Research Center, and written informed consent was obtained from all participants. All procedures were performed in accordance with the ethical standards of the Institutional Review Board of the Pennington Biomedical Research Center.

Anthropometric and body composition measurements

Weight, height, and waist circumference were measured with the use of standardized procedures. Body surface area was calculated by using the Du Bois and Du Bois equation (13). Whole-body percentage of body fat was measured by DXA. Three whole-body scanners were used across the studies [Lunar iDXA (GE Health Care) and Hologic QDR 2000 and QDR 4500A]. Conversion factors derived from internal validation analyses were used to make all measurements comparable to those taken with the Hologic QDR 4500A. Fat mass (FM) and fat-free mass (FFM) were calculated from the percentage body fat and metabolic weight measured on the morning of the chamber stay.

Indirect calorimetry

The 2 respiratory chambers at the Pennington Biomedical Research Center had a total volume of 27 m³ (3 × 3.7 × 2.4 m). The setting and measurement procedures of the chamber have previously been described (14). Briefly, concentrations of oxygen and carbon dioxide in the chamber were measured throughout the chamber stay, from which oxygen consumption, carbon dioxide production, and therefore EE and substrate oxidation (with the use of urea nitrogen excretion rate) were calculated every 10 s and the values were plotted at 10-min intervals. Microwave motion detectors were used to determine the percentage of time during which the subject was moving (Activity %). The Pennington Biomedical Standard Operating Procedure requires that participants enter the chamber at 0800 after an overnight fast and standardizes meal times and lights out. There was no mandatory activity protocol in the selected studies. Subjects could move freely while they were in the chamber, but they were not allowed to perform any exercise such as sit-ups or push-ups. For each subject, the daily energy requirement for weight maintenance was calculated by using predetermined equations (15–17), equations from personal communications (R Rising, 2002: RMR [kcal/d] = 538 + 25 [FFM (kg)] + 5.5 [FM (kg)]; PA Tataranni, 2002: 24EE [kcal/d] = 1294–7 [age (y)] + 33 [FFM (kg)] + 3 [FM (kg)] + 92 [male], or RMR measured by a metabolic cart (18) with activity factors of 1.3 or 1.4. The exact calculations of daily energy requirements were specific to each study as per the instructions from the principal investigators. The food (50–60% carbohydrate, 15% protein, and 25–35% fat as a percentage of total energy) was provided over 3 main meals: breakfast at 0900, lunch at 1330, and dinner and a snack at 1900, with a percentage of daily energy intake distribution of 25%/35%/40% over the 3 main meals. Calories spent during the first 3 and 7 h of the chamber stay were used to predict the final 24EE (14). When the prescribed energy intake fell outside ±100 kcal of the predicted 24EE after 7 h, the amount of calories provided in the dinner meal was adjusted (added or subtracted) in units of 100 kcal in an attempt to achieve energy balance. The participants exited the chamber at 0715 the next day. EE over the duration of the stay was extrapolated to obtain 24EE. PAL was defined as the ratio of 24EE to sleeping metabolic rate (SMR), whereas activity energy expenditure (AEE) was calculated as 24EE – (SMR + 0.1 × 24EE) (19).

Statistical analyses

Values are expressed as means ± SDs. Independent-samples t tests were used to determine the impact of sex and ethnicity. The data set was randomized such that half of the sample (test group) was entered in a stepwise linear regression model to develop equations for predicting 24EE. Variables were entered one by one, and those which significantly decreased the residual variance (root of the mean square errors) were retained. The final model included the panel of predictors that gave the maximal coefficient of determination (R²). Two models were derived for 24EE prediction by using 1) body composition and demographic variables and 2) anthropometric and demographic variables. The equations derived from the test group were then applied to the other half of the data set (validation group). Predicted EE and mean error were calculated. Paired-samples t test, Pearson correlation, and Bland-Altman analyses (20) were used to assess the relation between the predicted and measured 24EE. Prediction accuracy was defined as the percentage of the study population in which 24EE was predicted to within ±10% of the measured value (9) and was compared by using a chi-square test. Statistical analyses were performed by using the IBM SPSS Statistics.
(version 21; IBM Corporation). Significance was accepted at $P < 0.05$.

RESULTS

Study selection and characteristics

Between March 1998 and May 2013, 20 studies (with a total of 674 subjects) were conducted at the Pennington Biomedical Research Center that had measurements of 24EE in a respiratory chamber and body composition by DXA as part of the baseline testing. Subjects who were required to exercise during the chamber stay were excluded ($n = 111$). Four subjects were excluded because of invalid respiratory chamber data. A further 78 were excluded because of the following factors: 1) 13 had diabetes, 2) 30 were of an ethnicity other than white or African American, and 3) 35 were not in “reasonable” energy balance (energy intake ± 20% of 24EE) during the chamber stay. A total of 481 subjects entered the final analysis (Table 1). Because of protocol differences, not all subjects had data on the entire panel of measurements. The effective sample size for each variable is listed in Table 1 and again specified in each analysis in Results.

Equations for energy prescription

On average, the prescription of energy intake before the chamber stay was 2311 kcal, which was 12% (241 ± 345 kcal; $P < 0.0001$) higher than 24EE. After adjustments at 7 h into the measurement, the total energy intake was revised to 2238 ± 377 kcal and remained 8% (167 ± 133 kcal; $P < 0.001$) higher than 24EE (Figure 1). We used half of the data set (a random sample composed of 241 individuals) to develop equations to predict 24EE (Table 2; Energy prescription) as follows:

\[
\text{Body Composition Model: } 24\text{EE (kcal/d)} = 26.2([\text{FFM(kg)}]) +5.2([\text{FM(kg)}]) -2.32([\text{age(y)}]) -96([\text{African American}]) + 546
\]

\[
\text{Anthropometric Model: } 24\text{EE(kcal/d)} = 11.6([\text{weight(kg)}]) +8.03([\text{height(cm)}]) -3.45([\text{age(y)}]) +217([\text{male}]) -52([\text{African American}]) -235
\]

The above equations (body composition and anthropometric) were applied to the other half of the data set (validation group), and the agreement between the predicted and measured 24EE is shown in Figure 2. Predictions by body composition and anthropometric variables were both highly correlated with measured 24EE with Pearson coefficients of 0.896 (Figure 2A) and 0.893 (Figure 2B), respectively. At the group level, the mean prediction error of the body composition model was $-4 ± 152$ kcal/d and that of the anthropometric model was $-9 ± 155$ kcal/d. At the individual level, the 2 models had similar prediction accuracy (86% for the body composition model compared with 83% for the anthropometric model in Table 3; $P = 0.45$). Body surface area has also been implicated as an important determinant of EE (21). We modified our anthropometric model to include body surface area as a predictor, but that did not improve the 24EE prediction (mean error = $-9 ± 156$ kcal/d; prediction accuracy = 82%). The agreement between the predicted and measured 24EE is shown in Bland-Altman plots (Figures 2, C and D). The negative slope of the regression lines ($r = -0.148, P = 0.02$ for body composition; $r = -0.253, P < 0.001$ for anthropometry) and visual inspection of the distribution of plots categorized by activity level suggest that the equations tend to

<p>| TABLE 1 |
| Physical characteristics and measurements of energy metabolism in study participants |
| Sex | Ethnicity |</p>
<table>
<thead>
<tr>
<th>All ($n = 481$)</th>
<th>Male ($n = 211$)</th>
<th>Female ($n = 270$)</th>
<th>White ($n = 327$)</th>
<th>African American ($n = 154$)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (y)</td>
<td>481 34.5 ± 11.9</td>
<td>211 31.0 ± 11.2$^2$</td>
<td>270 37.1 ± 11.7</td>
<td>327 34.8 ± 12.5</td>
</tr>
<tr>
<td>Height (cm)</td>
<td>481 170.0 ± 9.4</td>
<td>211 177.8 ± 6.6$^2$</td>
<td>270 163.8 ± 6.1</td>
<td>327 170.8 ± 9.3</td>
</tr>
<tr>
<td>Weight (kg)</td>
<td>481 84.5 ± 20.2</td>
<td>211 88.2 ± 18.4$^2$</td>
<td>270 81.6 ± 21.0</td>
<td>327 83.3 ± 18.2</td>
</tr>
<tr>
<td>BMI (kg/m$^2$)</td>
<td>481 29.3 ± 7.0</td>
<td>211 27.9 ± 5.9$^2$</td>
<td>270 30.4 ± 7.6</td>
<td>327 28.6 ± 6.3$^3$</td>
</tr>
<tr>
<td>Waist circumference (cm)</td>
<td>477 92.0 ± 17.2</td>
<td>209 92.9 ± 14.2$^2$</td>
<td>268 91.3 ± 19.2</td>
<td>324 91.9 ± 17.2</td>
</tr>
<tr>
<td>Body surface area (m$^2$)</td>
<td>481 1.95 ± 0.22</td>
<td>211 2.05 ± 0.19$^2$</td>
<td>270 1.87 ± 0.21</td>
<td>327 1.95 ± 0.21</td>
</tr>
<tr>
<td>Body fat (%)</td>
<td>477 31.4 ± 10.6</td>
<td>210 33.0 ± 7.3$^2$</td>
<td>267 38.1 ± 7.6</td>
<td>326 30.8 ± 10.9</td>
</tr>
<tr>
<td>Fat-free mass (kg)</td>
<td>477 56.7 ± 11.8</td>
<td>210 66.6 ± 8.9$^2$</td>
<td>267 48.9 ± 7.0</td>
<td>326 56.9 ± 11.7</td>
</tr>
<tr>
<td>Fat mass (kg)</td>
<td>477 27.2 ± 13.5</td>
<td>210 21.0 ± 10.9$^3$</td>
<td>267 32.0 ± 13.5</td>
<td>326 26.5 ± 13.7</td>
</tr>
<tr>
<td>Fasting glucose (mg/dL)</td>
<td>480 92.7 ± 9.4</td>
<td>210 94.4 ± 8.5$^2$</td>
<td>270 91.3 ± 9.8</td>
<td>326 92.6 ± 9.2</td>
</tr>
<tr>
<td>24EE (kcal/d)</td>
<td>481 2071 ± 357</td>
<td>211 2316 ± 301$^2$</td>
<td>270 1879 ± 271</td>
<td>327 2093 ± 347</td>
</tr>
<tr>
<td>SMR (kcal/d)</td>
<td>481 1650 ± 281</td>
<td>211 1829 ± 230$^2$</td>
<td>270 1509 ± 225</td>
<td>327 1667 ± 266</td>
</tr>
<tr>
<td>Activity (%$^4$)</td>
<td>481 17.1 ± 10.0</td>
<td>211 18.9 ± 10.4$^2$</td>
<td>270 15.7 ± 9.5</td>
<td>327 17.8 ± 10.8$^3$</td>
</tr>
<tr>
<td>PAL (%)</td>
<td>481 1.26 ± 0.08</td>
<td>211 1.27 ± 0.08$^2$</td>
<td>270 1.25 ± 0.08</td>
<td>327 1.26 ± 0.08</td>
</tr>
<tr>
<td>AEE (kcal/d)</td>
<td>481 214 ± 127</td>
<td>211 255 ± 140$^2$</td>
<td>270 182 ± 105</td>
<td>327 216 ± 122</td>
</tr>
</tbody>
</table>

$^1$Values are means ± SDs unless otherwise indicated. AEE, activity energy expenditure; PAL, physical activity level; SMR, sleeping metabolic rate; 24EE, 24-h energy expenditure.

$^2,^3$Significantly different between sexes or ethnic groups (independent-samples $t$ test): $^2P < 0.001$, $^3P < 0.01$, $^4P < 0.05$.

$^4$Percentage of time during which the subject was moving in the respiratory chamber.
underestimate 24EE in subjects who had a high energy requirement and/or were more active during the chamber stay.

Validation of energy prescription equations

We validated the body composition model by comparing the 24EE prediction to that estimated by the Muller et al. (22) equation, which used body composition to predict RMR. An activity factor of 1.3 was used to account for physical activity during the chamber stay (23). The 24EE predicted by the Muller equation (2115 ± 6265 kcal/d) was not different from that estimated by our body composition model (2049 ± 320 kcal/d; P = 0.06) and the measured 24EE (2055 ± 343 kcal/d; P = 0.11). Both the body composition model and the Muller equation offered good fits with r = 0.896 and 0.887, respectively (Figure 3A). The body composition model had a smaller mean error (2446 kcal/d compared with 63616 kcal/d for Muller; P < 0.0001) and a higher prediction accuracy (86% compared with 75% for Muller; P = 0.004) when compared with the Muller equation.

We compared the anthropometric model with the Mifflin et al. (24) equation, which was reported as the best equation to estimate RMR based on the analysis from the American Dietetic Association (9). An activity factor of 1.3 was used. By using the Mifflin equation, the average predicted 24EE (2122 ± 330 kcal/d) was not different from that predicted by our anthropometric model (2046 ± 305 kcal/d; P = 0.10) and the measured 24EE (2055 ± 343 kcal/d; P = 0.10). The correlation between the measured 24EE and that predicted by the Mifflin equation (r = 0.887) and the anthropometric model (r = 0.893) was also not significantly different (Figure 3B). The anthropometric model had a smaller mean error (296 kcal/d compared with 666161 kcal/d for Mifflin; P < 0.0001), but the 2 equations had similar prediction accuracy at the individual level (83% compared with 79% for Mifflin; P = 0.29).

Equations for energy prescription adjustment

Next, we developed equations to adjust energy intake after the chamber study had begun. As shown earlier, both anthropometric

TABLE 2

<table>
<thead>
<tr>
<th>Predictor</th>
<th>RMSE</th>
<th>Adjusted R²</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Energy prescription</td>
<td>kcal/d</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Body composition model (n = 238)</td>
<td>24EE = 546 kcal/d</td>
<td>26.2 [FFM (kg)] + 5.2 [FM (kg)] −96 (African American) −2.32 [age (y)]</td>
<td>173 0.773 &lt;0.0001</td>
</tr>
<tr>
<td>Anthropometric model (n = 241)</td>
<td>24EE = −235 kcal/d</td>
<td>11.6 [weight (kg)] + 217 (male) + 8.03 [height (cm)] −3.45 [age (y)] −52 (African American)</td>
<td>259 0.514 &lt;0.0001</td>
</tr>
<tr>
<td>Energy adjustment</td>
<td>kcal/d</td>
<td></td>
<td></td>
</tr>
<tr>
<td>3-h Anthropometric model (n = 240)</td>
<td>24EE = −113 kcal/d</td>
<td>4.14 [3-h EE (kcal)] + 5.66 [weight (kg)] + 100 (male) + 2.86 [height (cm)]</td>
<td>146 0.844 &lt;0.0001</td>
</tr>
<tr>
<td>7-h Anthropometric model (n = 240)</td>
<td>24EE = −285 kcal/d</td>
<td>2.11 [7-h EE (kcal)] + 4.29 [weight (kg)] + 3.34 [height (cm)] + 48 (male)</td>
<td>115 0.903 &lt;0.0001</td>
</tr>
</tbody>
</table>

1 24EE SD = 371 kcal/d. FFM, fat-free mass; FM, fat mass; RMSE, root of the mean square error; 24EE, 24-h energy expenditure.

2 Determined by stepwise linear regression model.
and body composition variables are similar in predicting 24EE, but the former are easily available and are thus more practical to use in clinical settings. We therefore used anthropometry to develop the subsequent equations. We predicted 24EE by using EE measurements, after 3 and 7 h, respectively, into the chamber stay (Table 2; Energy adjustment) as follows:

3-h Anthropometric Model: \[24\text{EE} (\text{kcal/d}) = 4.14[3\text{hEE} (\text{kcal})] + 5.66[\text{weight (kg)}] + 2.86[\text{height (cm)}] + 100 (\text{male}) - 113 \tag{3}\]

7-h Anthropometric Model: \[24\text{EE} (\text{kcal/d}) = 2.11[7\text{hEE} (\text{kcal})] + 4.29[\text{weight (kg)}] + 3.34[\text{height (cm)}] + 48 (\text{male}) - 285 \tag{4}\]

None of the predictions was significantly different from the measured 24EE (Figure 4, A and B). Compared with the anthropometric model for initial energy prescription, the 3- and 7-h equations did not improve the prediction at the group level because average 24EE and mean errors were all similar (Table 3). The 24EE estimations at 3 and 7 h, however, significantly increased the prediction accuracy from 83% when using the anthropometric model for initial energy prescription to 92% with the 3-h equation \((P = 0.003)\) and to 96% with the 7-h equation \((P < 0.0001)\). Furthermore, Bland-Altman analysis also suggested that, in contrast to the anthropometric model for initial energy prescription, using either the first 3- or 7-h measurements to predict 24EE eliminated the bias of increased deviation from 24EE at either end of the EE spectrum (Figure 4, C and D). The correlation between AEE and measured 24EE \((r = 0.517, P < 0.0001)\) was similar to that between AEE and 24EE predicted by using 3-h EE \((r = 0.533, P < 0.0001)\) or 7-h EE \((r = 0.539, P < 0.0001)\) but not by using the anthropometric model \((r = 0.336, P < 0.0001)\). These data suggest that the improvement in 24EE prediction by using 3- or 7-h EE results from better estimation of AEE.
DISCUSSION

Measurements of energy metabolism in a respiratory chamber are the most sophisticated measures of energy homeostasis because they allow real-time simultaneous measurement of EE and substrate oxidation in an experimental setting that is close to free-living conditions. These data, however, can only be appropriately interpreted when energy requirement is accurately predicted, and therefore study participants are in “reasonable” energy balance. We developed and validated equations that predict sedentary 24EE both before and during the chamber measurement. Our data suggest that, in a respiratory chamber, our equations are at least comparable, if not better, when compared with predictive equations reported in the literature. Importantly, we showed that an energy prescription can be further refined during the chamber study (after 3 and/or 7 h of measurements) by allowing adjustment of food intake to better achieve the target energy consumption and therefore energy balance.

The strength of our anthropometric model for initially prescribing energy intake before entering the chamber is that our 24EE prediction is specific to the setting of a respiratory chamber study. The conventional way to prescribe energy intake requires RMR and an activity factor and both components may be subject to errors, especially if neither of the variables are directly measured. Most chamber studies estimate RMR by using predictive equations, but their applicability is highly variable (9, 25). The use of an activity factor of 1.3, which is the same for estimating EE of a sedentary lifestyle (26), to account for spontaneous physical activity in the respiratory chamber is probably inaccurate because activity in a chamber is limited as a result of the confined space and facilities. Exploratory behaviors associated with an unfamiliar environment also alter PAL. In the current study, PAL ranged from 1.04 to 1.51, which clearly suggests that it is inappropriate to assign one activity factor to all study participants. We decided not to incorporate any estimation of spontaneous physical activity because it is likely to introduce more errors in the predictive equations. Also, we expect study participants in similar 24-h respiratory chamber studies in other research facilities to fall within a comparable range of PAL given the similar size of the chambers.

TABLE 3
Summary of predicted and measured 24EE in a respiratory chamber

<table>
<thead>
<tr>
<th>Energy prescription</th>
<th>Anthropometric model</th>
<th>Energy prescription</th>
</tr>
</thead>
<tbody>
<tr>
<td>Body composition model (n = 238)</td>
<td>Müller (22) (n = 239)</td>
<td>Mifflin (24) (n = 240)</td>
</tr>
<tr>
<td>Predicted 24EE (kcal/d)</td>
<td>2049 ± 320</td>
<td>2046 ± 305</td>
</tr>
<tr>
<td>Correlation (R²)</td>
<td>0.803</td>
<td>0.797</td>
</tr>
<tr>
<td>Mean error (kcal/d)</td>
<td>−4 ± 152</td>
<td>−9 ± 155</td>
</tr>
<tr>
<td>Prediction accuracy (%)</td>
<td>86</td>
<td>83</td>
</tr>
<tr>
<td>Minimum error (%)</td>
<td>−19</td>
<td>−20</td>
</tr>
<tr>
<td>Maximum error (%)</td>
<td>41</td>
<td>32</td>
</tr>
</tbody>
</table>

1 Measured 24EE = 2055 ± 343 kcal/d. 24EE, 24-h energy expenditure.
2 Mean ± SD (all such values).
3 Significantly different from the energy prescription body composition model (paired-samples t test): P < 0.0001.
4 Significantly different from the energy prescription anthropometric model (paired-samples t test): P < 0.0001.
5 Significantly different from the energy prescription body composition model (chi-square test): P = 0.01.
6,7 Significantly different from the energy prescription anthropometric model (chi-square test): 6P < 0.01, 7P < 0.0001.

FIGURE 3. Correlation between 24EE measured in a respiratory chamber and that predicted by the Müller (A; n = 239) (22) and Mifflin (B; n = 240) (24) equations. Solid lines: predictive model with 95% CIs; dotted line: lines of identity. A Pearson correlation was used to assess the relation between the variables. 24EE, 24-h energy expenditure.
It is perhaps somewhat surprising that body composition did not yield a better prediction of 24EE than anthropometric variables. In the current study, FFM was by far the strongest determinant of 24EE and accounted for 77% of its variance. When predicting 24EE for energy prescription, FFM alone in the body composition model and the combination of weight, height, and sex in the anthropometric model achieved comparable reductions in the root of the mean square errors of 24EE (Table 2). This suggests that anthropometric variables, when used in combination with demographic characteristics, can sufficiently account for variations in FFM, and thus its effect on 24EE. In agreement with our findings, a study in 2528 subjects (22) also concluded that body composition had “no clear advantage” when compared with anthropometric variables as predictors of RMR (22).

We developed equations to improve 24EE prediction after the initial energy prescription. Researchers are well aware of the deviation of energy prescription from energy balance and the consequences on the respiratory quotient and macronutrient oxidation rates. Attempts to achieve energy balance included providing energy at a fraction of the predicted 24EE (27, 28), incorporating a mandatory exercise regimen (29, 30), or adjusting energy intake on the basis of 24EE measured during multiple chamber stays (31). de Jonge et al (14) showed in a small number of research volunteers that 24EE could be predicted with the EE measurement after 3 and 7 h into the chamber study with reasonable accuracy. We asked whether such predictions would estimate 24EE better than energy prescription before the study. The 3- and 7-h time points were chosen because they were at least 2 h after food consumption (ie, at a time at which approximately half of the thermic effect of food was captured) and would then allow adjustments to lunch and/or dinner meals if necessary. We showed that using the 3- and 7-h EE did not improve overall 24EE prediction at the group level but improved the prediction accuracy at the individual level when compared with the initial energy prescription (anthropometric model). More importantly, our data suggest that using 3- and/or 7-h EE to predict 24EE removes any

FIGURE 4. Relations between 24EE measured in a respiratory chamber and that predicted at 3 (A, C) and 7 (B, D) h into the chamber stay (n = 238). A, B: Pearson correlation; the solid and dotted lines indicate the predictive model with 95% CIs and the line of identity, respectively. C, D: Bland-Altman plot; the solid line indicates the regression model, and dotted lines indicate the mean difference and 95% limits of agreement. A Pearson correlation was used to assess the relation between the variables. O, subjects who were at the 0–25th percentile of time spent moving in the respiratory chamber; △, subjects who were at the 25–75th percentile of time spent moving in the respiratory chamber; x, subjects who were at the 75–100th percentile of time spent moving in the respiratory chamber; 24EE, 24-h energy expenditure.
systematic bias across the EE spectrum (ie, overestimating 24EE in subjects with a low energy requirement and underestimating 24EE at the other end of the spectrum) (32, 33). It is logical to hypothesize that, in comparison to the anthropometric model, using 3- or 7-h EE improves the prediction of spontaneous physical activity and therefore 24EE. Given that the subjects were in sedentary conditions, the activity level should be similar throughout the chamber stay (except during sleep) and therefore either 3- or 7-h EE provided an indication of the activity level over the 24 h. Our data suggest that 24EE predictions using 3- and 7-h EE were similar with no advantage of one over another. This allows flexibility when the equations are incorporated in research protocols, where in some cases it is preferable to adjust energy intake in a specific meal or meals.

It is important to note that, although the equations are developed using data from our laboratory only and are somewhat specific to the research setting and protocol, we anticipate the equations to be applicable to research facilities around the world. We expect study participants in other 24-h chamber studies (without prescription of exercise) to fall with a comparable range of activity level because our chamber is of a typical size to conduct similar studies (24–30 m³) and therefore no exercise-related adjustment of 24EE prediction would be necessary. We should emphasize, however, that the use of the 3- and 7-h EE to predict 24EE would require a meal schedule and percentage distribution of daily energy intake over the meals similar to what we currently have in our laboratory (refer to Subjects and Methods for details). Because similar meal schedules and energy distribution ratios are already commonly used (34, 35), we expect only minor adjustments to the research protocol should other facilities incorporate our equations into their study procedures.

The current work showed the feasibility of using real-time EE to adjust energy intake to achieve energy balance in a chamber study. This provides an important basis for further modifications of the equations to be used in chamber studies beyond investigating sedentary 24EE. Modifications are certainly possible that will adapt the equations to energy prescriptions and adjustments in exercise protocols. For example when sessions of treadmill/cycle ergometer are included in the chamber studies, the exercise EE can be easily calculated on the basis of the characteristics of the subjects and the specifics of the exercise (eg, duration and intensity) (36, 37), and that can then be added to the predicted 24EE. Similar modifications may also apply to over- or underfeeding studies. Furthermore, it may also be possible to modify the equations to predict 24EE in free-living conditions. Physical activity EE is the most variable component of 24EE, which can vary from 3% to 50% in free-living conditions (23) and which makes estimations of free-living EE very difficult. Interestingly, Snitker et al (19) showed that physical activity in a respiratory chamber significantly correlated with that in the free-living environment. Our equations may also inform on free-living 24EE when an index of spontaneous physical activity in the chamber is incorporated in the 24EE prediction models.

In summary, we developed and validated a set of equations that use anthropometric and demographic variables to prescribe and adjust energy intake to achieve energy balance in sedentary respiratory chamber studies over 24 h. These equations may be modified to adapt to a wide range of chamber protocols, including exercise, overfeeding, or underfeeding. PAL in the chamber may be important to extend the use of our equations to predict total daily EE in free-living conditions.

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