

## Assessment and prediction of thoracic gas volume in pregnant women: an evaluation in relation to body composition assessment using air displacement plethysmography

Pontus Henriksson, Marie Löf and Elisabet Forsum\*

Department of Clinical and Experimental Medicine, Faculty of Health Sciences, Linköping University, SE 581 85 Linköping, Sweden

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### Abstract

Assessment of body fat (BF) in pregnant women is important when investigating the relationship between maternal nutrition and offspring health. Convenient and accurate body composition methods applicable during pregnancy are therefore needed. Air displacement plethysmography, as applied in Bod Pod, represents such a method since it can assess body volume (BV) which, in combination with body weight, can be used to calculate body density and body composition. However, BV must be corrected for the thoracic gas volume (TGV) of the subject. In non-pregnant women, TGV may be predicted using equations, based on height and age. It is unknown, however, whether these equations are valid during pregnancy. Thus, we measured the TGV of women in gestational week 32 ( $n$  27) by means of plethysmography and predicted their TGV using equations established for non-pregnant women. Body weight and BV of the women was measured using Bod Pod. Predicted TGV was significantly ( $P=0.033$ ) higher than measured TGV by 6% on average. Calculations in hypothetical women showed that this overestimation tended to be more pronounced in women with small TGV than in women with large TGV. The overestimation of TGV resulted in a small but significant ( $P=0.043$ ) overestimation of BF, equivalent to only 0.5% BF, on average. A Bland–Altman analysis showed that the limits of agreement were narrow (from  $-1.9$  to  $2.9\%$  BF). Thus, although predicted TGV was biased and too high, the effect on BF was marginal and probably unimportant in many situations.

**Key words:** Air displacement plethysmography; Body composition; Pregnancy; Thoracic gas volume

The nutritional situation of pregnant women is considered to be important for offspring as well as for maternal health<sup>(1,2)</sup>. Therefore, to secure optimal health of the population, the WHO advocates application of a so-called life-course approach emphasising the importance of appropriate nutrition for pregnant women<sup>(3)</sup>. The reason is that women in populations where malnutrition is common tend to deliver infants with low birth weight, a factor associated with increased morbidity and mortality<sup>(4)</sup>. Also underweight pregnant women in well-nourished populations may deliver low-birth-weight babies<sup>(2)</sup>. On the other hand, overweight and obesity in women are also associated with adverse outcomes of pregnancy, with common examples being gestational diabetes, pre-eclampsia, large-for-gestational-age babies and possibly an increased risk for overweight and obesity in the offspring<sup>(2)</sup>. The detailed nature of the relationship between the nutritional situation of a pregnant woman and the

development of her offspring is, however, incompletely known. This is partly due to a lack of appropriate measures indicating the nutritional status of pregnant women. For example, the commonly used estimate of body fatness, BMI, is a relatively poor estimate of body composition, especially during pregnancy<sup>(5)</sup>. However, accurate estimates of body composition are important when studying the relationships between the maternal nutritional situation and pregnancy outcome.

There are many available methods to assess human body composition, but accurate methods tend to be expensive and complicated and therefore not suitable for large-scale studies of pregnant women. However, air displacement plethysmography can assess body composition in a quick, accurate, safe and non-invasive way<sup>(6,7)</sup>, and therefore has potential for studies during pregnancy. In this technique, body volume (BV) of the subject is assessed requiring a correction for his/her thoracic gas volume (TGV). TGV can be

**Abbreviations:** BF, body fat;  $BF_{\text{measTGV}}$ , body fat calculated using measured thoracic gas volume;  $BF_{\text{predTGV}}$ , body fat calculated using predicted thoracic gas volume; BV, body volume;  $BV_{\text{measTGV}}$ , body volume calculated using measured thoracic gas volume;  $BV_{\text{predTGV}}$ , body volume calculated using predicted thoracic gas volume; FRC, functional residual capacity; TGV, thoracic gas volume;  $TGV_{\text{meas}}$ , measured thoracic gas volume;  $TGV_{\text{pred}}$ , predicted thoracic gas volume.

\* **Corresponding author:** E. Forsum, fax +46 1033570, email [elisabet.forsum@liu.se](mailto:elisabet.forsum@liu.se)

assessed as part of the measurement procedure<sup>(8)</sup>, but this assessment may be difficult and some subjects are unable to produce satisfactory results. A more convenient approach commonly used in non-pregnant adults is to predict TGV from height and age using sex-specific equations<sup>(9,10)</sup>. McCrory *et al.*<sup>(11)</sup> found no mean difference between predicted and measured TGV in such subjects and concluded that predicted TGV can produce estimates of body composition which are satisfactory for many purposes. Similar results have been reported by Demerath *et al.*<sup>(12)</sup>. However, the validity of predicted TGV has not been evaluated in pregnant women. During pregnancy, physiological and anatomical changes, such as increased subcostal angle, dislocation of the diaphragm and a growing uterus, occur and may affect TGV<sup>(13,14)</sup>. The aim of the present study was to compare the measured and predicted TGV of healthy women in gestational week 32. Body composition and BV results, calculated using these two kinds of TGV, were also compared.

## Materials and methods

### Subjects

A total of 249 healthy pregnant women were recruited during 2008–2010 for a study where body composition was assessed in gestational week 32. A group of forty consecutive participants (non-smoking, non-asthmatic) were asked to join the present study. Of these forty subjects, two declined participation and eleven failed to produce three acceptable measurements of TGV. Thus, the study included twenty-seven women. The study was conducted according to the guidelines laid down in the Declaration of Helsinki, and all procedures involving human subjects were approved by the Ethical Review Board in Linköping, Sweden. Informed consent, witnessed and formally recorded, was obtained from all subjects.

### Study outline

The measurements were conducted in the morning after an overnight fast. Height was measured with a wall stadiometer to the nearest 0.5 cm. The women were then instructed and allowed to practise measurement of TGV and, subsequently, their body weight, BV and TGV were measured using Bod Pod (COSMED USA, Inc.). Estimates of BV were corrected using either measured TGV ( $BV_{\text{measTGV}}$ ) or predicted TGV ( $BV_{\text{predTGV}}$ ) using equations in Bod Pod software 4.2.4 (COSMED USA, Inc.). Body fat (BF) was calculated by means of this software using either measured TGV ( $BF_{\text{measTGV}}$ ) or predicted TGV ( $BF_{\text{predTGV}}$ ). Additional information regarding women and infants was recorded using questionnaires in gestational week 32 (weight before pregnancy, parity) and after delivery (gestational weight gain, infant birth weight).

### Thoracic gas volume

TGV corresponds to the amount of air in the body, primarily in lungs and thorax, during a test in Bod Pod, and it consists

of the functional residual capacity (FRC) and approximately half of the tidal volume<sup>(6,7)</sup>. TGV was measured while the subject was sitting in the Bod Pod using a technique comparable to that used to measure FRC in standard plethysmography, referred to as the 'panting manoeuvre'<sup>(6,7)</sup>. As applied in the present study, the technique measures the average amount of air in the body during tidal breathing. Women wore a nose clip and were breathing normally during the measurement, first without and then through a piece of tubing. After a few tidal breaths, at mid-exhalation, a shutter valve in the airway closed, and then the women panted gently three times using a force equivalent to that needed to fog up a pair of glasses before cleaning them. To assess validity of measurements, two variables, merit and airway pressure, were used<sup>(6,7)</sup>. High values of merit may be due to leakage of air at the mouth seal or nose, whereas a high airway pressure may indicate closure of the glottis or significant alveolar compression. Measurements were repeated if merit was  $>1.0$  and/or airway pressure was  $\geq 35$  cm H<sub>2</sub>O as recommended by the manufacturer<sup>(7)</sup>. Results from three acceptable measurements were used to calculate average measured TGV ( $TGV_{\text{meas}}$ ). Predicted TGV ( $TGV_{\text{pred}}$ ) was calculated by means of Bod Pod software 4.2.4 as  $FRC + 0.35$  litres, which is considered to represent 50% of the tidal volume<sup>(10)</sup>. FRC is calculated according to Crapo *et al.*<sup>(9)</sup> as  $0.036 \times \text{height (cm)} + 0.0031 \times \text{age (years)} - 3.182$ .

### Body composition

Body composition was calculated by means of Bod Pod software 4.2.4 using the equation of Siri<sup>(15)</sup>:

$$1/D_B = F_{BF}/D_{BF} + (1 - F_{BF})/D_{FFBW}.$$

In this equation,  $D_B$  is the body density,  $D_{BF}$  is the density of BF ( $0.9007$  g/cm<sup>3</sup>),  $D_{FFBW}$  is the density of fat-free body weight and  $F_{BF}$  is the fraction of fat in the body. We used  $1.092$  g/cm<sup>3</sup> as the density of fat-free body weight since this is appropriate for women in gestational week 32<sup>(16)</sup>.

### Statistics

Values are given as means and standard deviations. Linear regression and correlation analysis, paired *t* tests and comparison of slopes of regression lines were conducted as described in Kleinbaum *et al.*<sup>(17)</sup>. Standard error of the estimate was calculated as the square root of the mean square error. Technical error of measurement was calculated as described by Ulijaszek & Kerr<sup>(18)</sup>. Agreement between methods was evaluated as described by Bland & Altman<sup>(19)</sup>. In a Bland and Altman evaluation, the difference between results obtained by means of a new method and a reference method is plotted *v.* the average of these two methods. The mean difference and 2SD (limits of agreement) were calculated. This procedure also provides an opportunity to identify bias in the results across the interval of measurements.  $P < 0.05$  was considered statistically significant and all tests of hypothesis were two-sided. Statistical analysis was performed using PASW Statistics 18 (IBM) or Statistica software 9.1 (StatSoft, Inc.).



**Table 1.** Characteristics of pregnant women (*n* 27) participating in the study

	Mean	SD	Range
Age at measurement (years)	31	5	19–40
Stage of gestation at measurement* (weeks)	31.4	0.3	31.0–32.3
Height (m)	1.68	0.06	1.57–1.77
Weight before pregnancy† (kg)	66	12	48–98
BMI before pregnancy‡ (kg/m <sup>2</sup> )	23.5	3.9	19.5–36.9
Weight at measurement (kg)	75.3	11.5	54.4–106.5
Gestational weight gain‡§ (kg)	14	5	–1 to 24
Infant birth weight† (kg)	3.48	0.57	2.47–4.54

\* Calculated from a routine ultrasound examination in approximately gestational week 12<sup>(24)</sup>.

† Self-reported data.

‡ Calculation based on self-reported weight before pregnancy and height measured as described in the Materials and methods section.

§ Calculated as the last known weight in pregnancy minus weight before pregnancy.

## Results

### Characteristics of women

Characteristics of the twenty-seven women and their infants are presented in Table 1. Before pregnancy, twenty (74%) women were of normal weight ( $18.5 \leq \text{BMI} < 25.0$ ), five (19%) were overweight ( $25.0 \leq \text{BMI} < 30.0$ ) and two (7%) were obese ( $\text{BMI} \geq 30.0$ ). Their parity ranged between 0 and 4, and their gestational weight gain was 14 (SD 5) kg. Infants, all full term, healthy and of appropriate weight for gestational age at delivery, weighed 3.48 (SD 0.57) kg at birth. There was a significant linear relationship between weight in gestational week 32 (*y*) and height (*x*) ( $y = -59.847 + 80.464x$ ,  $r = 0.403$ ,  $P = 0.037$ ).

### Thoracic gas volume

As shown in Table 2, average  $\text{TGV}_{\text{meas}}$  varied between 2.404 and 3.980 litres for the twenty-seven women. For these women, mean  $\text{TGV}_{\text{meas}}$  was 3.127 litres (SD 0.488). The average within-subject standard deviation was 0.184 litres, with

**Table 2.** Predicted and measured thoracic gas volume (TGV) as well as body volume (BV) and body fat (BF) calculated using predicted and measured TGV for women (*n* 27) in gestational week 32

	Mean	SD	Range
$\text{TGV}_{\text{pred}}$ (litres)	3.314*	0.206	2.915–3.628
$\text{TGV}_{\text{meas}}$ (litres)	3.127	0.488	2.404–3.980
$\text{BV}_{\text{predTGV}}$ (litres)	74.211†	11.919	53.047–106.700
$\text{BV}_{\text{measTGV}}$ (litres)	74.136	11.878	52.855–106.371
$\text{BF}_{\text{predTGV}}$ (%)	35.0‡	5.3	24.0–44.7
$\text{BF}_{\text{measTGV}}$ (%)	34.5	5.2	21.9–44.2

$\text{TGV}_{\text{pred}}$ , predicted TGV;  $\text{TGV}_{\text{meas}}$ , measured TGV;  $\text{BV}_{\text{predTGV}}$ , BV calculated using  $\text{TGV}_{\text{pred}}$ ;  $\text{BV}_{\text{measTGV}}$ , BV calculated using  $\text{TGV}_{\text{meas}}$ ;  $\text{BF}_{\text{predTGV}}$ , BF calculated using  $\text{TGV}_{\text{pred}}$ ;  $\text{BF}_{\text{measTGV}}$ , BF calculated using  $\text{TGV}_{\text{meas}}$ .

\* Mean value was significantly different from  $\text{TGV}_{\text{meas}}$  ( $P = 0.033$ ).

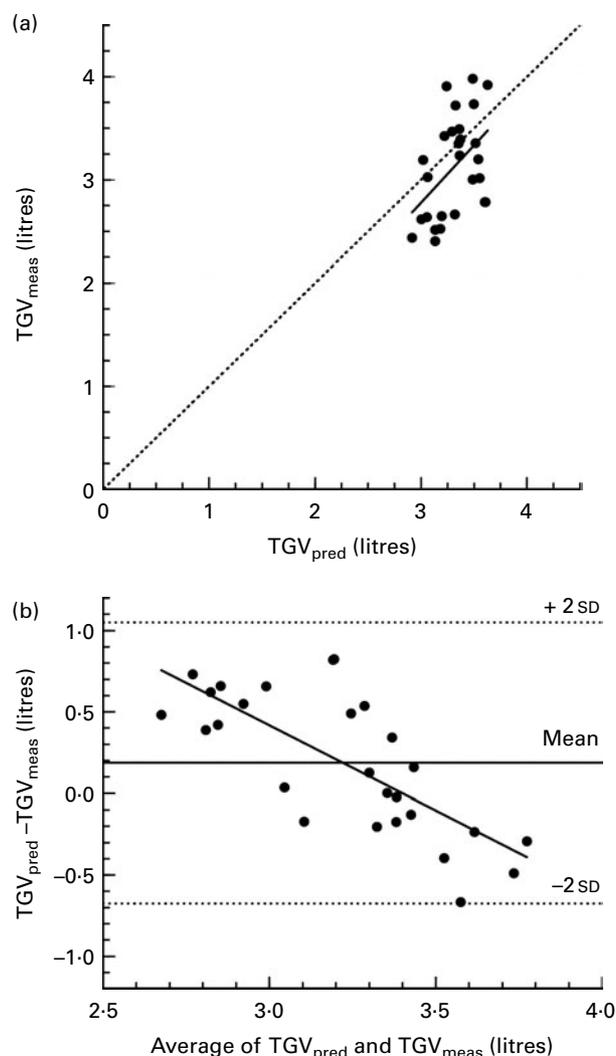
† Mean value was significantly different from  $\text{BV}_{\text{measTGV}}$  ( $P = 0.033$ ).

‡ Mean value was significantly different from  $\text{BF}_{\text{measTGV}}$  ( $P = 0.043$ ).

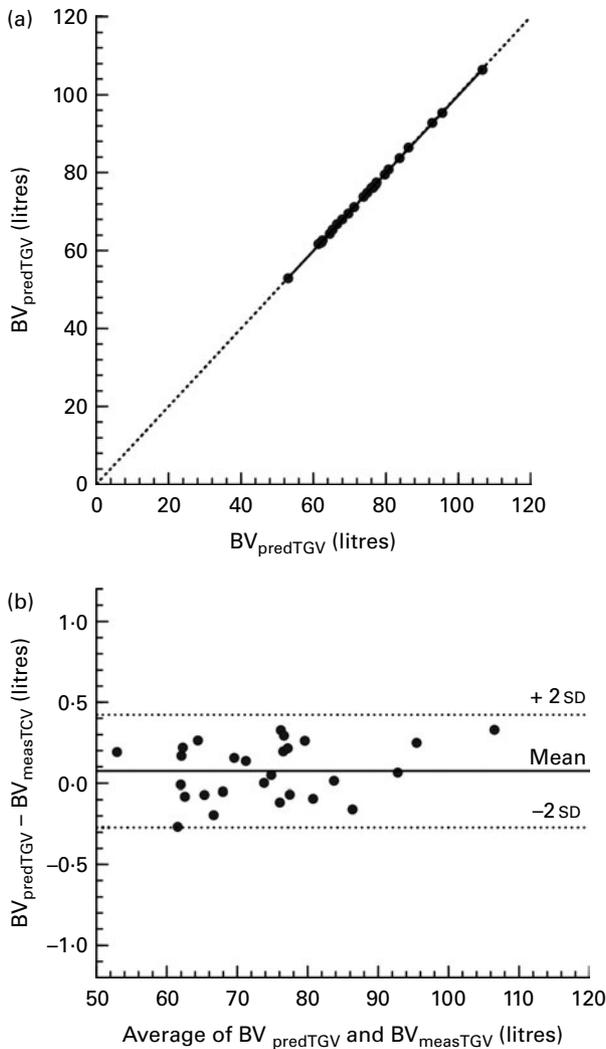
the corresponding CV being 5.9%. The technical error of measurement was 0.210 litres or 6.7%.

### Predicted and measured thoracic gas volume

Values for  $\text{TGV}_{\text{pred}}$  and  $\text{TGV}_{\text{meas}}$  are presented in Table 2.  $\text{TGV}_{\text{pred}}$  (3.314 litres) was significantly higher, by 6.0% or 0.187 litres, than  $\text{TGV}_{\text{meas}}$  (3.127 litres).  $\text{TGV}_{\text{pred}}$  and  $\text{TGV}_{\text{meas}}$  were significantly correlated (Fig. 1(a)). A Bland–Altman scatter plot (Fig. 1(b)) demonstrates a significant negative linear relationship when the regression of the difference between  $\text{TGV}_{\text{pred}}$  and  $\text{TGV}_{\text{meas}}$  *v.* the average of  $\text{TGV}_{\text{pred}}$  and  $\text{TGV}_{\text{meas}}$  is performed. The limits of agreement are wide (from –0.675 to 1.049 litres) corresponding to –21.0 and 32.6% of the



**Fig. 1.** (a) Regression of measured thoracic gas volume ( $\text{TGV}_{\text{meas}}$ , *y*) *v.* predicted TGV ( $\text{TGV}_{\text{pred}}$ , *x*). The slope of the regression line ( $y = -0.568 + 1.115x$ ,  $r = 0.471$ , standard error of the estimate 0.439,  $P = 0.013$ ) is not significantly ( $P = 0.786$ ) different from the line of identity ( $y = x$ ). (b) Bland–Altman scatter plot; regression of  $\text{TGV}_{\text{pred}} - \text{TGV}_{\text{meas}}$  (*y*) *v.* the average of  $\text{TGV}_{\text{pred}}$  and  $\text{TGV}_{\text{meas}}$  (*x*). The solid line represents the mean difference between  $\text{TGV}_{\text{pred}}$  and  $\text{TGV}_{\text{meas}}$  (0.187 litres) and the dashed lines are the limits of agreement ( $2\text{SD} = 0.862$  litres). The regression equation is  $y = 3.546 - 1.043x$ ,  $r = -0.741$  ( $P < 0.001$ ). Data collected from twenty-seven women in gestational week 32.



**Fig. 2.** (a) Regression of body volume (BV) calculated using measured thoracic gas volume (TGV) ( $BV_{measTGV}$ ,  $y$ ) v. BV calculated using predicted TGV ( $BV_{predTGV}$ ,  $x$ ). The slope of the regression line ( $y = 0.186 + 0.996x$ ,  $r > 0.999$ , standard error of the estimate 0.171,  $P < 0.001$ ) is not significantly ( $P = 0.194$ ) different from the line of identity ( $y = x$ ). (b) Bland–Altman scatter plot; regression of  $BV_{predTGV} - BV_{measTGV}$  ( $y$ ) v. the average of  $BV_{predTGV}$  and  $BV_{measTGV}$  ( $x$ ). The solid line represents the mean difference between  $BV_{predTGV}$  and  $BV_{measTGV}$  (0.075 litres) and the dashed lines are the limits of agreement ( $2SD = 0.346$  litres). The regression equation is  $y = -0.179 + 0.003x$ ,  $r 0.236$  ( $P = 0.235$ ). Data collected from twenty-seven women in gestational week 32.

average of  $TGV_{pred}$  and  $TGV_{meas}$ . The bias in TGV ( $TGV_{pred} - TGV_{meas}$ ) in gestational week 32 was correlated neither with height ( $r 0.028$ ,  $P = 0.888$ ) nor with weight ( $r 0.234$ ,  $P = 0.24$ ) of the women.

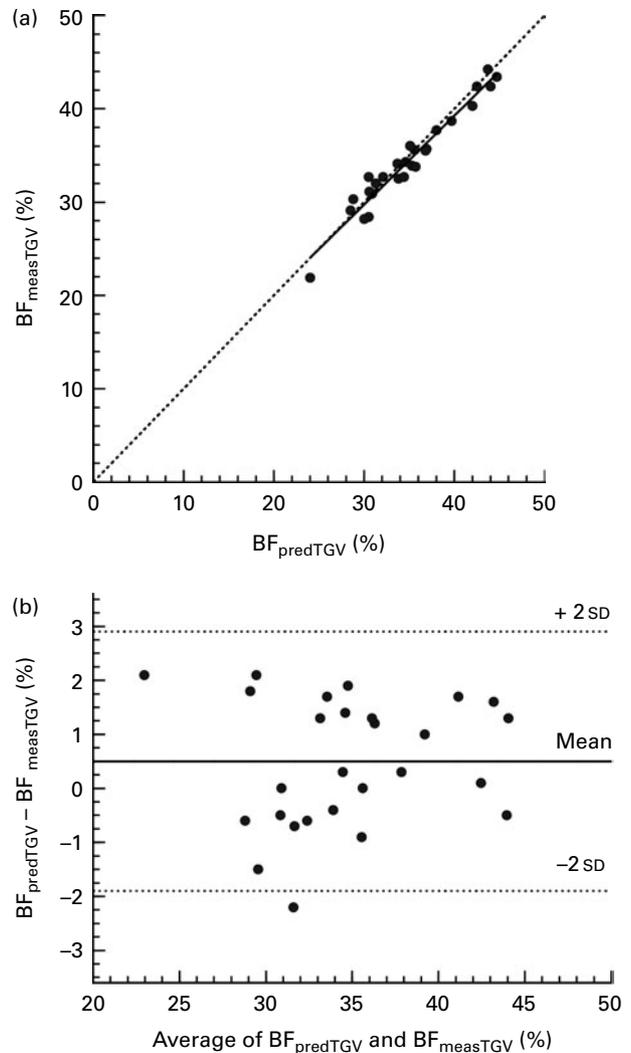
*Body volume calculated using predicted v. measured thoracic gas volume*

Estimates of  $BV_{predTGV}$  and  $BV_{measTGV}$  are presented in Table 2. Average  $BV_{predTGV}$  (74.211 litres) was slightly but significantly higher than average  $BV_{measTGV}$  (74.136 litres). There was a significant correlation between  $BV_{predTGV}$  and  $BV_{measTGV}$  (Fig. 2(a)). Fig. 2(b) shows that the average difference

between  $BV_{predTGV}$  and  $BV_{measTGV}$  is 0.075 litres. The limits of agreement are very small, from  $-0.271$  to  $0.421$  litres equivalent to  $-0.4$  to  $0.6\%$  of the average of  $BV_{predTGV}$  and  $BV_{measTGV}$ . No significant correlation was found when the regression of the difference between  $BV_{predTGV}$  and  $BV_{measTGV}$  v. the average of  $BV_{predTGV}$  and  $BV_{measTGV}$  was performed.

*Body composition calculated using predicted v. measured thoracic gas volume*

Estimates of  $BF_{predTGV}$  (%) and  $BF_{measTGV}$  (%) are presented in Table 2. Average  $BF_{predTGV}$  (35.0%) was slightly (0.5% BF) but significantly higher than average  $BF_{measTGV}$  (34.5%). There



**Fig. 3.** (a) Regression of body fat (BF) calculated using measured thoracic gas volume (TGV) ( $BF_{measTGV}$ ,  $y$ ) v. BF calculated using predicted TGV ( $BF_{predTGV}$ ,  $x$ ). The slope of the regression line ( $y = 1.139 + 0.953x$ ,  $r 0.974$ , standard error of the estimate 1.2,  $P < 0.001$ ) is not significantly ( $P = 0.296$ ) different from the line of identity ( $y = x$ ). (b) Bland–Altman scatter plot; regression of  $BF_{predTGV} - BF_{measTGV}$  ( $y$ ) v. the average of  $BF_{predTGV}$  and  $BF_{measTGV}$  ( $x$ ). The solid line represents the mean difference between  $BF_{predTGV}$  and  $BF_{measTGV}$  (0.5% BF) and the dashed lines are the limits of agreement ( $2SD = 2.4\%$  BF). The regression equation is  $y = -0.267 + 0.022x$  ( $r 0.095$ ,  $P = 0.638$ ). Data collected from twenty-seven women in gestational week 32.

was a significant correlation between  $BF_{predTGV}$  (%) and  $BF_{measTGV}$  (%) (Fig. 3(a)). The Bland–Altman scatter plot in Fig. 3(b) shows that the limits of agreement are small, from  $-1.9$  to  $2.9\%$  BF, equivalent to  $-5.5$  to  $8.3\%$  of the average of  $BF_{predTGV}$  and  $BF_{measTGV}$ . No significant correlation was found when the regression of the difference between  $BF_{predTGV}$  (%) and  $BF_{measTGV}$  (%) *v.* the average of  $BF_{predTGV}$  (%) and  $BF_{measTGV}$  (%) was performed.

**Discussion**

The women in the present study were recruited from a well-educated middle-class population. They were not randomly selected, but the proportion of them being overweight or obese before pregnancy as well as their weight gain during pregnancy were similar to comparable data previously reported for Swedish women<sup>(20,21)</sup>. Furthermore, the mean and variability in birth weight of their infants were in good agreement with Swedish reference data<sup>(22)</sup>. Therefore, we consider that the women in the present study are likely to be representative of Swedish women while in the pregnant state.

Although no study has examined the validity of TGV, assessed by means of Bod Pod, this set-up has been shown to produce very accurate estimates of FRC which is the major component of TGV<sup>(8)</sup>. The CV of our TGV measurements,  $5.9\%$ , was slightly lower than a comparable value ( $7.2\%$ ) reported by Crapo *et al.*<sup>(9)</sup>. The technical error of measurement in the present study was  $0.210$  litres while the corresponding value reported by Minderico *et al.*<sup>(23)</sup> was  $0.201$  litres. These findings indicate that the repeatability of our measurements was satisfactory. The correlation between  $TGV_{pred}$  and  $TGV_{meas}$ ,  $0.47$ , is lower in the present study than the comparable value,  $0.79$ , reported by McCrory *et al.*<sup>(11)</sup> for non-pregnant healthy adults. The lower value observed for pregnant women is hardly surprising since

pregnancy may well influence TGV in ways not related to body height or age. Furthermore, the value reported by McCrory *et al.*<sup>(11)</sup> was assessed in a population consisting of men and women having a variation in TGV larger than in our sample. The correlation between  $BF_{predTGV}$  (%) and  $BF_{measTGV}$  (%) in the present study,  $r\ 0.97$ , is comparable to corresponding values reported by McCrory *et al.*<sup>(11)</sup> ( $r\ 0.99$ ) and by Demerath *et al.*<sup>(12)</sup> ( $r\ 0.97$ ). Furthermore, the standard error of the estimate of these regression equations was similar in the three studies, i.e.  $1.36$ <sup>(11)</sup> and  $1.88$ <sup>(12)</sup> *v.*  $1.2\%$  BF in the present study. When calculations were based on  $TGV_{pred}$ , the present study and the study by McCrory *et al.*<sup>(11)</sup> classified a large proportion of the subjects ( $> 80\%$ ) within  $\pm 2\%$   $BF_{measTGV}$ .

Our Bland–Altman scatter plot in Fig. 1(b) demonstrates a significant relationship when the regression of  $TGV_{pred} - TGV_{meas}$  *v.* the average of  $TGV_{pred}$  and  $TGV_{meas}$  is performed, suggesting that the overestimation introduced when prediction equations are used to calculate TGV is influenced by the size of TGV. Similar observations have been made by Minderico *et al.*<sup>(23)</sup>. Considering that the height of a woman is the main predictor of TGV, we performed the calculation presented in Table 3 where the average biases in TGV, BV and BF, respectively, are estimated for three hypothetical women with short, average and tall body height. The mean height of the women in our sample was  $168$  (SD  $6$ ) cm. Thus, we considered an average woman to be  $168$  cm, a small woman  $156$  ( $168 - 2 \times 6$ ) cm and a tall woman  $180$  ( $168 + 2 \times 6$ ) cm. All three women in Table 3 were assumed to be 30 years old and to contain  $35\%$   $BF_{predTGV}$ . Their  $TGV_{pred}$  was calculated using height and age and average bias using the regression equation given in Fig. 1(a), as indicated in Table 3. For a short woman, estimating TGV from height and age introduced an average bias in this estimate of  $0.237$  litres, whereas the corresponding values for a woman of average height and for a tall woman were  $0.187$  and  $0.138$  litres, respectively. Furthermore, as shown in

**Table 3.** Predicted thoracic gas volume (TGV), body volume (BV) and body fat (BF), and estimated biases in these variables for three hypothetical women with small, average and large TGV, respectively, 30 years old, being in gestational week 32 and containing  $35\%$  BF

	Small $TGV_{pred}^*$	Average $TGV_{pred}^\dagger$	Large $TGV_{pred}^\ddagger$
$TGV_{pred}$ (litres)§	2.877	3.309	3.741
Average bias (litres)	0.237	0.187	0.138
$BV_{predTGV}$ (litres)¶	64.636	74.139	83.641
Average bias (litres)**	0.095	0.075	0.055
$BF_{predTGV}$ (%)	35.0	35.0	35.0
Average bias (%)††	0.8	0.5	0.3

$TGV_{pred}$ , predicted TGV;  $BV_{predTGV}$ , BV calculated using  $TGV_{pred}$ ;  $BF_{predTGV}$ , BF calculated using  $TGV_{pred}$ .  
 \* Representing a woman containing  $35\%$   $BF_{predTGV}$ , being  $1.56$  m tall and weighing  $65.7$  kg (predicted using the equation:  $-59.847 + 80.464 \times 1.56$  as described in the Results section).  
 † Representing a woman containing  $35\%$   $BF_{predTGV}$ , being  $1.68$  m tall and weighing  $75.3$  kg (predicted using the equation:  $-59.847 + 80.464 \times 1.68$  as described in the Results section).  
 ‡ Representing a woman containing  $35\%$   $BF_{predTGV}$ , being  $1.80$  m tall and weighing  $85.0$  kg (predicted using the equation:  $-59.847 + 80.464 \times 1.80$  as described in the Results section).  
 § Calculated from height and age using Bod Pod software as described in the Materials and methods section.  
 || Calculated as  $TGV_{pred}$  minus a TGV value calculated as follows:  $-0.568 + 1.115 \times TGV_{pred}$  (Fig. 1(a)).  
 ¶ Calculated as body weight divided by body density corresponding to  $35\%$  BF ( $1.016$  litres/kg).  
 \*\* Calculated as  $BV_{predTGV}$  minus a value for BV calculated using a TGV value calculated as follows:  $-0.568 + 1.115 \times TGV_{pred}$  (Fig. 1(a)).  
 †† Calculated as  $BF_{predTGV}$  (%) minus a value for BF (%) calculated using a TGV value calculated as follows:  $-0.568 + 1.115 \times TGV_{pred}$  (Fig. 1(a)).

Table 3, the average biases in BV and BF (%) were small and not very different between a short woman with a small TGV and a tall woman with a large TGV. These observations are due to the fact that TGV is small in relation to BV and that only 40% of TGV enters the equation when correcting BV for TGV<sup>(6,7)</sup>. Thus, although it is certainly true that predicting TGV when studying pregnant women in Bod Pod introduces a bias in the estimates of BV and BF (%), this bias is small and unlikely to be of practical significance in most situations. Thus, for example, for the average woman in Table 3, the value obtained for BF when predicted TGV is used in the calculations is 35.0% while the 'true' value is 34.5%.

There is a continuous decrease in TGV throughout pregnancy<sup>(13,14)</sup>. Therefore, it is likely that BV and BF can be calculated from TGV<sub>pred</sub> without any risk for a large bias in pregnant women also before gestational week 32. During the last few weeks of pregnancy, TGV is known to be further decreased with an average maximal decrease of approximately 300 ml<sup>(13,14)</sup>, which could possibly increase the bias in TGV<sub>pred</sub>. However, the average bias in BV in such a situation would still be quite moderate, 120 ml (40% of 300 ml). This would in turn lead to an average bias in BF (%) of 0.9% for the short woman in Table 3, whereas the average bias for the average and tall women would be 0.8 and 0.7% BF, respectively. Therefore, we consider that the average bias introduced by using prediction equations is likely to be small also during the last weeks of gestation.

It is relevant to point out, however, that caution is needed if the Bod Pod procedure is used during pregnancy to assess BV in a study where a three- or four-compartment model is used to obtain reference body composition data. The bias introduced by predicting TGV may then be important, perhaps especially in short women. In such a situation, it seems well motivated to measure rather than to predict TGV.

In summary, predicting TGV in gestational week 32 using height and age results in estimates of TGV that are biased and too high on the average. However, using such predicted TGV when assessing BV and BF by means of air displacement plethysmography (Bod Pod) gives results with very slight, and for most purposes unimportant, bias. Thus, predicted TGV is appropriate in most situations when studying body composition of pregnant women at least before gestational week 32.

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