# EVALUATION OF ERRORS CAUSED BY PULMONARY AND CUTANEOUS EVAPORATION IN BODY COMPOSITION ASSESSMENTS BY REPEATED MEASURES PROTOCOLS OF AIR DISPLACEMENT PLETHYSMOGRAPHY

P. MUNTEAN<sup>\*</sup>, A. PETRAȘ<sup>\*</sup>, SANDRA POPA<sup>\*</sup>, IRINA SIMA<sup>\*</sup>, B. GLIȘICI<sup>\*</sup>, O. CÎRJA<sup>\*</sup>, F. SCHICK<sup>\*</sup>, A. NEAGU<sup>\*,\*\*</sup>, OANA MUNTEANU<sup>\*</sup>, V. PUPĂZAN<sup>\*</sup>, MONICA NEAGU<sup>\*#</sup>

\*Discipline of Biophysics, Department of Functional Sciences, "Victor Babeş" University of Medicine and Pharmacy, Timişoara, Romania, <sup>#</sup>e-mail: neagu.monica@umft.ro \*\*Department of Physics & Astronomy, University of Missouri, Columbia, U.S.A.

Abstract. Air displacement plethysmography (ADP) evaluates body fat percentage (%BF) by measuring body mass (BM) and body volume (BV). ADP performed by the BOD POD [6] demonstrated excellent repeatability, with a technical error of measurement (TEM) of the order of 1 %BF. Repeated measures protocols were proposed for spotting unreasonable results, as well as for improving overall reliability. One such protocol asks to perform at least two consecutive ADP tests and take their mean if they differ by at most 1 %BF; otherwise, to perform a third test and take the mean of the two closest %BF values. Such a protocol was found to significantly improve the reliability of body composition assessments in middle-aged women. Nevertheless, a study of the BOD POD's reliability indicated that BM underwent a statistically significant drop between two consecutive tests. A steady loss of BM of the order of 10 g/h is mainly attributed to pulmonary and cutaneous evaporation. Therefore, the drop of BM between successive tests might cause a systematic error in %BF assessments by repeated measures protocols of ADP. The present study aims to assess this error and to compare it to the TEM of %BF estimates using the BOD POD. Our study was performed on a highly heterogeneous sample of 65 healthy adults (38 men and 27 women), aged 18-49 years, who spanned a wide range of BV (41-130 L) and BMI (16.2-41.5 kg/m<sup>2</sup>). We performed two successive BOD POD tests for each subject and calculated the rate of change in body mass, obtaining  $-68\pm59$  g/h. Since, on the average, one test lasted about 10 minutes, we estimated that the mean drop in body mass between two consecutive tests was of about 11 g, causing an increase in the measured % BF of the order of 0.1%. To further evaluate the impact of pulmonary and cutaneous evaporation on the results of repeated BOD POD measurements, we also performed a Bland-Altman analysis by plotting the differences vs. the means of BM, BV, and %BF values recorded in two successive tests. In conclusion, the errors caused by loss of body mass between consecutive measurements are about an order of magnitude smaller than the TEM of %BF assessments via ADP.

Key words: BOD POD reliability, body fat percentage, technical error of measurement.

Received: August 2018; in final form September 2018.

ROMANIAN J. BIOPHYS., Vol. 28, No. 4, P. 137-146, BUCHAREST, 2018

### **INTRODUCTION**

The BOD POD® Body Composition Tracking System assesses the amount of full body fat (BF) by measuring body mass (BM) and body volume (BV) via the air displacement plethysmography (ADP) technique [6].

The BOD POD displayed excellent repeatability [1, 4, 5, 8, 10, 11, 14], with a technical error of measurement (*TEM*) of 0.8 %*BF* [5] and mean within-subject coefficients of variation (*CVs*) ranging from 1.7% to 4.5% [8]. Nevertheless, to further improve reliability, Collins and McCarthy suggested to perform at least two complete trials, followed by a third one if the first two were not within 0.5 %*BF* [5]. Tucker *et al.* proposed to conduct a pair of trials, followed by a third one if the first two differed by more than 1 %*BF*, and evaluated the reliability benefits of their protocol on a sample of 283 middle-aged women [14].

Such repeated measures protocols have the potential to improve the precision of body composition assessments by ADP provided that the subjects' bodies do not change from one trial to another.

An extensive study of the BOD POD's reliability revealed statistically significant differences between BM values recorded in two successive trials [11]. Body volume also decreased, marginally missing statistical significance, whereas body density (and, thus %BF) did not change significantly [11]. Hence, loss of BM between trials in a repeated measures protocol might cause systematic errors in the results of body composition assessments. The present work seeks to evaluate these errors and to compare them with the typical error made in individual ADP tests. To this end, we performed duplicate trials on a heterogeneous sample of subjects and estimated the rate of BM loss, the change in BV, as well as their impact on %BF, resting metabolic rate (RMR) and total energy expenditure (TEE).

# MATERIALS AND METHODS

## STUDY POPULATION

A sample of 65 healthy adults (38 men and 27 women) gave informed consent to participate in our study.

This study was conducted in accord with the Declaration of Helsinki and was approved by the Ethics Committee of the "Victor Babeş" University of Medicine and Pharmacy, Timişoara, Romania.

#### ADP MEASUREMENTS

ADP measurements were performed using a BOD POD<sup>®</sup> Gold Standard Body Composition Tracking System (COSMED USA, Inc., CA, USA), with BOD POD software version 5.3.2. Scale calibration and system quality check were done daily, before each set of measurements. ADP tests were scheduled either in the morning (after overnight fasting), or around noon. Subjects were asked to refrain from eating and drinking for at least 4 hours before being tested. Also, they avoided alcohol consumption and intense exercise for at least 12 hours before the tests. Upon their arrival to the body composition laboratory, they were asked to use the restroom if they did not declare to have done so during the last 30 minutes.

The operator measured the subject's standing height to the nearest 0.5 cm. Then she/he entered the subject's data into the BOD POD software and ran two successive ADP tests, with an average duration of about 10 minutes each. Both tests were conducted by the same operator for a given subject.

In the BOD POD measurement chamber, the subjects wore swim caps provided on site and form-fitting swimsuits of their own. To avoid variability related to posture [12], the subjects were instructed to adopt a precise position in the BOD POD chamber, with hands resting on their knees and straightened back without leaning on the backrest of the seat.

The BOD POD software was used to predict thoracic gas volume, to measure body mass and body volume, to calculate % BF using the Siri formula [13], and to estimate the resting metabolic rate (*RMR*) and total energy expenditure (*TEE*).

#### STATISTICAL ANALYSIS

For data analysis, we used the Statistics Toolbox of MATLAB 7.13 (The MathWorks, Natick, MA, USA).

We calculated the rate of change in each subject's *BM* during the time interval between two consecutive trials,  $(BM_2 - BM_1)/(t_2 - t_1)$ , where  $t_1$  is the moment of time when the first trial was started and  $BM_1$  is the body mass recorded during the first trial, whereas  $t_2$  and  $BM_2$  are the corresponding quantities regarding the second trial. We also computed the mean and standard deviation (*SD*) of these rates.

We performed Bland-Altman analyses of various body composition parameters by plotting the differences,  $D_i$  (where the index *i* labels subjects), *versus* the mean of the pair of values obtained for each subject. More precisely, the Bland-Altman analysis consists in (i) plotting the difference *versus* the mean of pairs of recorded values, (ii) calculating the bias, defined as the mean difference  $(\overline{D})$ , and (iii) calculating the 95% limits of agreement,  $\overline{D}\pm 1.96SDD$ , where *SDD* denotes the standard deviation of the differences [3]. We also computed 95% confidence intervals (*CI*) for the bias and for the limits of agreement. To this end, we calculated the standard error of the bias,  $SDD/\sqrt{n}$ , and the *t*-value at which Student's probability density function with n-1 degrees of freedom takes the value 0.05 (for a 95% level of confidence). Then we expressed the *CI* of the bias in terms of these quantities as  $\overline{D}\pm t\cdot SDD/\sqrt{n}$ . For the limits of agreement, the standard error is  $SDD\sqrt{3/n}$ ; the corresponding *CI* is given by  $LLA\pm t\cdot SDD\sqrt{3/n}$  for the lower limit of agreement (*LLA*), and  $ULA \pm t \cdot SDD\sqrt{3/n}$  for the upper limit of agreement (*ULA*) [3, 9].

## RESULTS

This study was performed on a sample of 65 healthy adults (38 men and 27 women), which spanned a wide range of body compositions (Table 1). The parameters listed in Table 1 are the mean of the pair of values obtained in two successive ADP trials.

## Table 1

Characteristics of the study population described in terms of mean values  $\pm$  standard deviation (*SD*) and the range of values (minimum – maximum) of the age, body mass index (*BMI*), body mass (*BM*), body volume (*BV*) and body fat percentage (%*BF*).

Subjects	All		Men		Women	
	Mean±SD	Range	Mean±SD	Range	Mean±SD	Range
Age (years)	26.6±9.1	18.0-49.2	26.3±8.4	18.0-48.2	26.9±10.3	18.5-49.2
<b>BMI</b> (kg/m <sup>2</sup> )	24.4±5.3	16.2-41.5	26.5±5.2	20.2-41.5	21.5±4.0	16.2-33.8
<b>BM</b> (kg)	73.5±20.7	44.1-130	85.0±17.9	57.3-130	57.3±11.6	44.1–97.5
<b>BV</b> (L)	70.1±20.3	41.4–130	80.6±18.4	52.6-130	55.3±12.1	41.4–97.7
% <b>BF</b> (%)	21.6±10.2	4.4-45.9	18.2±10.1	4.4-45.9	26.5±8.3	14.5-45.9

The rate of change in *BM* for our sample was  $-68 \pm 59$  g/h (mean  $\pm SD$ ). This is a typical value of the rate of *BM* loss attributed to the elimination of water vapours via the lungs and the skin [2].

Assuming that the BV does not change during the time interval elapsed between the two trials, one can estimate the impact of evaporation on the measured %BF. In our study, the average duration of one test was about 10 min, leading to an average BM loss of 11.3 g. For a generic person that weighs 73.5 kg and has 21.6 %BF (our sample averages), we can express the body fat percentage recorded in the first trial using the Siri formula [13]:

$$\frac{\% BF_1}{100\%} = \frac{4.95BV}{BM_1} - 4.5,\tag{1}$$

where BV is the body volume. Similarly, the body fat percentage recorded in the second trial can be expressed as

$$\frac{\% BF_2}{100\%} = \frac{4.95BV}{BM_1 - m} - 4.5 \tag{2}$$

with m denoting the *BM* loss between the two tests. Subtracting Eq. (1) from Eq. (2) and expressing *BV* from Eq. (1), one obtains

$$\% BF_2 - \% BF_1 = \frac{m}{BM_1 - m} \left(\frac{\% BF_1}{100\%} + 4.5\right) 100\%,$$
 (3)

which, for our sample averages,  $\% BF_1 = 21.6\%$ ,  $BM_1 = 73.5$  kg,  $m = 11.3 \times 10^{-3}$  kg, gives  $\% BF_2 - \% BF_1 = 0.073\%$ .

We further present the results of the Bland-Altman analysis of various body composition parameters recorded for each subject in a pair of tests. In the plot of Fig. 1, a circular marker represents the difference vs. the mean of the pair of BM values recorded for a given subject in two consecutive trials. Note that, for about 90% of the investigated subjects, the body mass measured during the first trial  $(BM_1)$  was larger than the one recorded in the second trial  $(BM_2)$ .



Fig. 1. Bland-Altman plot of differences *vs.* means of the body mass values  $BM_1$  and  $BM_2$  recorded in the first and second trial, respectively. The solid horizontal line represents the bias, whereas dotted horizontal lines represent the limits of agreement. Error bars on the right depict the 95% confidence intervals (*CI*) of the statistical quantities represented by the corresponding horizontal lines.

The bias, of about 10 g, defined as mean value of  $(BM_1 - BM_2)$ , is statistically significant: the value zero lies outside of the corresponding *CI*, depicted as an error bar intersecting the solid horizontal line that represents the bias.

In Fig. 1, the circular markers are non-uniformly distributed around the solid line that represents the bias. The data points of subjects with relatively small *BM* lie mainly below the line of bias, whereas data points of relatively massive subjects have an opposite tendency. The larger is the *BM*, the larger is the drop of *BM* from one trial to another.

There is no such tendency in the Bland-Altman plot of the change in BV between successive tests (Fig. 2). Here the bias is -81 mL, marginally different from zero, and individual data points are uniformly distributed around the line that represents the bias.



Fig. 2. Bland-Altman plot of differences *vs.* means of *BV*s recorded in two successive BOD POD trials. (Notations are explained in the caption of Fig. 1.)

Figure 3 is the Bland-Altman plot of differences vs. means of %BF values assessed in two successive BOD POD trials. The bias, of about -0.6 %BF, is significantly different from zero. The data points are uniformly distributed around the solid horizontal line that depicts the bias.



Fig. 3. Bland-Altman plot of differences vs. means of  $\% BF_1$  and  $\% BF_2$  recorded during the first and second trial, respectively. (See the caption of Fig. 1 for notations.)

Figure 4 shows the Bland-Altman analysis of the estimated resting metabolic rate (*RMR*) in two contiguous ADP tests. The bias of 10 kcal is significantly different from zero, with *CI* ranging from 5.5 to 14.5 kcal.



Fig. 4. Bland-Altman plot of differences vs. means of estimates of *RMR* in two successive trials. (Notations are described in the caption of Fig. 1.)



Fig. 5. Bland-Altman analysis of total energy expenditure (*TEE*) estimates in consecutive ADP trials. (Notations are presented in the caption of Fig. 1).

The BOD POD software provides an estimate of the total energy expenditure (*TEE*) based on the measured %BF and the level of physical activity declared by the subject. Figure 5 shows the Bland-Altman plot of differences vs. means of *TEE*<sub>1</sub>

and  $TEE_2$  obtained in the first and second trial, respectively. The bias of 15.5 kcal is statistically different from zero.

#### DISCUSSION

In pairs of contiguous ADP trials, performed on a sample of healthy adults of diverse body compositions, we observed that body mass drops systematically from the first trial to the second by 68 g/h on the average. This value is consistent with the one reported by Benedict and Root in their study of alterations in body weight due to cutaneous and pulmonary evaporation (70 g/h) [2]. Nevertheless, the relatively large *SD* (59 g/h) indicates a large individual variability in *BM* alterations due to evaporation.

To estimate the impact of BM loss on %BF measurements by repeated ADP tests we first assumed that BV does not change in the time interval of about 10 min between two successive tests. Under this assumption, we estimated that %BF increased by 0.073% due to a BM loss of 11.3 g, observed, on the average, between two tests in our sample.

The Bland-Altman analysis, on the other hand, pointed out a mean difference between successive %BF estimates (bias) of 0.6%. In their vast study of the reliability of ADP, Noreen and Lemon reported a statistically significant difference between the mean BM recorded during test and retest [11]. Body density did not change significantly between successive tests, presumably because the drop in BM was accompanied by a drop in BV. The P values, however, were different (0.001 for BM and 0.08 for BV), indicating that the drop in BM was highly significant, whereas the change in BV was marginally insignificant. In our study, BM decreased significantly, whereas BV did not. On the contrary, the bias in volume measurements was negative (-81 mL) suggesting that BV increased from the first trial to the second. Consequently, we observed a statistically significant change both in BM and %BF.

This discrepancy between our results and those of Noreen and Lemon might originate from errors involved in BV measurements. Indeed, Dewitt *et al.* reported a precision of 72 mL for BV assessments by the BOD POD [7]. Due to errors in BV measurements, the precision of percent body fat assessments using the BOD POD was estimated to be about 0.8 %BF [5]. In the light of these works, it seems reasonable to conclude that the 0.6% bias in %BF found in our study stems mainly from measurement errors, which overwhelm the contribution of BM loss between tests.

Body volume measurements by ADP are subject to several sources of error, such as fluctuations of temperature and humidity in the measurement chamber due to unsteady breathing of the subject, or due to environmental changes. While environmental factors were carefully controlled in our experiments, the subject's behavior remained a source of biological variability in spite of the explanations given while obtaining the informed consent. Moreover, the measurements were influenced by the presence of body hair, a known source of error in ADP [8].

ADP is also used for assessments of food energy requirements of the human body. Although successive trials resulted in biases of *RMR* and *TEE* that were statistically different from zero, their values, of the order of 10 kcal, are small from the point of view of nutritional practice.

## CONCLUSIONS

Body composition assessments using the BOD POD are remarkably precise. They are mildly affected by the loss of body mass between consecutive trials, but the corresponding error is of the order of 0.1% BF, i.e. about 8 times smaller than the technical error of measurement of the BOD POD.

In conclusion, the reliability benefits of repeated measures protocols are practically unaffected by the drop of body mass incurred between successive trials as a result of pulmonary and cutaneous evaporation.

Acknowledgements. We thank Horia Hărăguş for helpful discussions on statistical methods used for evaluating reliability. The authors have not received any funds or benefits from the producers of the instruments used in this study.

#### REFERENCES

- 1. ANDERSON, D.E., Reliability of air displacement plethysmography, *The Journal of Strength & Conditioning Research*, 2007, **21**, 169–172.
- BENEDICT, F.G., H.F. ROOT, Insensible perspiration: Its relation to human physiology and pathology, Arch. Intern. Med., 1926, 38, 1–35.
- BLAND, M.J., D.G. ALTMAN, Statistical methods for assessing agreement between two methods of clinical measurement, *The Lancet*, 1986, 327, 307–310.
- BOSY-WESTPHAL, A., M. MAST, C. EICHHORN, C. BECKER, D. KUTZNER, M. HELLER, et al., Validation of air-displacement plethysmography for estimation of body fat mass in healthy elderly subjects, *Eur. J. Nutr.*, 2003, 42, 207–216.
- COLLINS, A.L., H.D. MCCARTHY, Evaluation of factors determining the precision of body composition measurements by air displacement plethysmography, *Eur. J. Clin. Nutr.*, 2003, 57, 770–776.
- 6. DEMPSTER, P., S. AITKENS, A new air displacement method for the determination of human body composition, *Med. Sci. Sports Exerc.*, 1995, **27**, 1692–1697.
- DEWIT, O., N.J. FULLER, M.S. FEWTRELL, M. ELIA, J.C.K. WELLS, Whole body air displacement plethysmography compared with hydrodensitometry for body composition analysis, *Arch. Dis. Child.*, 2000, 82, 159–164.
- 8. FIELDS, D.A., M.I. GORAN, M.A. MCCRORY, Body-composition assessment via air-displacement plethysmography in adults and children: A review, *Am. J. Clin. Nutr.*, 2002, **75**, 453–467.
- 9. GIAVARINA, D., Understanding bland altman analysis, *Biochemia Medica*, 2015, 25, 141–151.

- GREER, A.E., K.M. EDSALL, A.E. GREER, Reliability of bod pod measurements remain high following a short duration low-carbohydrate diet, *Int. J. Sport Nutr. Exerc. Metab.*, 2016, 26, 145–149.
- 11. NOREEN, E.E., P.W.R. LEMON, Reliability of air displacement plethysmography in a large, heterogeneous sample, *Med. Sci. Sports Exerc.*, 2006, **38**, 1505–1509.
- 12. PEETERS, M.W., Subject positioning in the BOD POD® only marginally affects measurement of body volume and estimation of percent body fat in young adult men, *PLoS One*, 2012, 7, e32722.
- SIRI, W.E., Body composition from fluid spaces and density: Analysis of methods, In: J. Brozek, A. Hanschel, (Eds.), *Techniques for measuring body composition*. National Academy of Sciences, Washington, DC, 1961, pp. 223–244.
- TUCKER, L.A., J.D. LECHEMINANT, B.W. BAILEY, Test-retest reliability of the bod pod: The effect of multiple assessments, *Percept. Mot. Skills*, 2014, 118, 563–570.