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Evaluation of the TOBEC method for calculating fat mass in Tree Sparrows *Passer montanus* and House Sparrows *Passer domesticus*

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Abstract. Total body electrical conductivity (TOBEC) is the name of a non-invasive method for investigating total body fat (TBF) in vertebrates. The error of measurement depends on body mass (for large animals the relative error is small), body shape and other factors.
The ACAN-2 apparatus operating on the basis of the TOBEC method shows integer numbers (readings) correlated with lean body mass (LBM). From the series of these readings (measurements) TOBEC can be calculated in many ways.
The error for LBM and TBF measurements in Tree Sparrows (of masses 22.5 ± 1.7 g) and House Sparrows (of masses 29.8 ± 2.0 g) was 1.19 g. This error may be reduced by repeating the TOBEC measurement and calculating the arithmetic mean of readings from the apparatus obtained 1 second after the commencement of measurement. Readings making up a single measurement series showed periodic irregular fluctuations of average amplitude 3 units in the case of Tree Sparrows and 5 units for House Sparrows — corresponding to errors of 0.5 g LBM in both species. Given individuals of both species were characterised by similar differences between the first and second TOBEC measurements. The TOBEC value obtained in a measurement during which a bird defecated in the chamber of the apparatus was significantly higher than that for a bird in a clean chamber. The orientation of the head in the chamber did not influence the repeatability of the TOBEC measurement. In Tree Sparrows, the relationship between TOBEC and LBM differed between those captured and held for one night prior to measurement and those measured for TOBEC immediately after capture.

Key words: body fat, Tree Sparrow, *Passer montanus*, House Sparrow, *Passer domesticus*, TOBEC

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INTRODUCTION

The total body electrical conductivity (TOBEC) method was first applied in estimating the fat mass of minced meat (Harker 1973). However, because it allowed for estimates of amounts of fat in live vertebrates, it soon found application in scientific research on: human adult males (Presta et al. 1983a, b) and infants (Cochran et al. 1989), pigs (Fiorotto et al. 1987, Cochran et al. 1989), rabbits (Fiorotto et al. 1987), rodents (Bracco et al. 1983, Piasecki et al. 1995, Koteja 1996, Zuercher et al. 1997), fish (Fisher et al. 1996) and birds (Walsberg 1988, Castro et al. 1990, Morton et al. 1991, Roby 1991, Scott et al. 1991, Skagen et al. 1993, Conway et al. 1994, Asch & Roby 1995, Lyons & Haig 1995, Staudinger et al. 1995, Burger 1997, Roby et al. 1997).

With assumptions based on the patent from Harker (1973), the TOBEC method allows the gross chemical composition of the body of an organism to be determined. The scientific basis for this is the fact that the dielectric constant (describing the electrostatic polarisation of a body in an electric field) and electrolytic conductivity are several tens of times

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lower in fat than in remaining body components. An apparatus operating on the basis of the TOBEC method is able to indicate a function for the lean body mass (LBM) of an organism, thereby allowing total body fat (TBF) to be calculated if total body mass (TBM) is known. However, as indications from the apparatus are influenced by an object's temperature (Scott et al. 1991), and shape (Fiorotto et al. 1987, Castro et al. 1990, Roby 1991), there is no universal formula which would allow LBM to be calculated for all animals as a function of TOBEC.

To determine FBM in some species the Harker patent recommends:

1) the choice of a population sample, for which we make TOBEC measurements and carry out gross chemical composition analysis;

2) the calculation of the linear regression for TOBEC against LBM coefficients and inversion of the formula;

3) the calculation of TBF as the difference between TBM and LBM (Morton et al. 1991).

Research into organisms of one species will not avoid errors in estimates of LBM and TBF from TOBEC measurements resulting from small differences in body shape. In animals with small amounts of fat these errors may reach 200% of real fat mass (Skagen et al. 1993, Conway et al. 1994, Zuercher et al. 1997). However, in such cases it is possible to make intraspecific comparisons using the TOBEC-estimated mean fat mass of an appropriately-large group of animals, especially where individuals in these groups do not differ in mean body mass.

The aim of the present study was to assess the reliability of the TOBEC method in the measurement of LBM and TBF in Tree Sparrows and in House Sparrows. To this end:

1) checks were made on factors linked to measurement technique that might influence the repeatability of results;

2) a regression of TOBEC against LBM was calculated for 21 Tree Sparrows and 48 House Sparrows in which gross chemical composition was determined, along with standard deviations of LBM assessed using TOBEC;

3) checks were made regarding the factors associated with measurement technique whose influence on the TOBEC/LBM regression was significant.

MATERIALS AND METHOD

The work was carried out in Dziekanów Leśny near Warsaw (52°20'N, 20°50'E). The two stages involved checks on the repeatability of TOBEC using measurements of the same bird and comparison of TOBEC and LBM in 69 birds for which gross chemical composition was analysed.

Repeatability of measurements

House and Tree Sparrows were mist-netted between January 1995 and March 1996. All were weighed to an accuracy of 0.01 g using electronic balances, measured for tarsus length to an accuracy of 0.1 mm and then subjected to TOBEC measurement using the ACAN-2 apparatus. This apparatus — operating on the basis of the TOBEC method — was constructed by Techmex International of Kraków (Poland), in co-operation with the Department of Biophysics of the Jagiellonian University (Froncisz et al. 1994, Piasecki et al. 1995, Koteja 1996, Zuercher et al. 1997). Indications of TOBEC may be compared with LBM without transformation. A further advantage is ease of use in field conditions. The ACAN-2 apparatus has a removable measuring chamber with a movable blockade to immobilise the animal studied. In our research, the diameters of the chamber were 33 and 35 mm for Tree and House Sparrows, respectively. The blockade had a scale from 1 to 6, with an increase of 1 corresponding to a 4 mm decrease in cylinder circumference.

Analysis of the repeatability of TOBEC measurements for the same individuals in the ACAN-2 apparatus involved 101 Tree Sparrows and 326 House Sparrows. Measurements were made in the following, uniform manner: an empty chamber was placed in the apparatus prior to each measurement to zero the indicator. The bird was then placed in the chamber, along its axis, dorsal side up, and the diameter of the cylinder reduced appropriately to the bird's circumference, such that it was immobilised. The number on the blockade scale was noted, along with the orientation of the bird's head (to the left or right or facing straight ahead). The occupied chamber was placed in the apparatus and 10 readings taken at 1-second intervals. Measurement was followed by a check for the presence of excreta in the chamber. The bird was then removed, and the measuring chamber ventilated or cleaned if necessary, before a second, or sometimes a third, measurement was made. Repeated measurements utilised the same blockade in all but 18 cases (in which there was a 1-point difference on the scale not found to influence the measurement). The ACAN-2 apparatus was not used during rain or periods of high humidity.

A consistent distinction was drawn between the terms "reading" and "measurement":

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— "reading" was the positive whole number indicated by the apparatus following placement of the chamber containing the bird;
— "measurement" entailed a series of readings at ca 1-second intervals noted following insertion of the chamber containing the bird.

The results of two successive measurements thus took the form of two series of numbers: $Y_{1,I}$, ... to $Y_{10,I}$ and $Y_{1,II}$, ... to $Y_{10,II}$ was adopted, where arabic numerals indicated consecutive readings and roman numerals successive measurements.

The designation $\bar{x}_i$ was adopted for the mean of the "i" (from first to i-th) readings calculated separately for first ($\bar{X}_{i,I}$) and second ($\bar{X}_{i,I}$) mesurement (eq., $X_{3,I} = 1/3\ (Y_{1,I} + Y_{2,I} + Y_{3,I})$.

The designations $Min_{i,\ Z}$ and $Max_{i,}$ were adopted for the minimum and maximum values among the "i" (from first to i-th) readings from the the first ($Min_{i,I}$, $Max_{i,I}$) and second ($Min_{i,II}$, $Max_{i,II}$) mesurement (eq., $Min_{5,I}$ is the minimum value among $Y_{1,I}$, $Y_{2,I}$, $Y_{3,I}$, $Y_{4,I}$, $Y_{5,I}$).

The 20 numbers: $Y_{1,I}$, $Y_{2,I}$, $Y_{3,I}$, $\bar{X}_{3,I}$, $\bar{X}_{5,I}$, $\bar{X}_{10,I}$, $Min_{5,I}$, $Min_{10,I}$, $Max_{5,I}$, $Max_{10,I}$,$Y_{1,II}$, $Y_{2,II}$, $Y_{3,II}$, $\bar{X}_{3,II}$, $X_{5,II}$, $X_{10,II}$, $Min_{5,II}$, $Min_{10,II}$, $Max_{5,II}$, $Max_{10,II}$, obtained and calculated in the TOBEC measurement for a given bird were termed the "TOBEC values" for that bird.

The difference between maximal ($Max_{10,Z}$) and minimal ($Min_{10,Z}$) readings obtained in the course of one measurement was termed the range of variation in readings.

Two measurements each were made for 83 Tree Sparrows and 255 House Sparrows in which unambiguous determination of the orientation of the head in relation to the axis of the apparatus chamber was possible (to the left, to the right or straight ahead). Thirteen House Sparrows and two Tree Sparrows defecated in the chamber in the course of a measurement. This led to the dirtying and moistening of plumage in the tail area and adjacent parts of the chamber. Defecation mostly occurred (in all but 3 House Sparrows) during the first measurement, thereby reducing mass by between 0.07 and 0.1 g.

Comparing tobec with gross chemical composition

In 1994 year 39 Tree Sparrows were captured, weighed, measured for tarsal length and TOBEC and then killed by anaesthetisation so that their gross chemical composition could be determined. The 18 Tree Sparrows caught at 18.00 on 30 Aug. were kept in a darkened cage overnight until 05.00, after which they were weighed (mean mass – 20.2 g) and subjected to TOBEC measurement. The remaining 21 Tree Sparrows caught at 16.00 were weighed and subjected to TOBEC measurements immediately after capture. TOBEC was read off only once and these measurements were not repeated. Immediately after TOBEC measurement the birds were killed, and taken for analysis of body composition by way of the Soxhlet Method with an ether solvent.

Birds were homogenised in a mortar following drying to constant mass at 60°C. Determinations of fat content were then made on three samples of homogenate from each. The error in the estimation of total body fat was most often below 3% (or 4% in 3 cases).

A further 48 House Sparrows were captured between 07.00 and 12.00 in January and December 1995, as well as in February 1996. These were immediately weighed and subjected to TOBEC determinations. The 15 caught in January 1995 were measured for TOBEC only once, while the 33 captured during the next winter were measured 2–4 times, being removed between measurements while the chamber of the apparatus was ventilated. The birds were then killed and prepared for analysis in the same way as the Tree Sparrows. The House Sparrows were dried to constant mass at a temperature of between 105°C and 110°C. They were analysed intact, except that breasts were cut to permit more rapid extraction. The dried corpses were subject to ether extraction, with the extracted fat being transferred quantitatively to test-tubes and the ether evaporated off to constant mass.

In line with the user instructions for the ACAN-2 apparatus, TOBEC is considered directly proportional to the LBM of the animal studied (Morton et al. 1991, Froncisz et al. 1994, Piasecki et al. 1995, Koteja 1996). Each species has certain constants C_1 and C_2, such that:

[1] $$Y = C_1 \times LBM + C_2$$

where LBM = fresh lean body mass and Y = TOBEC.

The constants C_1 and C_2 were calculated from the linear regression of TOBEC on LBM.

The LBM of the birds for which TOBEC measurements were made may be calculated from the formula:

[2] $$LBM = (Y - C_2) / C_1$$

Standard errors to the estimation of LBM were calculated, with the aid of formula [2], from the following formula:

[3] $$SE_{-r} = \left(\frac{1}{n-2} \sum_i \left(LBM_i - \frac{Y_i - C_2}{C_1} \right)^2 \right)^{\frac{1}{2}}$$

where LBM_i = fresh lean body mass of the i-th bird and Y_i the TOBEC value of the i-th bird.

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The 20 numbers comprising "TOBEC values" were calculated for each of the 33 House Sparrows of known body composition that had been subjected to two TOBEC measurements of 10 readings each. A check was made to determine which of the values was best correlated with the actual lean body mass.

The measurements of tarsi and noted blockade numbers were the only measured values to be linked with a bird's shape. We checked whether these data in multifactor correlation analysis would increase the significance of the correlation between fresh lean body mass and TOBEC. The Tree Sparrows analysed for body composition were confined by the same number blockade (5), so only two-factor correlation and regression analyses were calculated — between TOBEC and the linear combination of LBM and tarsal length. In turn, the correlations and regressions for House Sparrows were those between TOBEC and a linear combination of LBM, tarsal length, and blockade number.

A check was made for differences between the regression of TOBEC on LBM in the case of the 18 Tree Sparrows kept on the premises overnight and only then subjected to TOBEC measurements, and that obtained for the Tree Sparrows analysed directly after capture.

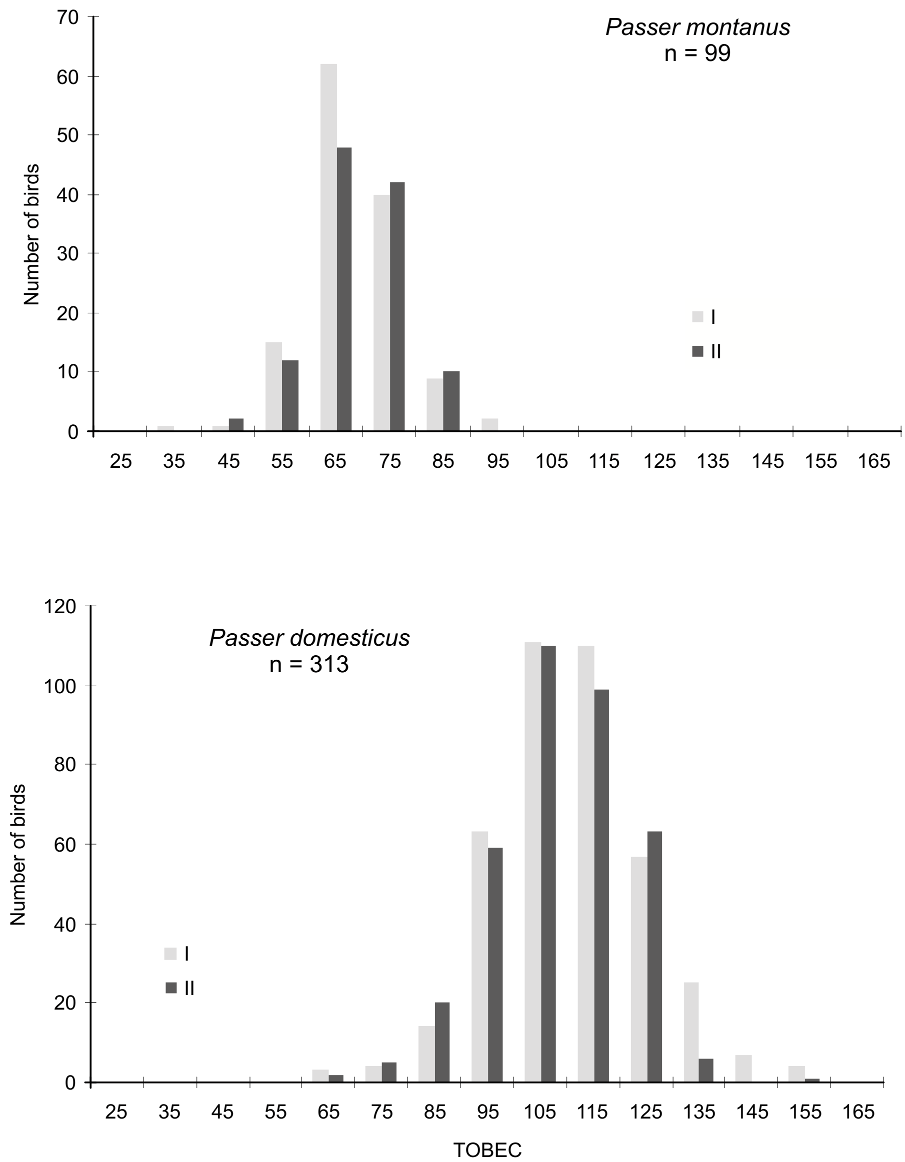

Fig. 1. Distribution of TOBEC readings ($Y_{1,I}$ and $Y_{1,II}$) obtained in two (I and II) measurements.

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RESULTS

Repeatability of measurements

ACAN-2 aparatus readings varied between 33 and 95 in Tree Sparrows, and between 55 and 187 in House Sparrows. The distributions of values for the two species did not depart significantly from the normal (Fig. 1, measurements made for birds which defecated were excluded). The mean readings from first measurements was 68.7 units for Tree Sparrows (SD = 8.5, median 69) and 111.2 for House Sparrows (SD = 15.9, median 111). The respective means from second measurements were 69.3 (SD = 7.9, median 70) and 110.5 (SD = 14.1, median 110).

Readings making up a single measurement showed periodic irregular fluctuations. The differences between maximal and minimal readings were in the range 0 to 11 (most often 2) in Tree Sparrows, and 1 to 16 (most often 5) in House Sparrows (Fig. 2, measurements made for birds which defecated were excluded). The ranges of variation in readings were greater in House Sparrows than in Tree Sparrows (K-S test: d = 5.01, p < 0.01).

For no "TOBEC values" did the probability of a first measurement being greater than a second exceed the opposite probability (of a second measurement being greater than a first). Absolute values for differences between "TOBEC values" in

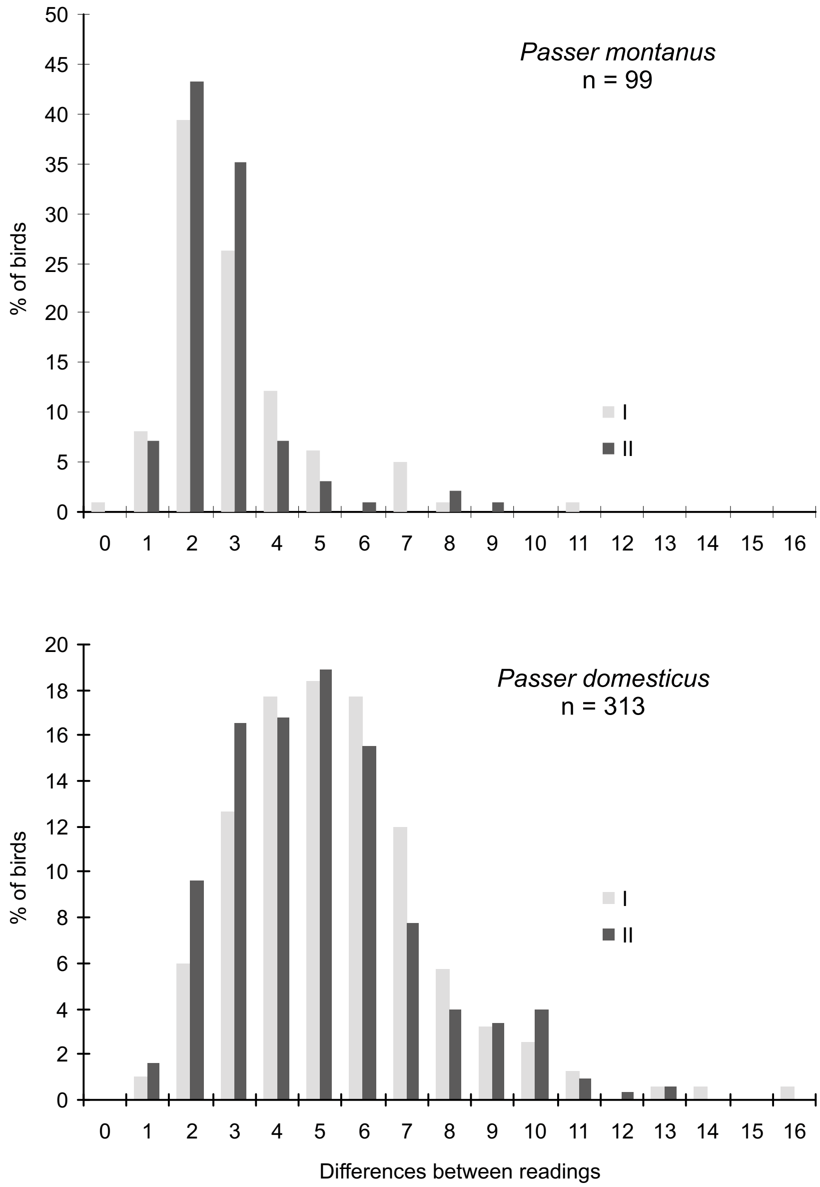

Fig. 2. Distributions of differences between maximum and minimum readings obtained during two (I and II) TOBEC measurements.

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the first measurement and corresponding values in the second measurement ranged from 0 to 14 in Tree Sparrows (average 3) and from 0 to 23 in House Sparrows (average 4).

A change in the position of the head within the chamber of the apparatus was not found to reduce or increase "TOBEC values".

Table 1. Body parameters of sparrows studied.

	Passer montanus (n = 21)		*Passer domesticus* (n = 48)	
	Mean ± SD	Range	Mean ± SD	Range
Total body mass [g]	22.49 ± 1.66	19.10–25.98	29.81 ± 1.96	26.42–34.08
Tarsus length [mm]	17.6 ± 0.82	16.0–19.0	19.5 ± 0.91	17.2–21.4
Total body fat [g]	1.108 ± 0.097	0.962–1.426	1.941 ± 0.360	1.178–2.731
Dry lean body mass [g]	6.289 ± 0.541	5.145–7.314	9.234 ± 0.768	6.679–10.906
Total body water [g]	15.09 ± 1.13	12.89–17.49	18.64 ± 1.31	15.58–21.41
Lean body mass [g]	21.38 ± 1.65	18.03–24.80	27.87 ± 1.80	23.974–31.349

In every case in which birds defecated in the apparatus chamber (2 for Tree Sparrows and 13 for House Sparrows), the "TOBEC values" was greater after soiling of the chamber. It is not very likely that this happened by chance (test of signs, $p < 0.01$). In relation to the "TOBEC values", the difference between these values reached 2–7 units in Tree Sparrows and 6–21 units in House Sparrows.

Relationship between TOBEC and lean body mass

The total mass of fat in Tree Sparrows (not kept overnight) was between 4.21 and 6.79% of body mass. The mean total mass of fat among House Sparrows was between 4.06 and 8.31% of body mass. The masses, tarsal lengths, and body compositions of these birds are presented in Table 1.

In line with expectations, any species showed correlation between TOBEC and TBF (Table 2). TOBEC was most correlated with LBM in House Sparrows, and with dry mass in Tree Sparrows (Table 2).

In the case of the 21 Tree Sparrows for which only one measurement with one reading was taken, the correlation coefficient between LBM and TOBEC was $r = 0.83$ (coefficient of determination $R^2 = 0.68$). The regression of TOBEC (Y) against LBM took the form:

[4] $Y = (4.71 \pm 0.74)LBM - (27.44 \pm 15.84)$

The standard deviation to estimates of LBM was 1.19 g.

For the 48 House Sparrows studied, the coefficient for the correlation between LBM and the first readings of the first TOBEC measurement ($Y_{1,I}$) was the same $r = 0.83$ (coefficient of determination $R^2 = 0.68$). The regression of $Y_{1,I}$ against LBM took the form:

[5] $Y_{1,I} = (5.48 \pm 0.55)LBM - (44.10 \pm 15.37)$

The standard deviation to estimates of LBM was 1.20 g.

Table 2. Correlations between TOBEC (first reading of the first measure) and body parameters for sparrows studied. * — $p < 0.001$.

Correlations with:	*Passer montanus* (n = 21)		*Passer domesticus* (n = 48)	
	r	ANOVA	r	ANOVA
Total body mass	0.84	*	0.78	*
Lean body mass	0.83	*	0.83	*
Total body water	0.81	*	0.80	*
Dry total body mass	0.87	*	0.54	*
Dry lean body mass	0.83	*	0.57	*
Total body fat	0.31	ns	0.14	ns

The regression of TOBEC against LBM obtained for House Sparrows (formula [5]) did not differ significantly from that obtained for Tree Sparrows (formula [4]) — the straight-line regression for one species was contained within the 95% confidence interval of the regression obtained for the other.

After combining sets of data for Tree Sparrows (one readout from a single measurement) and House Sparrows (first readout from first measurement — $Y_{1,I}$), the correlation coefficient between LBM and TOBEC was $r = 0.95$ (coefficient of determination $R^2 = 0.90$), The regression of TOBEC (Y) against LBM had the form:

[6] $Y = (5.42 \pm 0.22)LBM - (42.52 \pm 5.86)$

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Deriving from formula [6] are the following formulae for the calculation of LBM and TBF:

[7] $LBM = (Y + 42.52)/5.42$

[8] $TBF = TBM - (Y + 42.52)/5.42$

The standard deviation to estimates of LBM was calculated in accordance with formula [3] and amounted to 1.19 g. The maximal absolute value for the difference between the real LBM and that calculated from formula [7] was of 2.11g in the case of Tree Sparrows and 3.79 g in the case of House Sparrows (mean for the two species was 0.95 g).

In the case of the 33 House Sparrows for which 2 measurements of 10 readings each were made, lean body mass was most correlated with the mean from the first readings of the two TOBEC measurements (Table 3). The correlations between mean "TOBEC values" in the first and second measurements were higher than those for the first measurement and second measurement separately (Table 3). Standard errors to the estimations of fresh LBM estimated in accordance with formula [3] were equal to 1.06–1.12 g.

Table 3. Coefficients for the correlations between different TOBEC values and lean body mass (LBM) in 33 House Sparrows, along with SE_r [g] to the estimations. I, II — measurements.

TOBEC	I		II		Mean	
	r	SE_r	r	SE_r	r	SE_r
Y_1	0.81	1.32	0.80	1.50	0.87	1.06
Y_2	0.83	1.24	0.76	1.72	0.85	1.13
Y_3	0.82	1.28	0.78	1.61	0.85	1.11
$\bar{X}_3$	0.82	1.26	0.78	1.60	0.86	1.10
$\bar{X}_5$	0.82	1.28	0.79	1.58	0.86	1.10
$\bar{X}_{10}$	0.82	1.28	0.78	1.59	0.86	1.11
Max_5	0.81	1.35	0.77	1.64	0.85	1.14
Max_{10}	0.81	1.35	0.78	1.59	0.85	1.13
Min_5	0.83	1.24	0.80	1.51	0.86	1.06
Min_{10}	0.82	1.26	0.80	1.53	0.86	1.09

The regression for the mean of the first readings of the two measurements $Y_1 = 0.5\times(Y_{1,\,I} + Y_{1,\,II})$ against LBM took the form:

[9] $Y_1 = (5.92 \pm 0.61)LBM - (56.39 \pm 16.92)$

The standard deviation of the LBM estimate was 1.06 g.

In Tree Sparrows, the square of the correlation coefficient between the mean of the first TOBEC readings (Y) and the linear combination of LBM and tarsus length (TL) was $R^2 = 0.68$, while the equation for the multiple regression of TOBEC against LBM and TL was of the form:

In this equation, the slope coefficient for the regression relating to tarsal length did not differ significantly from.

[10] $Y = (4.13 \pm 0.84)LBM + (2.33 \pm 1.69)\times TL - (56.09 \pm 25.90)$

In this equation, the slope coefficient for the regression relating to tarsed lenght did not differ significantly from 0.

In House Sparrows, the square of the correlation coefficient between the mean from the first readings of the two TOBEC measurements and the linear combination of LBM, tarsal length (TL), and blockade number (BL) was $R^2 = 0.74$. The equation for the multiple regression of TOBEC (Y) against LBM, TL, and BL had the form:

[11] $Y = (5.63 \pm 0.59)LBM + (0.14 \pm 1.13)\times TL - (0.36 \pm 1.75)\times BL - (50.75 \pm 19.13)$

In this equation, the slope coefficients for the regressions against tarsal length and blockade number did not differ significantly from 0.

In the case of the Tree Sparrows kept overnight ($n = 18$), the correlation coefficient between TOBEC and LBM was only $r = 0.49$ (coefficient of determination $R^2 = 0.24$). For these birds, the regression of TOBEC against LBM (Fig. 3) was described by the equation:

[12] $Y = (1.51 \pm 0.54)LBM + (31.10 \pm 21.12)$

LBM estimated according to this formula differed from the real value by 1.70g on average (significantly more than in the case of the Tree Sparrows that were not kept overnight and using formula [4]: t test — $t = 2.24$, $df = 37$, $p < 0.03$), and maximally by 4.61 g.

The slope coefficient to the straight-line regression for confined Tree Sparrows (formula [12]) was significantly smaller than that for the birds not kept overnight (formula [4], $t = 2.850$, $df = 35$, $p < 0.01$, Fig. 3).

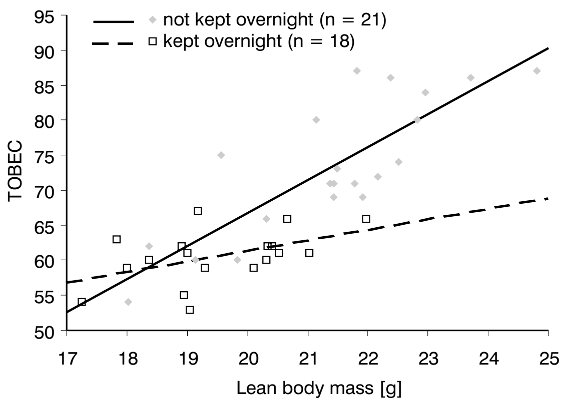

Fig. 3. Regression of TOBEC against LBM for Tree Sparrows which were or not kept overnight.

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In the case of confined Tree Sparrows weighing more than 20g, the attempt to estimate LBM with the aid of formulae [4] or [6] would have resulted in an error taking the form of an underestimation of real LBM (Fig. 3).

DISCUSSION

In small birds, fat is first and the most important reserve of energy allowing for the maintenance of constant body temperature and for the survival of the night or other breaks in feeding. Increased demands for energy in birds arise during the migration period, in winter, in the period of breeding and as young become independent. Thus the survival of birds at these times depends on the amount of fat accumulated (e.g. King 1972, Dolnik & Gavrilov 1975, Blem 1980, 1990, Schifferli 1980). On the other hand, an excess of fat reduces a bird's ability to fly and increases the risk that it will be taken by predators (Gosler et al. 1995). Calculation of the mass of fat in birds and the search for an *in vivo* method by which to estimate it fairly accurately are of importance in studying the condition of birds and estimating their probability of survival.

The TOBEC method is the best non-invasive way of measuring LBM and TBF in vertebrates. However, it depends on many factors, i.e. body shape (Fiorotto et al. 1987, Castro et al. 1990, Roby 1991) and body temperature (Scott et al. 1991). Studies of live homoeothermic animals of a given species, or species of very similar body shape, can by convention assume constant body temperature and shapes throughout a sample.

Many studies of TOBEC have been carried out using one individual to make several measurements. In estimating the amount of fat in people, Presta et al. (1983a) made 10 measurements and noted 10 readings over a 10-second period. They then took the mean of the total of 100 readings. Conway et al. (1994), working on the Wood Thrush *Hylocichla mustelina*, repeated TOBEC measurements 16 times, and recommended that an average of 9 measurements be made. Roby et al. (1997), and Zuercher et al. (1997), applied 6 repetitions of TOBEC measurements, while Burger (1997) used 7 measurements per bird. It was possible to conclude from the latter that a TOBEC measurement of the same individual is of limited repeatability irrespective of species and type of apparatus used.

In our studies, the repetition of TOBEC measurements were associated with average variations across a range of 3 or 5 units in the cases of Tree Sparrows and House Sparrows, respectively. Formulae [7] and [8] indicate that the former range corresponds to 0.55 g of fat (or 50% of total body fat), and the latter to 0.92 g of fat (47% of total body fat).

Best correlated with LBM in our work was the mean of first reading of TOBEC obtained ca 1 second after placing the bird in the apparatus (Table 3). Similar procedures have been applied in the measurement of much larger birds (*Calidris pusilla, C. fuscicollis*), with similar results being obtained (Skagen et al. 1993). Besides means of readings from repeated measurements, minimal values have also been taken into consideration (Zuercher et al. 1997). As our calculations show, these are better correlated with lean body mass than maximal values (Table 3).

This study found no relationship between the orientation of the head in the measuring chamber and the repeatability of TOBEC measurements. Castro et al. (1990) held that the mean coefficient of variability associated with position in the chamber was of 6.8%, while Roby (1991) gave a value of only 1.2%. Completely new findings concern the increase in TOBEC when a bird defecates into the measuring chamber, as well as the different relationship between TOBEC and LBM to be noted in birds kept caged overnight, as opposed to being analysed directly after capture (Fig. 3). The period of confinement was associated with a change in the body composition of birds (authors' unpublished data) — and probably also with changes in electrolytic conductivity properties — which brought about a reduction in the TOBEC value for Tree Sparrows weighing more than 20 g.

The correlations obtained here for LBM against TOBEC ($R^2 = 0.68$ for both species) were lower than those noted generally in the literature, where R^2 often exceeds 0.90. For instance Bracco et al. (1983) gave $R^2 = 0.99$ for the 50 Sprague-Dawley rats of masses 197–433 g, while Castro et al. (1990) reported $R^2 = 0.95$ for 38 analysed birds of masses 18–50 g, and Froncisz et al. (1994) had an $R^2 = 0.93$ for 22 mice of the species *Apodemus agrarius* and *A. flavicollis* weighing between 16 and 28 g. The cause was first and foremost the small range of variability noted for LBM among the birds studied (18.03–24.08 g in Tree Sparrows and 23.97–31.35 g in House Sparrows). In the case of the correlation between TOBEC and LBM for the both sparrow species combined (and hence a greater range of variability in LBM from 18.03 to 31.35 g), the R^2 value rose to 0.90. The relationship between the value for the TOBEC/LBM correlation and the range of variability in lean body mass of the analysed species of bird was also indicated

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by Lyons & Haig (1995), who found respective coefficients of determination of $R^2 = 0.92$, 0.79 and 0.35 for Short-billed Dowitchers *Limnodromus griseus*, Dunlins *Calidris alpina* and Semipalmated Sandpipers *C. pusilla*, with LBM values having ranges 70–170, 43–68 and 13–21 g respectively.

We obtained a positive correlation between TOBEC and the total body masses of birds, LBM, and TBW (Table 3). However, there was no significant relationship between TOBEC and TBF in either species of sparrow. The analogous correlations in birds twice as heavy as sparrows was much weaker, but also significant in the case of fat (Conway et al. 1994).

During the time in which the authors were making measurements using the ACAN-2 apparatus, works appeared discussing the limitations of the TOBEC method in regard to small lean birds and mammals of mass 20 g (Skagen et al. 1993, Conway et al. 1994, Asch & Roby 1995). For northern Red-backed Vole *Clethrionomys rutilus* weighing 14.31 to 40.26 g, but with small masses of fat (in the range 0.19 to 5.84 g), the mean percentage error for fat was of 120% (Zuercher et al. 1997).

The results obtained by us for House Sparrows are very similar to those from Asch & Roby (1995), who measured TOBEC in 25 House Sparrows using the SA-1 Small Animal Body Composition Analyser. With $R^2 = 0.54$, the conformity between their TOBEC measurements was lower than that obtained here for 33 House Sparrows using the ACAN-2 apparatus ($R^2 = 0.75$). At 1.33 g, the standard deviation to Asch & Roby's estimated LBM of House Sparrows was rather greater than that found by us (1.06 g).

Our results demonstrate that the present level of development of the TOBEC instrument tested here leaves the TOBEC method unsuitable for individual LBM and TBF measurements in the cases of House Sparrows and Tree Sparrows. The 1.19 g standard deviation associated with estimates of LBM from TOBEC is comparable with the total fat content in these birds. We believe that the TOBEC method may be a good instrument for calculating the mass of fat in other species of bird with masses similar to those of sparrows (20–35 g), but with a greater mass of fat (5 g on average). Indeed the method has proved its worth in the assessment of the fat mass (range 5–10 g) in Yellow-necked Mouse *Apodemus flavicollis*, weighing between 25 and 27 g (Froncisz et al. 1994).

The TOBEC method is a good technique for comparing mean masses of fat for a satisfactorily-large group of birds in which the calculation of standard errors for these means accounts for individual errors in assessing the masses of fat of particular individuals (cf. Burger 1997).

CONCLUSIONS

1) The error for the estimation of lean body mass (LBM) in Tree and House Sparrows was equal to 1.19 g when the TOBEC method was applied using ACAN-2 apparatus. This may be reduced by repeating the TOBEC measurement and calculating the arithmetic mean of indications from the apparatus obtained 1 second after the commencement of measurement.

2) The indications of the apparatus obtained c. 1, 2,..,10 second after commencement of measurement showed periodic irregular fluctuations of average amplitude 3 units in the case of Tree Sparrows and 5 units for House Sparrows — corresponding to errors of 0.5 g LBM in both species.

3) The first measurement differed from the second of the same bird on average differences between "TOBEC values" were equal to 3 units for Tree Sparrows and 4 for House Sparrows. There was the same probability that the first measurement was greater than the second as that the second was greater than the first.

4) Different orientations of the head had no influence on the repeatability of the TOBEC measurement.

5) The TOBEC value obtained in a measurement during which a bird defecated in the chamber of the apparatus was significantly higher than that for a bird in a clean chamber.

6) In Tree Sparrows, the relationship between TOBEC and LBM differed between those captured and held for one night prior to measurement and those measured for TOBEC immediately after capture.

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STRESZCZENIE

[Ocena TOBEC jako metody szacowania masy tłuszczu zawartego w ciele wróbli i mazurków]

TOBEC jest nieinwazyjną metodą pomiaru zawartości tłuszczu w ciele kręgowców opartą na patencie Harkera z 1973 roku. Aparaty działające w oparciu o tą metodę pokazują wartości pomiaru skorelowane liniowo z mokrą masą beztłuszczową (LBM) badanego zwierzęcia, co po dokładnym zważeniu go pozwala na określenie zawartości tłuszczu. Jednakże pomiar ten zależy od kształtu zwierzęcia i sposobu rozmieszczenia tkanki tłuszczowej w jego ciele, stąd dla każdego gatunku równanie liniowe, pozwalające na przeliczenie wskazań aparatu na ciężar mokrej masy beztłuszczowej, jest inne.

W badaniach sprawdzano przydatność metody TOBEC do oszacowania masy tłuszczu wróbla i mazurka przy użyciu aparatu ACAN-2 dostosowanego do badania niewielkich zwierząt (o masie od 15 do 100 g). Dla 101 mazurków i 326 wróbli przeprowadzono analizę powtarzalności pomiaru TOBEC. Wskazania aparatu przy pomiarze mazurków wynosiły średnio 68.7 jednostek, a wróbli domowych średnio 110.5 jednostek (Fig. 1). Pomiar TOBEC tych ptaków wykonywany był dwukrotnie. Przy każdym pomiarze notowano 10 kolejnych wskazań aparatu z częstością ok. 1 s. Wskazania te wahały się nieregularnie z amplitu-

dą równą średnio 2 jednostki dla mazurka i 5 jednostek dla wróbla (Fig. 2), co odpowiadało różnicy w oszacowaniu mokrej masy beztłuszczowej oraz tłuszczu o około 0.5 g. Różnicę pomiędzy dwoma kolejnymi pomiarami analizowano badając 10 różnych wielkości charakteryzujących pojedynczy pomiar (pierwsze, drugie i trzecie wskazanie aparatu, średnią z początkowych trzech, pięciu i wszystkich dziesięciu wskazań, maksymalne wskazanie z początkowych 5 i ze wszystkich dziesięciu wskazań oraz minimalne wskazanie z początkowych pięciu i wszystkich dziesięciu wskazań) (Tab. 3). Różnice miedzy pierwszym i drugim pomiarem wynosiły dla mazurka średnio 3 jednostki a dla wróbla domowego 4 jednostki. Ułożenie głowy nie miało wpływu na wynik pomiaru natomiast zanieczyszczenie komory pomiarowej kałem ptaka powodowało istotny i znaczący wzrost wartości wskazań aparatu.

W celu znalezienia równania pozwalającego na przeliczenie wskazań aparatu TOBEC na zawartość masy tłuszczu po pomiarze TOBEC 69 ptaków (48 wróbli i 21 mazurków) uśpiono i metodą Soxcleta oznaczono masę tłuszczu zawartą w ich ciele (Tab. 1). Korelacje wskazań aparatu z mokrą masą beztłuszczową wynosiły dla obu gatunków r = 0. 83 (Tab. 2). Korelacja ta była najwyższa gdy TOBEC wyznaczono jako średnią arytmetyczną z pierwszych wskazań aparatu (po 1 sekundzie od włożenia komory z ptakiem do aparatu) podczas obu pomiarów (Tab. 3). Równanie regresji TOBEC od mokrej masy beztłuszczowej mazurków nie różniło się istotnie od równania regresji TOBEC od mokrej masy beztłuszczowej wróbli. Można zatem do wyznaczania mokrej masy beztłuszczowej oraz tłuszczu u wróbli i mazurków używać tego samego wzoru [7] i [8]. Wykonano jeszcze dodatkowy eksperyment polegający na odłowie 18 mazurków, lecących na nocleg w godzinach przedwieczornych, przetrzymaniu ich przez noc, a nad ranem, po zważeniu i zmierzeniu TOBEC, ptaki uśpiono i oznaczono metodą Soxhleta masę tłuszczu w ich ciele. Ptaki przetrzymane przez noc różniły się od ptaków nieprzetrzymanych nie tylko składem ciała ale także równaniem zależności TOBEC od mokrej masy beztłuszczowej (Fig. 3). Wynika stąd, że innych wzorów umożliwiających oszacowanie mokrej masy beztłuszczowej należy używać dla ptaków, których TOBEC mierzono zaraz po złowieniu, a innych dla ptaków trzymanych kilka godzin w niewoli.

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