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Lab Fattening and Non-invasive Estimates of Body Composition in Deer Mice

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ABSTRACT--Total body electrical conductivity measurements and lipid composition were determined for the deer mouse (*Peromyscus maniculatus*) to derive species specific calibration equations for use with EM-SCAN estimates of lean and fat tissue. For each individual, total body electrical conductivity was measured by EM-SCAN, and actual lipid content was determined by chemical extraction. Then, using estimated and actual lipid values, separate calibration equations were generated for freshly captured (lean) and laboratory maintained (fat) individuals, and a combined equation was derived for all individuals. These equations were variable in the accuracy of lipid estimates; the lowest relative error estimate (percent body fat) was obtained with the equation for fat individuals while the highest error (percent body fat) was associated with the lean condition. Although high average error rates for lipid might preclude the use of this approach when absolute accuracy is necessary with lean individuals, estimates of lean tissue were very accurate regardless of body composition condition. When removed from the field and maintained in the laboratory, body composition changed significantly and quite rapidly with relative body fat doubling in six weeks. Thus, maintenance under laboratory conditions might affect physiologic and behavioral parameters in such subjects.

Key words: body composition, deer mice, laboratory fattening, *Peromyscus maniculatus*.

Non-invasive estimates of body composition have been conducted on a wide variety of vertebrates, including birds (Castro et al. 1990, Morton et al. 1991, Roby 1991, Scott et al. 1991, 1996, Skagen and Knopf 1993, Osborne et al. 1996), small mammals (Bachman 1994, Voltura 1997, Voltura and Wunder 1998), and fish (Fischer et al. 1996). In many cases, such studies have employed total body electrical conductivity (TOBEC) devices such as EM-SCAN (Voltura 1997, Voltura and Wunder 1998, Zuercher et al. 1999, Unangst and Wunder 2001) to estimate lean and lipid components of total body composition. Quantitative differences in divalent cations present in lean (fat free) versus lipid (fat) tissue enable the EM-SCAN device to estimate lean tissue with high precision and accuracy and fat tissue with lesser absolute accuracy (Walsberg 1988, Morton et al. 1991, Voltura and Wunder 1998, Zuercher et al. 1999, Wunder et al. 2000, Unangst and Wunder 2001). As recommended by the manufacturer and confirmed for mammals by Unangst and Wunder (2001), the accuracy and precision of the body-composition estimation are improved by developing species-specific calibration equations.

In addition, the change in body composition documented in wild animals

in all runs. In addition, an assessment of estimation accuracy between models was performed by comparing the average error estimate for each model.

RESULTS

The body composition of the 41 specimens used in equation derivation is summarized in Table 1. For all conditions, body mass was very close (lean 18.65 ± 0.67 g, fat 18.83 ± 0.51 g, combined 18.69 ± 0.41; $F_{2,37} = 0.74$, $P = 0.53$), however lipid mass was nearly four times greater in the fat (2.64 ± 0.24 g) versus lean (0.65 ± 0.03 g) data set ($P = 0.0002$). During the six week fattening period, the nine deer mice increased relative body fat from 8% to 14% ($P = 0.0004$), with an increase in total body mass of 1.2 g, primarily due to gains of 1.2 g in lipid mass and a relatively constant fat free mass (Table 2). Calibration equations for the EM-SCAN device to estimate either lipid or fat free mass of deer mice are presented in Table 3, with the absolute average error and relative average error for the multiple regression equations shown in Table 4. Estimates of fat free mass were strong with R^2 values ranging from 0.87 to 0.99 and error estimates for fat free mass ranging from 0.1 to 0.7 g. Lipid estimates were less accurate (R^2 varied from 0.53 to 0.69), but error estimates were strong and ranged from 0.3 to 0.7 g. In all cases, absolute average error estimates for both lipid and fat free mass were smallest in the lean condition and largest in the combined condition.

DISCUSSION

Our data indicated that the EM-SCAN provided a good estimation of body composition (fat and lean content) in deer mice with error estimates consistent with those of other rodent models (Zuercher et al. 1997, Voltura and Wunder 1998, Zuercher et al. 1999, Unangst and Wunder 2001). Errors in lipid estimates were smallest in the lean condition (0.29 ± 0.03 g) and largest in the combined condition (0.68 ± 0.07 g). Deer mice in the lean condition had an average fat mass of 0.65 ± 0.03 g, which equated to an average error of 45%. In contrast, the combined condition had body fat of 2.64 ± 0.40 g and an estimation error of 0.68 ± 0.07 g, which resulted in an average error of 35%. The most accurate average error for lipid was 19% in the fat condition. This was expected because with decreasing fat quantities, the average error will be larger because fat makes up an extremely small proportion of total body mass (less than 8%). Using a 0.3 g error in a hypothetical wild caught 20 g deer mouse with an actual fat content of 0.7 g would result in an increase in estimated total body fat from 3.5% to 5%. Although this high level of error might influence its use as an absolute measure, this deer mouse could still be classified as relatively lean. Such error rates might preclude the use of EM-SCAN to predict absolute lipid content on lean individuals but still could provide valuable information concerning relative body conditions. As individuals increased body fat, the average error in percent lipid improved with EM-SCAN and could be very meaningful. Error estimates for fat free mass were very similar to lipid but the average error was considerably less (1% to 4%) because the subject animals' total body mass ranged from 85% to 93% lean tissue. Thus, EM-SCAN more accurately estimated lean tissue, but still provided accurate and reliable estimation performance over a wide range of body composition conditions for both lean and fat tissue.

Table 1. Body composition comparison (mean ± 1SE) for deer mice used in model derivations for complete (n = 41), fat (n = 27) and lean (n = 14) data.

Condition	Body Mass (BM) (g)	Lipid Mass (LM) (g)	Fat-free Mass (FFM) (g)	Percent Body Fat (% of BM)	Percent Fat-free (% of BM)
lean	18.65 ± 0.67	0.65 ± 0.03	17.94 ± 0.66	3.50 ± 0.10	96.19 ± 0.21
fat	18.83 ± 0.51	2.64 ± 0.24	16.18 ± 0.36	13.62 ± 1.02	86.35 ± 1.03
combined	18.69 ± 0.41	1.96 ± 0.22	16.71 ± 0.36	10.14 ± 1.02	89.74 ± 1.01

BM = wet body mass, LM = lipid mass, FFM = fat free mass

Table 2. Change in body composition (mean ± SE) for fat deer mice over six week period (n = 9).

Week	Body Mass (BM) (g)	Lipid Mass (LM) (g)	Fat-free Mass (FFM) (g)	Relative Body Fat (% of BM)	Relative Body Fat (% of FFM)
0	17.99 ± 0.64	1.43 ± 0.18	16.56 ± 0.52	7.9	8.6
2	18.32 ± 0.82	2.27 ± 0.35	16.06 ± 0.61	12.4	14.1
4	19.46 ± 0.90	2.63 ± 0.40	16.82 ± 0.58	13.5	15.6
6	19.21 ± 0.83	2.60 ± 0.33	16.61 ± 0.59	13.5	15.6

BM = wet body mass, LM = lipid mass, FFM = fat free mass

Table 3. Calibration equations estimating lipid or fat free mass of deer mice for complete (n = 41), fat (n = 27), and lean (n = 14) data.

Condition	Equation*	R^2
Complete	[1]LM = -1.24 + 0.62M - 0.27CI	0.66
	[2]FFM = 1.27 + 0.37M + 0.27CI	0.87
	[3]FFM = CI - 0.56/1.82	0.77
fat	[1]LM = -1.61 + 0.55M - 0.20CI	0.69
	[2]FFM = 1.75 + 0.45M + 0.20CI	0.86
	[3]FFM = CI - 6.48/1.45	0.68
lean	[1]LM = 0.04 + 0.07M - 0.03CI	0.53
	[2]FFM = -0.04 + 0.92M + 0.03CI	0.99
	[3]FFM = CI - 3.87/2.07	0.78

*LM = lipid mass in g, FFM = fat free mass in g, M = wet mass in g, CI = conductive index, defined as (average EM-SCAN reading x body length)$^{0.5}$

[1,2] = multiple regression

[3] = inverse regression two stage

Table 4. Average error estimates (mean ± 1 SE) from cross validation for multiple regression models estimating fat free or lipid content on deer mice from complete (n = 41), fat (n = 27), and lean (n = 14) data.

Parameter	Condition		
	lean	fat	combined
Average error (g LM)*	0.29 ± 0.03	0.54 ± 0.09	0.68 ± 0.07
Average error (% lipid)	45	19	35
Average error (g FFM)*	0.11 ± 0.03	0.53 ± 0.09	0.67 ± 0.07
Average error (% fat-free)	<1	3	4

*LM = lipid mass, FFM = fat free mass

Derivation of separate calibration equations for each sample condition (lean, fat, combined) provided flexibility for the researcher to more closely match the experimental design. If a research design included only wild caught research animals or only laboratory maintained or laboratory reared animals, then the model specifically derived for a particular situation might be most appropriate. Overall, the utility of EM-SCAN to predict lean tissue was high; however, if precise estimates of lipid in lean individuals were necessary, then performance was less accurate.

The doubling of fat tissue in deer mice removed from the wild and maintained in the laboratory for only six weeks were consistent with those reported by Hayward (1965). In his study, six subspecies of wild *P. maniculatus* had between 2.7 and 9.1% body fat when expressed as a mean percent of fat free tissue. Using the same ratio in our study, the 14 deer mice in the lean condition, and the 27 deer mice at the start of the fattening experiment had 3.6 and 8.5% body fat respectively (Table 2). However, when kept in the laboratory, fat percent increased to 15.6% in our study compared to levels ranging from 11.6 to 52.2% after six months in the laboratory in Hayward's study (Hayward 1965). Such evidence confirmed that lipid deposition associated with laboratory maintenance in *Peromyscus* was significant and quite rapid. Thus, the body composition change from the lean, wild caught deer mouse to the laboratory fattened deer mouse in a short period of time cannot be discounted. These body fat increases might affect various physiologic and behavioral parameters and should be considered when designing studies of laboratory kept wild animals, especially if such results were to be extrapolated to field conditions.

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