Pellet Morphometry as a Tool to Distinguish Age and Sex in the Mule Deer

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By examining the morphometry (i.e., length, width, length-to-width ratio, and volume) of pellets in three different categories (adult males, adult females, and yearlings) mule deer (Odocoileus hemionus), we were able to distinguish via discriminant function and fuzzy clustering analyses the age and sex of these animals. To determine a priori the identity of the pellet samples and evaluate the accuracy of our methods, we obtained samples from individuals in captivity. The discriminant function allowed us to correctly assign 100% of adult males, 91.66% of adult females, and 75% of yearlings to an age class, using previous information. The fuzzy clustering method enabled us to correctly distinguish 100% of adult males, 83.3% of adult females, and 75% of yearlings. The methods are based upon different assumptions. An important assumption with the discriminant function method is that the membership of each pellet group must be established a priori. This may be a disadvantage in certain cases, such as when pellet samples are gathered for an indirect population assessment procedure. Despite this drawback, however, both methods appear to be highly accurate. Zoo Biol 23:139-146, 2004. © 2004 Wiley-Liss, Inc.

Key words: discriminate function analyses; fuzzy clustering analyses; pellet group; classification of age and sex; mule deer; *Odocoileus hemionus*

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INTRODUCTION

Dung pellets have a cylindrical form, consist of nonassimilated vegetal material, are compacted, and, in the case of mule deer, can be distinguished by color, shape, or position, depending on the social group of the animals that produced them [Aranda, 1981]. The use of pellets to infer ecological parameters in mammals may be an important tool for researchers.

Pellet group surveys have been used to estimate distribution, habitat use, and density in many deer populations [Neff, 1968; Collins and Urness, 1984; Ezcurra and Gallina, 1981; Leopold et al., 1984; Rowland et al., 1984; Gallina, 1990, 1994; Gallina et al., 1991; Alvarez-Cárdenas et al., 1999; Sánchez-Rojas and Gallina, 2000a, b].

Other studies have shown that it is possible to identify sex and age (with a 90% confidence level) for three different deer species: wapiti (*Cervus elaphus*) [Bubenik, 1982], moose (*Alces alces*) [MacCracken and van Bellenberghe, 1987], and white-tailed deer (*Odocoileus virginianus*) [Ezcurra and Gallina, 1981]. However, these findings have not been extended to other animals.

Sexual segregation (i.e., differences between adult males and females in habitat use) is a common phenomenon in deer populations [Bower, 1984; Main and Coblentz, 1990, 1996; Bleich et al., 1997]. Therefore, it is important to distinguish between males and females when characterizing habitat use.

The main objective of this study was to determine whether it is possible to determine the age and sex of mule deer (*Odocoileus hemionus*) using the morphometric attributes of pellets. Our main assumptions were that the age of individuals is an indirect indicator of body size, and that pellet shape is sex-specific [Bubenik, 1982]. To investigate this, we sampled pellet production in captive animals, and explored multivariate statistical tools (the discriminant function and fuzzy clustering analyses) as a means of recovering population data from indirect information.

MATERIALS AND METHODS

Pellets were obtained from captive mule deer, which were divided into three categories: bucks (adult male, >3 years old), does (adult female, >3 years old), and yearlings (<1.5 year old). To maximize the variance for the measurements of the pellets, and to increase sample size, samples were obtained from three different sources: the Pocatello Idaho Zoo; the wildlife experimental enclosure, School of Natural Renewable Resources, University of Arizona, Tucson; and the Zoo of Centro Ecológico de Sonora, Mexico. Mule deer from these locations are recognized as the subspecies *hemionus* [Hall, 1981].

Pellets were collected in a single day at each location, shortly after the animals defecated. Consequently, we had a priori information regarding the origin of each pellet group. From each pellet group we randomly chose 15 individual pellets. Each pellet was measured to obtain the following information: maximum length (L), maximum width (W), length-to-width ratio $(\frac{L}{W})$, and the volume (V) estimated as $V = \pi(\frac{W}{2})^2 L$, where $(\frac{W}{2})$ is the radius and L is the height. To avoid any interobserver bias, all measurements were made by the same observer using a digital calliper (Mitutoyo[®]) with 0.01 accuracy.

We obtained the mean, median, standard error, and coefficient of variation for each variable, for each fecal group. The mean and median values were plotted for each variable, and the degrees of overlap among age/sex classes were examined. We used analyses of variance (ANOVAs) to investigate possible differences among groups.

To resolve the problem of separating two or more groups of items given several variables, we used the step-by-step discriminant function analysis [Manley, 1994]. This test enabled us to 1) determine whether differences arise between the mean values of variables among the age and sex classes; 2) identify the variables responsible for these differences; and 3) develop a classification function, based on the discriminant variables, to accurately assign questionable pellet groups to particular age and sex classes.

We also used a fuzzy set clustering technique to approach the same problem (pellet allocation among age/sex groups). We compared the efficacy of this technique with that of the discriminant function analysis, which is not applicable without a priori group information.

In ordinary set theory, an element does or does not belong to a particular set (an indicator variable can be associated with each element to assign its membership status, taking a value of one if the element is a member and zero if it is not). The idea is extended to allow the indicator variable to have any real value within the interval (0,1). In this way, elements have a degree of membership, and the set, the boundaries of which are thus not sharply defined, is termed "fuzzy." To obtain the value of membership for each class, we used the fuzzy k-means algorithm, which is based on minimizing the within-group sum-of-square J_m (U, V, A), which is given by

$$J_m(U, V, A) = \sum_{i=1}^n \sum_{j=1}^c (U_{ij})^m (d_{ijA})^2$$
(1)

where

$$(d_{ijA})^2 = x_i - v_j|_A^2 = (x_i - v_j)^T A(x_i - v_j)$$

is a measurement of distance calculated as a norm-induced metric, **A** is the norminducing matrix (any matrix of dimension p, where p is the number of attributes considered), m is the "fuzziness parameter" ($1 \le m \le \infty$), x_i is the vector attribute measurement made on an individual i (i=1,2,...,n), the vector v_j is the "center of cluster" j (j=1,2,...,c), j, u_{ij} is the membership value of individual i on the cluster j, **U** is the membership matrix, and **V** is the matrix of the cluster center ($U = \{u_{ij}\}$ and $V = \{v_j\}$) [Equihua, 1990, 1991].

Equation [1] defines the clustering techniques isodata and k-means [Saber, 1984] when m=1. The partition is hard when m=1, and becomes fuzzier as m approaches ∞ [Bezdeck, 1984]. Although m may have a wide range of values, it has been found, purely on the basis of empirical results, that values of ~ 2 produce satisfactory results (see Equihua [1990] and references therein for details).

This algorithm is used in the Fuzzy Clustering Tool program [Equihua, 2000], which provides the value of membership of each individual pellet group. We decided to assign each pellet group to a specific age and sex class based

on the maximum value of membership to some class (adult male, adult female, and yearling) to accurately assign hard-to-classify pellet groups to a specific age and sex class.

RESULTS

We collected 26 different pellet groups: six samples from bucks (90 pellets total), 14 samples from does (210 pellets total), and eight samples from yearlings (120 pellets total). However, in this last category, two females (1.5 years old) were at the age limit between adult and yearling.

The highest values across all variables in the samples were for adult males, the lowest were for yearlings, and the intermediate values were for females. Pellet shape did not significantly differ between sexes. However, the yearling pellets were more rounded than the adult pellets. As such, the value of ratio $\binom{L}{W}$ was smaller than in the case of adults (Table 1), although the differences was not significant.

Each measurement showed a coefficient of variation of 5-23%, which indicated a variance from small to medium among the individual pellet groups. All of the variables for volume showed a low degree of overlap among classes, and the width had the lowest coefficient of variation value.

We decided to use the median instead of the mean in both methods because the former is less sensitive to extreme values (measurements of individual pellets can be affected by various factors, such as environmental humidity, the kind of food consumed by the animal, and the position and place of the pellet when it is excreted). Using the median for each measurement as a factor in the stepwise discriminant analysis, the estimated U (Wilks' Lambda) was 0.076, which approximates an F (4, 44) of 28.9. This value suggests a significant difference between age/sex classes (P < 0.0001). We found that volume and width explained 89.9% of the variation in the data.

Using the discriminant classification function (Table 2), we were able to identify correctly all adult males, 91.66% of adult females, and 75% of yearlings. To use the discriminant coefficient classification, we multiplied the value of the median of the two significant variables (volume and width) by the corresponding coefficient for each class. This product is the sum of the constant, and the result is a discriminant value for each sex and age class.

The fuzzy k-mean algorithm estimated the value of the membership for each pellet group and plotted the value of membership of the two classes, in this case the value of female membership and the value of yearling membership. With this technique, we were able to identify the age and sex category for each pellet (see Fig. 1). Furthermore, we were able to correctly assign 100% of adult males, 83.33% of adult females, and 75% of yearlings.

DISCUSSION

We were able to show significant differences in pellet dimensions (volume and width) among adult males, adult females, and yearlings. These results are in agreement with previous studies of other deer species [Ezcurra and Gallina, 1981; Bubenik, 1982; MacCracken and van Bellenberghe, 1987].

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de Sono	Young $(N=8)$ rank	SD	0.39	0.14	0.03	0.06	N, samj
ológico (Yoı	Σ̈́	10.77	7.25	0.45	1.49	riance;
in capti ntro Eco		Μ	10.66	7.22	0.44	1.48	nt of va
l the Ċe		CV	0.07	0.02	0.16	0.07	coefficie
ona, and	rank	Min	11.35	6.58	0.41	1.26	e; CV, (
l young of Arizo	N = 14)	SD Max Min	13.74	9.33	0.93	1.89	um valu
iversity	emale (1	SD	0.34	0.10	0.03	0.05	minim
dult fem urce Uni	Adult female ($N = 14$) rank	χ	12.43	7.81	0.61	1.63	e; Min,
ole Reso		Μ	12.17	7.83	0.60	1.60	um valu
Renewal		CV	0.12	0.07	0.22	0.12	maxim
TABLE 1. Description of measurement of pellets from adult males, adult females, and young individuals kept in captivity from the Pocatello Zoo (Idaho), the wildlife experimental enclose School of Natural Renewable Resource University of Arizona, and the Centro Ecológico de Sonora	Adult male $(N = 6)$ rank	Min	13.49	9.53	1.08	1.21	n; Max,
		Max	16.87	11.32	1.44	1.76	leviation
		SD	0.79	0.30	0.12	0.08) dard
		χ	15.75	10.13	1.28	1.57	SD, star
escripuol xperimen		Μ	15.7	10.1	1.26	1.54	, mean;
the wildlife e.			Length (L)	Width (W)	Volume	Ratio: L/W	M median; $\bar{\chi}$, mean; SD, standard deviation; Max, maximum value; Min, minimum value; CV, coefficient of variance; N, sample size.

TABLE 1. Description of measurement of pellets from adult males, adult females, and young individuals kept in captivity from the Pocatello Zoo (Idaho), the wildlife experimental enclose School of Natural Renewable Resource University of Arizona, and the Centro Ecológico de Sonora	
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	Age and sex classes					
Measurement	Adult male	Adult female	Young			
Width Volume Constant	29.1839 -0.1129 -78.4019	$\begin{array}{r} 46.6721 \\ -0.2485 \\ -108.9146 \end{array}$	50.3467 -0.2779 -121.6607			

TABLE 2. Discriminant classification function coefficient for the mule deer pellet groups

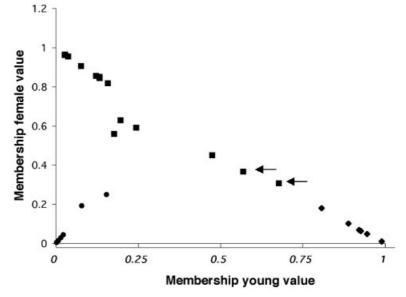


Fig 1. Values of membership of female, and young according to the fuzzy clustering algorithm. Squares represent female, circles are bucks and rhombi are young animals. The two squares with an arrow indicate two young at the age limit (1.5 years).

However, since we did not include all possible gradients of age in the sample, the medium-sized male pellet may have fallen within the female pellet category. It is important to emphasize, however, that this was not the case for bucks or younger animals.

Dimensional differences among pellet groups in relation to sex may be attributed to body size differences [MacCracken and van Ballenberghe, 1987]. This is particularly true for the mule deer, as this species exhibits a strong sexual dimorphism (i.e., males are heavier than females [Anderson and Wallmo, 1984]).

The values of the variables between sexes show a degree of overlap, but this is low in the case of volume and width variables with significant values from the stepby-step discriminant analysis. According to these measurements and the statistical procedure, adult males showed the highest values, followed by adult females and then yearlings. The fuzzy set analysis was more sensitive and was able to detect the two females that were at the limit of their age class. Although they were originally misclassified in the yearling category, the results were similar to those obtained with the discriminant classification method. Nevertheless, both methods were able to recover, from each fecal group, most of the age and sex categories. However, each method employs different assumptions: the discriminant function method requires, at least at the calibration stage, that the age or sex of origin of each pellet group be known a priori, which is not the case with the fuzzy set method. This is especially important for studies in which pellets are collected from animals of unknown age or sex. In such cases, the fuzzy set method should prove to be a useful tool.

The ability to determine the age and sex structure of a mule deer population based on pellet dimensions has important implications for management and conservation strategies. The use of pellet counts to estimate deer density would be more relevant if we could classify them according to age and sex classes. This could be especially important when the animals are difficult to track because it is difficult to establish visual contact, such as when an individual is under very dense vegetation, or a population has a very low density. The statistical methods we have examined here have additional advantages: 1) they are inexpensive; 2) they do not disturb the animals; 3) with a good sample design, they can provide valuable information about the sex ratio and the age structure of the population); and 4) they can provide information, such as regarding habitat use, by different age and sex classes and thus help researchers assess the animals' ecological preferences.

CONCLUSIONS

1. We found differences in the morphometry of pellets among bucks, does, and yearlings in captive mule deer.

2. The discriminant function and fuzzy clustering analyses were both highly accurate in detecting these differences. However, some loss of information can not be avoided, and thus the estimate produced should be regarded as only a gross approximation.

3. The discriminant function method requires, at some point, a priori knowledge of the age or sex of the animal producing the pellet, which is a disadvantage.

4. The classification of pellets using these tools optimizes the information gathered by pellet survey methods, because it allows us to obtain a gross estimate of demographic parameters and to identify differences in habitat use between sexes.

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