COMPLEXITY OF BEHAVIOURAL SEQUENCES AS A MEASURE OF PHYSIOLOGICAL STRESS: ASSESSMENT OF A TECHNIQUE

Alejandro Frid
Boreal Research Associates, Site 20, Comp. 357, RR 1, Whitehorse, YT,
Y1A 4Z6, Canada, email: afrid@yknet.yk.ca

and

Stuart Alexander
Alexander Ecological Services, 14 Oak St., Whitehorse, YT, Y1A 4B1,
Canada, email stuarta@yknet.yk.ca

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INTRODUCTION

Our primary objective was to evaluate the technique developed by Alados et al. (1996), which uses complexity of vigilance-feeding sequences as a measure of physiological stress in free-living animals. According to these authors, their method can quantify stress from mere observations of feeding behaviour. By eliminating the need to capture and handle animals, it is potentially useful for testing predictions on how helicopter disturbance stresses wildlife (Frid 1996).

Alados et al. (1996) tested their method with Spanish ibex (*Capra pyrenaica*). They found that the vigilance-feeding sequences of individuals that were pregnant or infected with parasites were significantly less complex, or more predictable, than those of unstressed individuals.

Their analyses were based on fractal theory, which provides a metric for complexity called fractal dimension. Of the analytical techniques that they used, spectral analysis was most applicable to this study because it is more likely to yield useful results when applied to relatively short samples of behavioural sequences (Alados et al. 1996; C. L Alados pers. comm.). Spectral analysis is the decomposition of wave patterns into component sine and cosine waves, one for each possible frequency within the time period of the sequence. The slope of the plot of each component wave's power (squared amplitude) against frequency (both transformed to $\log_e$) provides a measure that is inversely related to fractal dimension. That is, a steeper slope implies lower behavioural complexity in the behavioural sequence. Lower complexity of behaviour has been inferred to reflect greater physiological stress in the animal (Alados et al. 1996).

The use of behavioural complexity, or unpredictability, as an indicator of stress is based on the assumption that complexity is evolutionarily advantageous. In the case of vigilance, a behaviour that is largely aimed at looking for predators, unpredictability presumably increases the difficulty for a predator to remain undetected while attempting to approach potential prey (Quenette & Desportes 1992, and references cited within). The complexity of these patterns seems to break down when physiological stress alters biochemical processes in the nervous system (Alados et al. 1996, and references cited within). According to Alados et al., it is this break down of behavioural complexity that can be measured by the slopes of power spectrums.

Even though Alados et al. published their results in a leading refereed journal for behavioural studies, we had several apprehensions about applying their method. The first was that the technique is in its infancy and, as far as we are aware, it has been used only in their one study. Second, the analysis is mathematically complex and potentially subject to computational quirks. Third, it is best if samples of behavioural sequences are at least 30-min long (CL Alados pers. comm.), and large sample sizes are usually needed for any analysis of vigilance, which implies considerable field effort. Finally, independently of physiological stress, vigilance decisions are usually affected by myriad factors related to predation risk, food supply, and sociality (reviewed in Elgar 1989). Alados et al (1996) did not address whether any of the latter factors would have short-term effects on the complexity of vigilance-feeding sequences (Quenette &

Desportes 1992), and we wondered whether not controlling for at least the most important of these factors would confound interpretations, or at least reduce statistical power.

Thus, before considering using the technique of Alados et al (1996) in new studies designed to test predictions on effects of helicopter disturbance on wildlife (Frid 1996), we analyzed observations on Dall's sheep (*Ovis dalli dalli*) already available to us. These data did not contain samples of disturbed animals, but by analyzing them we hoped to answer the following questions:

1) Can our results be interpreted similarly to the results of Alados et al.? In other words, do vigilance-feeding sequences that appear less complex in descriptive plots actually have power spectrums with steeper slopes? And if so, do lactating females have power spectrums with steeper slopes than those of females without young?
2) Does complexity of vigilance-feeding sequences respond to short-term changes in environmental and social factors related to predation risk which are known to affect conventional measures of vigilance time budgets?
3) If complexity does respond to these factors, how does the response differ from that of the percent of time spent vigilant by foraging animals?

In the process of answering the latter question, we gained insights which have implications for sampling conventional measures of vigilance time budgets. The latter are useful for quantifying effects of human disturbance on the foraging efficiency of wildlife (Berger et al. 1983; Stockwell et al. 1991; Frid 1996).

METHODS

Defining concepts and terms related to vigilance

Vigilance, which in ungulates and other terrestrial vertebrates is recognized by the head-up position, is largely aimed at looking out for predators. This behaviour, however, is generally mutually exclusive with feeding (reviews in Elgar 1989 and Lima & Dill 1990; Illius & FitzGibbon 1994). While less vigilant individuals are more likely to be taken by a predator (FitzGibbon 1989; Krause & Godin 1996), individuals that are too vigilant will forage less efficiently. Thus, making decisions that optimize the trade-offs between vigilance and feeding has been a major evolutionary force shaping the behaviour of prey (reviews in Elgar 1989 and Lima & Dill 1990). As a result, vigilance responds to risk-related factors, such as group size, distance to refuge, and predator presence (review in Elgar 1989), and can respond to interactions between factors (Frid 1997).

The predation risk factors that we analyzed are known to affect vigilance time budgets (either vigilance rate or percent time spent vigilant) as follows:

1) Vigilance decreases as group size becomes larger (review in Elgar 1989), but this relationship can be strongest when animals are far from a refuge, and can be weakest when animals are near a refuge (Frid 1997). Increasing group size reduces predation risk because it dilutes the probability of an individual being preyed on, increases the number of sensory organs for

detecting a predator, and increases confusion for a predator trying to focus on individual prey (review in Roberts 1996a).

2) Vigilance increases as distance to a refuge becomes larger (review in Elgar 1989), but this relationship can be strongest when animals are in very small groups, and weakest when animals are in very large groups (Frid 1997).
3) Vigilance decreases as distance to obstructive cover (tall dense vegetation which could hide a predator) becomes larger (review in Elgar 1989; Lazarus & Symonds 1992).
4) Mothers with young offspring are more vigilant than females without young or with older offspring (e. g. FitzGibbon 1993; Burger & Gochfeld 1994).

Study site, seasons and animals

Observations on adult female Dall's sheep were made during the last part of the 1995 lambing season (18 May to 5 June) at Killermun Lake, southwest Yukon. Details on this population can be found in Frid (1995).

Data were collected on lactating individuals, which had lambs that were < 5 weeks old, and on non-lactating adult females (which had no lambs). For the latter, the ratio of barren to pregnant individuals was unknown. Also, observations were made when plant phenology was advancing rapidly, and animals were experiencing fast changes in food quality and density.

Natural predation risk was certainly present. Five species of carnivores (including one raptor) known to prey on sheep were found in the study site, and several interactions between predators and sheep were observed. See Frid (1995) for details.

Collecting behavioural data

Data on vigilance-feeding sequences were collected with continuous recording of focal individuals (reviewed in Martin & Bateson 1993). These records were timed to the nearest second. A notebook computer (TRS-80 Model 100) programmed as an event recorder was the recording medium. To minimize disturbance to the focal animal and potential predators, observers used spotting scopes and were over 500 m from focal animals.

Simultaneously with the continuous recording, a second observer recorded the focal animal's group size, and distances to the nearest cliffs and nearest obstructive cover with an instantaneous scan (review in Martin & Bateson 1993) at the start of the sample and every subsequent 3 min. These measurements were averaged to describe the general conditions that may have affected focal animal's vigilance. To estimate distances, observers used known reference points on the landscape (e. g. sheep torso lengths, flagging, the length of cliff bands) and a 1:50,000 topographic map. During scans, observers also recorded whether an animal's position in the group was central or peripheral. When group size was less than five or when group geometry was linear, all animals were classed as peripheral. Cliffs were defined as in Frid (1997). For non-lactating and lactating individuals, respectively, group sizes ranged from 1-

35 and 3-35, distances to cliffs ranged from 1-750 m and from 1-250 m, and distances to obstructive cover ranged from 0-750 m and from 0-1000 m.

Recording sessions began when sheep were either handling or searching for food, and ended after 6-min or when animals stopped handling food plants for 1 min. We assumed that the latter criteria limited sampling to animals that were feeding intensively, and that individual differences in short term hunger caused little variability of vigilance (Krebs & Kacelnik 1991). Although the shortest samples used for analyses were about 3 min long (one sample was 1 min long), 89 % of samples were $\geq$ 5 min.

If lactating individuals nursed with their head up during observations, the sample was either eliminated or truncated to delete this behaviour and the portion of the sample that preceded or followed it (depending which option gave the longest sample). Our criteria here was that an animal nursing with its head up is effectively vigilant, but the decision to stop feeding was not made by her, but rather by the lamb requesting to suckle.

Sheep were not marked. To minimize the potential problem of individuals contributing more than one observation to the data set (Machlis et. al 1985), we considered observations to be biologically independent only if they involved individuals that could be temporarily distinguished by their position in the landscape or if they occurred on different days.

Observations in which the vigilance of focal animals was potentially affected by social dynamics, the presence of predators (mainly golden eagles [*Aquila chrysaetos*]), or aircraft were excluded from analyses. Sample sizes were insufficient for analyzing these factors.

Analyzing complexity of vigilance feeding sequences

According to Alados et al. (1996), the complexity of a vigilance-feeding sequence is inversely related to the slope (β) of the log-log (base e) power spectrum from a discrete Fourier transform of the 0-1 sequence (0 = feeding, 1 = vigilant; in this study, measured with a sampling frequency of 1 second over an interval of n seconds). Following Hastings & Sugihara (1993), a Fourier transform decomposes the wave pattern of a sequence of n observation points into a set of sine and cosine waves, one for each of n possible frequencies. The Fourier transform returns a complex number

$$z = x + iy$$

for each frequency (f), from which the amplitude of the corresponding wave is calculated as

$$|z| = r = (x^2 + y^2)^{1/2}.$$

The power $E(f)$at a given frequency is the squared amplitude of the wave at that frequency. The power spectrum is a plot of power against frequency f, over the interval

$$0 < f < n/2.$$

With discrete Fourier transforms, power values are symmetrical about the mid-point of the set of sequences (Hastings & Sugihara 1993:70). Frequency values can be nominal (e.g., 1, 2, 3, ...). Least-squares regression was used to estimate the slope (β) of the log-log transformed (base e) power spectrum, such that for some constant k

$$E(f) = kf^{-b}$$

Fourier transforms were performed using the *cfft* function in Mathcad Plus 5.0 for Windows (MathSoft Inc. 1994). This function has the advantage that it works on samples of any length. Many computer programs provide a fast Fourier transform function (e.g., Microsoft Excel, Systat), that requires the interval n to be a power of 2. Regression slopes (β-values) were calculated with Mathcad's *slope* function.

Statistical analyses

Multiple regressions were used to analyse the independent (additive) and interactive (multiplicative) effects of predation risk factors on the slopes (β) of power spectrums, and on percent time spent vigilant. The independent factors analyzed were group size, distance to cliffs, distance to obstructive cover, and reproductive status (a dummy variable with 1 = lactating, 0 = non-lactating). Position in group is also known to have a strong effect on vigilance (Berger & Cunningham 1988; review in Elgar 1989; Burger & Gochfeld 1994), but most samples (83 % for lactating individuals, 94 % for non-lactating individuals) consisted of peripheral individuals and position in group was not analyzed nor controlled. (In most cases group geometry did not allow the possibility for animals being central.)

Transformations were used to meet the assumptions of normality and homoscedasticity (Kleinbaum & Kupper 1978; McCullagh & Nelder 1983; Zar 1984). Diagnostic tests were used to confirm the success of transformations and to check other assumptions of regression (Kleinbaum & Kupper 1978; Wilkinson 1990).

Regression models were reduced to their most significant form with backward step-wise procedures (Kleinbaum & Kupper 1978; McCullagh & Nelder 1983; Wilkinson 1990). Stepping was non-automated and the criteria for removing or re-entering variables into the model were based both on significance values of 0.05 and on tolerance values. (Tolerance is 1 minus the multiple correlation between a predictor and the remaining predictors in the model. Setting a minimum tolerance value prevents the construction of highly multicollinear models.) Variables could not become part of the reduced model unless their tolerance was greater than 0.1 (Wilkinson 1990).

Prior to regression, Pearson correlation matrices were used to look for correlations between independent variables. Correlated variables were excluded from analyses.

Meaningful power spectrums can be generated only from true behavioural sequences, not from single behavioural events. Thus, for analyses of β-values of power spectrums, samples with single or no vigilance events were excluded from analyses.

RESULTS

For brevity, throughout the remainder of this report "β-value" refers to the slope of the log-log plot of the power spectrum (see Methods).

Behavioural complexity in relation to β-values

A visual inspection of power spectrums and descriptive plots (Fig. 1) suggested that intervals between vigilance events were not any less predictable for sequences with the lowest β-values than for sequences with the highest β-values. Furthermore, durations of vigilance events seemed *more* complex (less predictable) for sequences with the greatest β-values than for sequences with the lowest β-values. In other words, β-values appeared to be *directly* related to the unpredictability of durations of vigilance events, and to not be related to the unpredictability of intervals between such events. This visual inspection, which needs to be corroborated with some other measure of complexity (e. g. Roberts 1994, but see Kramer & Bell 1996 and Roberts 1996b), cautions that our results may not interpretable in the same way as those of Alados et al..

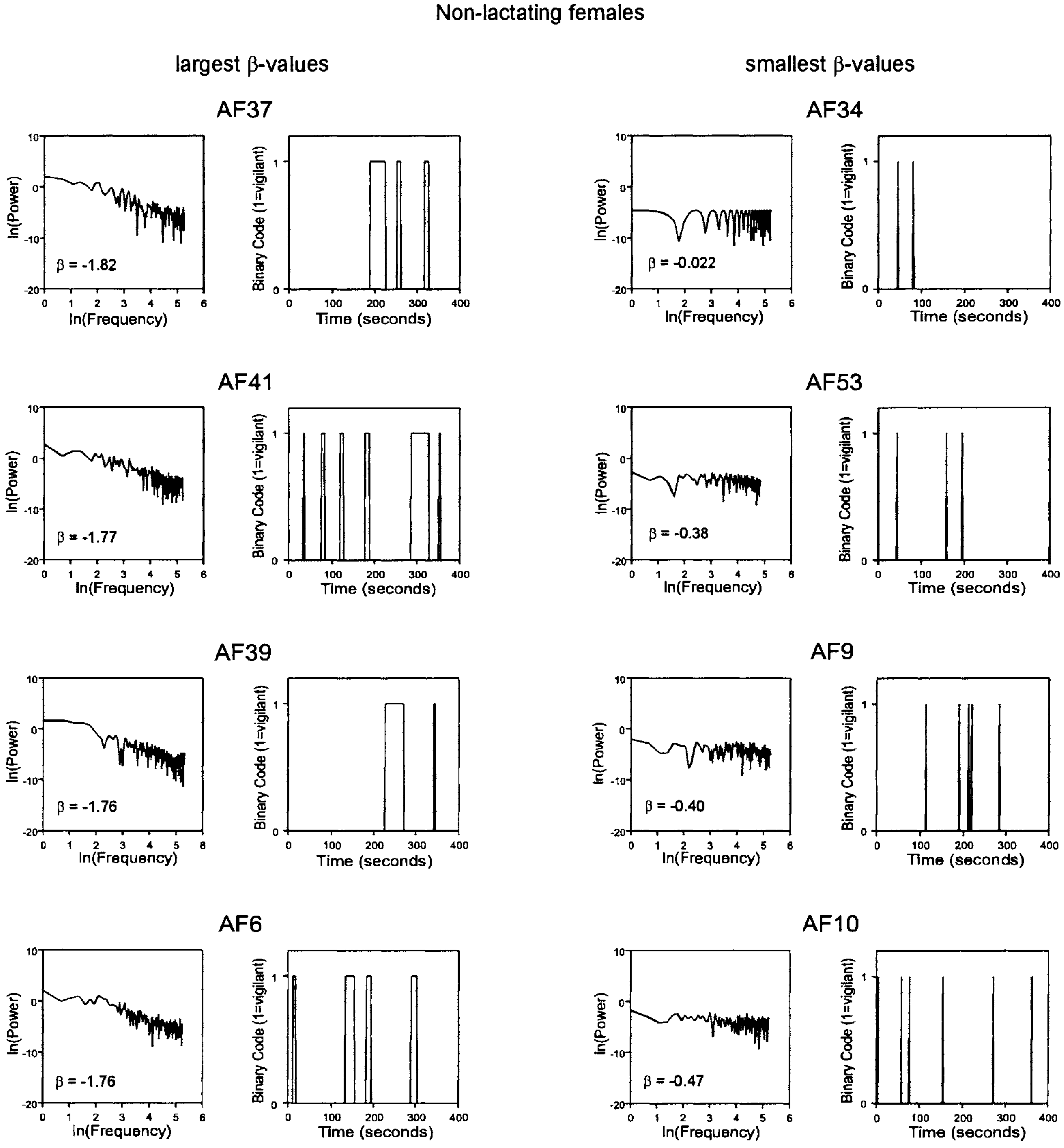
Reproductive status and β-values

Our analyses did not detect any differences in complexity of vigilance-feeding sequences between lactating (N = 49) and non-lactating individuals (N = 48) (mean ± SD of β-values, respectively, -1.19 ± 0.43, -1.21 ± 0.38; T = 0.31, DF = 95, P = 0.76). This lack of difference may have been partly due to the confounding effects of other variables known to affect conventional measures of vigilance time budgets, such as group size, distance to cliffs, and distance to obstructive cover. All such variables, however, were correlated with reproductive status, precluding statistical controls through multiple regression.

Comparing responses of β-values and of time budgeting to short-term changes in predation risk

For non-lactating individuals, the percent of time spent vigilant was affected by the interaction of group size and distance to obstructive cover, but was unaffected by distance to cliffs. Although there was considerable variation in the relationship ($R^2 = 0.13$), animals became less vigilant as group size increased, but the response was stronger for animals that were farther from obstructive cover (Table 1; Figs. 2 and 3). In contrast, none of the predation risk factors analyzed affected the β-values of vigilance-feeding sequences. This was the case when, prior to being reduced, the regression model included group size, distance to cliffs, and their interaction ($R^2 = 0$, all F's ≤ 1.80, all P's ≥ 0.19, N = 48), and when it included group size, distance to obstructive, and their interaction ($R^2 = 0$, all F's ≤ 2.76, all P's ≥ 0.10, n = 48). (The two regressions were necessary because distance to obstructive cover and distance to cliffs were correlated [r = -0.68, P <0.001], and could not be tested in the same model.)

The percent of time spent vigilant by lactating individuals was unaffected by group size, distance to obstructive cover, and the interaction between these



Non-lactating females

largest β-values

smallest β-values

AF37

AF34

AF41

AF53

AF39

AF9

AF6

AF10

Fig. 1a. Examples of descriptive plots of vigilance-feeding binary sequences (0=feeding, 1=vigilant) and corresponding power spectrums for non-lactating female Dall's sheep. Left-hand panels represent the sequences with the largest absolute β-values (steepest power spectrums) while right-hand panels represent the sequences with the smallest absolute β-values (flattest power spectrums) in the data set.

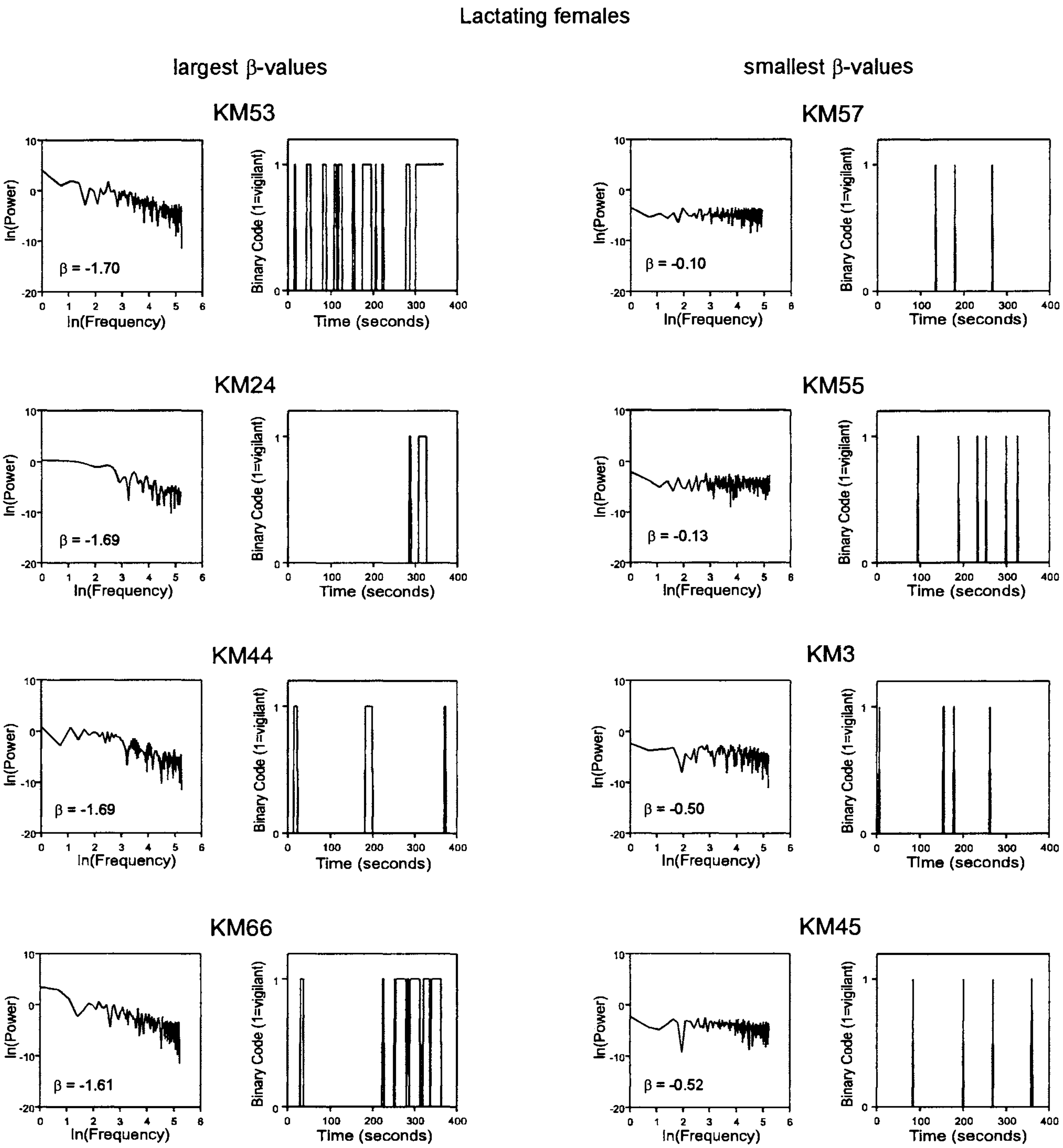


Fig. 1b. Examples of descriptive plots of vigilance-feeding binary sequences (0=feeding, 1=vigilant) and corresponding power spectrums for lactating female Dall's sheep. Left-hand panels represent the sequences with the largest absolute β-values (steepest power spectrums) while right-hand panels represent the sequences with the smallest absolute β-values (flattest power spectrums) in the data set.

Fig. 2. Estimated response of % time vigilant to the interaction of group size and distance to obstructive cover for non-lactating females. The broken line represents sheep that are 5 m from obstructive cover. The solid line represents sheep that are 50 m from obstructive cover. Regression lines are generated from the reduced model in Table 1a ($R^2 = 0.13$).

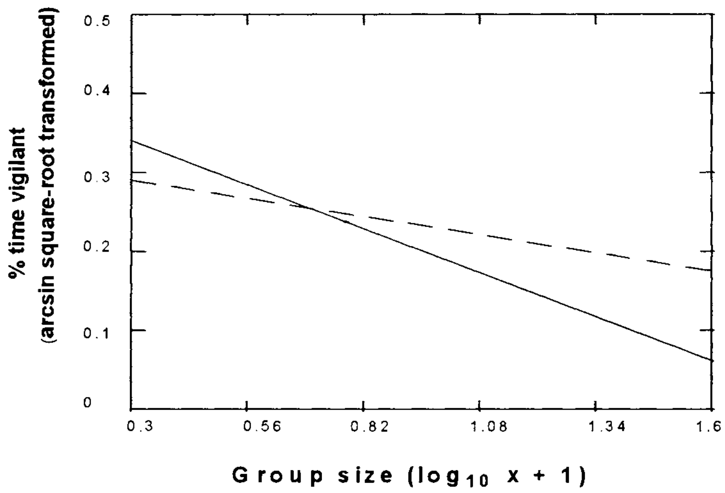


Fig. 3. Examples of the actual response of vigilance by non-lactating females to the interaction of group size and distance to obstructive cover. Black squares represent sheep that were farther than 10 m from obstructive cover. Clear squares represent sheep that were within 10 m of obstructive cover. Because continuous, three-dimensional data had to be categorized to be shown in two dimensions, these examples are only for descriptive purposes; see Fig. 2 for the statistically significant trends.

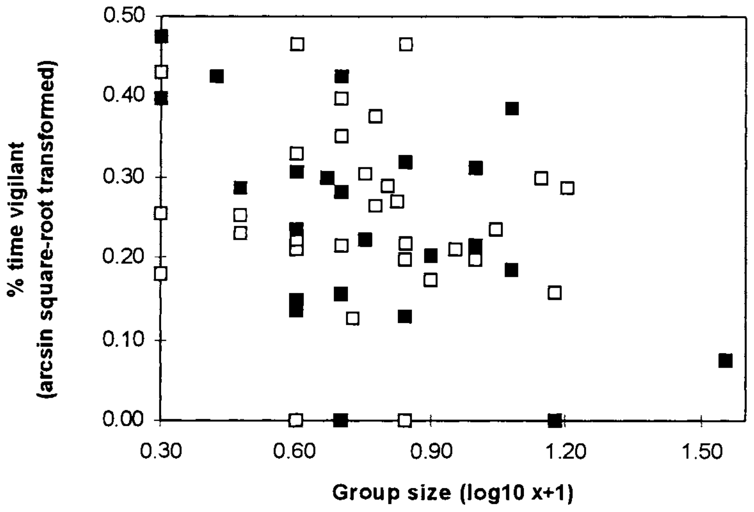


Table 1. Reduced multiple regression models analysing % time spent vigilant (arcsin square-root transformed) by foraging non-lactating individuals. Distance to cliffs and distance to obstructive cover were correlated (r = -0.68, P <0.001) and could not be included in the same model.

Table 1a. Reduced model that, prior to stepping procedures, included group size, distance to obstructive cover, and their interaction

Variable ($\log_{10}$ transformed)	Coefficient	Tolerance	T	P
constant	2.55		9.27	<0.001
distance to obstructive cover	0.088	0.27	2.052	0.045
group size X distance to obstructive cover	-0.126	0.27	2.72	0.009

ANOVA summary: R^2 = 0.13, F = 3.8, DF = 2, 51, P = 0.028

Table 1b. Reduced model that, prior to stepping procedures, included group size, distance to cliffs, and their interaction.

Variable ($\log_{10}$ transformed)	Coefficient	T	P
constant	0.35	7.12	<0.001
group size	-0.15	-2.32	0.024

ANOVA summary: R^2 = 0.09, F = 5.38, DF = 2, 52, P = 0.024

variables ($R^2 = 0$, all F's ≤ 0.86, all P's ≥ 0.36, $n = 59$). (Distance to cliffs was excluded from analyses because it was correlated with group size [$r = 0.38$, $P = 0.003$] and with distance to obstructive cover [$r = -0.61$, $P = <0.001$]). In contrast, absolute β-values decreased as group size increased, but there was considerable variation in this relationship ($R^2 = 0.09$; Table 2).

DISCUSSION

Are β-values inversely related to complexity?

If a time series can be modelled by a fractal process, then the slope of the power spectrum should be inversely related to the fractal dimension of the process (Hastings & Sugihara 1993). Alados et al. (1996), following others cited therein, interpreted a higher fractal dimension (i.e. a lower β-value) as indicative of higher behavioural complexity. A visual inspection of descriptive plots, however, suggested that our data appeared to yield β-values that were *directly* related to unpredictability of duration of vigilance events, and which appeared to be unrelated to predictability of the intervals between such events. The discrepancy is not a computational artifact. We sent samples of our raw data to C. L. Alados, who kindly took the time to analyze them herself. The β-values she calculated were very similar to ours. Unfortunately, she offered no explanation for why we found a relationship which is opposite to that expected. Obviously, an alternative measure of complexity (e. g. Roberts 1994, but see Kramer & Bell 1996 and Roberts 1996b) is needed to corroborate our visual interpretation of descriptive plots.

One important difference between the data of Alados et al. (1996) and our data is sample duration. By being only 3-6 min long, our samples had a maximum of about 360 data points (one for each second), while those analyzed by Alados et al (1996) had 1024 to 2048 data points. We do not know whether our samples were too short to produce meaningful power spectrums.

Do β-values reflect the physiological stress of lactation?

Our analyses did not detect any effect of reproductive status on β-values of vigilance-feeding sequences, which is inconsistent with the findings of Alados et al (1996). Although these authors did not study lactating animals, they did find, significant differences between pregnant and non-pregnant animals. Furthermore, they obtained these results without controlling for any predation risk factor known to affect vigilance time budgets.

The discrepancy between their results and ours could be due to a number of factors. First, as described above, our samples may have been too short to produce meaningful power spectrums. Second, for non-lactating individuals we do not know the ratio of pregnant to barren individuals. Given that late pregnancy increases energy requirements for female ungulates by 40 % (Loudon 1985), there may have been substantial variation in the body condition of non-lactating individuals. Third, the age of lambs ranged from less than a

Table 2. Reduced multiple regression model analysing β-values of vigilance-feeding sequences by lactating female. Prior to being reduced, the model included group size, distance to obstructive cover, and their interaction. Distance to cliffs was excluded from analyses because it was correlated with obstructive cover ($r = -0.57$, $P < 0.001$) and with group size ($r = 0.42$, $P = 0.003$)

Variable ($\log_{10}$ transformed)	Coefficient	T	P
constant	-1.74	-6.30	<0.001
group size	-0.52	2.12	0.04

ANOVA summary: $R^2 = 0.09$, $F = 4.48$, $DF = 1, 46$, $P = 0.04$.

week to perhaps 5 weeks, and younger lambs impose greater lactation demands on the mother (Shackleton & Haywood 1985). Thus, variability in lamb age may have increased variability in the condition of lactating individuals. Fourth, plant phenology was rapidly advancing during the 3 weeks of sampling, which likely increased temporal variability in the body condition of both reproductive classes.

β-values and % time vigilant in relation to predation risk factors.

One result suggested that β-values, similarly to vigilance time budgets, can respond to predation risk factors. For lactating individuals, increasing group size brought a decrease in absolute β-values. In contrast, the percent time spent vigilant by this reproductive class was unaffected by predation risk factors. For non-lactating individuals, β-values were unaffected by predation risk factors, but the percent of time spent vigilant was affected by the interaction between group size and distance to obstructive cover. These results suggest that although complexity of vigilance-feeding sequences can respond to short term changes in predation risk, its sensitivity could differ from that of conventional measures of vigilance time budgets.

Some insights on vigilance time budgets

Although the focus of this project was on vigilance complexity, not time budgeting, some comments on the latter are warranted. These insights are important because time budgeting of vigilance also has applications for studying the effects of disturbance on wildlife (Berger et al. 1983; Stockwell et al. 1991; Frid 1996).

For adult females without young, animals became less vigilant as group size increased, but this response was stronger for animals that were far from obstructive cover. The decrease of vigilance with group size has been found in a large number of vertebrate studies, and is consistent with the interpretation that animals in larger groups are under less predation risk (review in Roberts 1996a). Why this relationship was weaker when animals were near obstructive cover likely was unrelated to predation risk. We suggest that near the shrubs and trees that create obstructive cover, microclimates may have been more favourable for forage growth (warmer temperatures and lower desiccation due to wind). Also, during focal animal samples sheep fed on *Salix* shrubs, which made up most of the obstructive cover and whose leaves emerged during the study, for an average of 8 % of observation time. Thus, food density and quality might have been greatest in or near obstructive cover. Previous studies have found that feeding rate increases with food density, which in turn results in a decrease on the amount of time allocated to vigilance (review in Elgar 1989). Thus, near obstructive cover the effects of group size may have been diluted by the effects of food density.

The percent of time lactating individuals spent vigilant did not respond to any predation risk factors. The lack of a group size effect, which is almost ubiquitous across vertebrates (Elgar 1989; Roberts 1996a), is particularly puzzling. It is also surprising that, the vigilance of non-lactating females did not decrease in response to greater distance to cliffs, as has been found by other sheep studies (Risenhoover & Bailey 1985; Frid 1997). Possible explanations for these negative results, and which may also account for the large amount of unexplained variability in significant regression models, are as follows:

1) Theory predicts that animals in poorer condition make lower investments in antipredator behaviour (McNamara & Houston 1987). As explained in the previous section, differences between pregnant and barren individuals, as well as changes in plant phenology and lactation demands, likely increased variability in body condition, which may have increased variability in vigilance.
2) Plant phenology may have brought rapid changes in food density and quality, which, may have increased variability in vigilance (review in Elgar 1989).
3) Older offspring are less vulnerable to predation than younger offspring, and mothers decrease their vigilance as their offspring become older (FitzGibbon & Lazarus 1995). Thus, variability in lamb age likely increased variability in the vigilance of lactating individuals.
4) Behavioural samples lasted only 3 to 6 min, and the results may have been partly an artifact of short sample lengths. Vigilance is a relatively rare event, and it may be that the shorter the sample, the more inaccurate the estimate. We explored this possibility by taking the data of Frid (1997), truncating his original 13-15 min samples down to 6 min, and reanalyzing the data with stepwise regression (as done for this study) Truncating the sample durations did not affect which factors were significant in the reduced model, but did reduce the R^2 value from 0.24 to 0.18 (Appendix A). This result suggested that, though probably a contributing factor, sample duration likely was not the main explanation for lack of significant results in our data. Furthermore other studies of ungulate vigilance obtained strong results with sample lengths of only 1 min (Burger & Gochfeld 1994) and 3 min (Berger & Cunningham 1988).
5) Statistical power (the probability of detecting a significant effect) increases with sample size. Given all the sources of variability discussed above, plus others reviewed by Elgar (1989), any effect on vigilance of Dall' s sheep is bound to be small, and sample sizes may have been insufficient to detect the smaller effects. Unfortunately, power analysis (see Thomas & Juanes 1996) could not be done within the time constraints of this contract.

CONCLUSIONS AND RECOMMENDATIONS

Alados et al. (1996) used fractal dimensions as measures of complexity of vigilance-feeding sequences to quantify stress in free-living ungulates. Because such measures can be obtained from mere observation, eliminating the need to capture and handle animals, the technique can potentially be very useful. In our assessment of the technique, however, we found several complications.

Our principal results on how β-values relate to complexity of vigilance-feeding sequences, and on how this behavioural complexity relates to physiological stress (as determined by reproductive status), were inconsistent with those of Alados et al. (1996). Although the contrast between studies may have been due to sampling artifacts (mainly short durations of our focal animal samples), our results do suggest that their method requires further testing before it can be considered to have general applicability. One of our results (absolute β-values of lactating individuals decreased with group size) also suggested that behavioural complexity may be affected by short-term changes in predation risk, which could complicate interpretation of results, as well as increase the need for statistical controls.

On the basis of these conclusions, we make the following recommendations:

1) Do not invest substantial field effort in applying the method of Alados et al. to studies of human disturbance on wildlife until the technique has been tested and developed further. The technique, however, could be very useful, and we do recommend doing the latter. Some critical tests would be to compare β-values with measures of physiological stress obtained directly from blood or urine and feces samples (see Miller et al. 1991).
2) We do not know whether the apparent direct relationship between β-values and complexity was an artifact of our short samples. Alados et al. (1996) obtained meaningful results with 17 min samples, and the 13-15 min samples of Frid (1997) likely will provide a more useful test of their technique. Thus, we recommend using the data of Frid (1997) to continue assessing the relationship between complexity and β-values. As part of this process, β-values should be compared to other measures of behavioural complexity (e. g. Roberts 1994, but see Kramer & Bell 1996 and Roberts 1996b).
3) For statistical tests involving β-values in this report and in any future work with the data of Frid (1997), power analyses should be used to differentiate between statistical and biological significance (see Thomas & Juanes 1996).
4) Until further testing determines the general applicability of β-values as measures of stress, conventional measures of vigilance time budgets, (e. g. % time vigilant), which are substantially easier to compute and which require less field effort (they provide useful data with shorter sample lengths), could be used to test predictions on how disturbance affects foraging efficiency (Berger et al. 1983; Stockwell et al. 1991; Frid 1996). The latter could also be complementary to analyses of β-values.
5) For analyses of β-values and of % time vigilant, we suggest that stronger results will be obtained if sampling is restricted to periods or situations in which rapid changes in plant phenology, demands of lactation, and vulnerability of lambs are unlikely to increase variability in the data.

LITERATURE CITED

Alados, C. L., J. M. Escos, & J. M. Emlen. 1996. Fractal structure of sequential behaviour patterns: an indicator of stress. Anim. Behav. 51: 437-433.

Berger, J., D. Daneke, J. Johnson, S. H. Berwick. 1983. Pronghorn foraging economy and predator avoidance in a desert ecosystem: implications for the conservation of large mammalian herbivores. Biol. Conserv. 25: 193-208.

Burger, J., & M. Gochfeld. 1994. Vigilance in African mammals: differences among mothers, other females, and males. Behaviour 131:153-169.

Elgar, M. A. 1989. Predator vigilance and group size in mammals and birds: a critical review of the empirical evidence. Biol. Rev., 64, 13-33.

FitzGibbon, C. D. 1989. A cost to individuals with reduced vigilance in groups of Thomson's gazelles hunted by cheetahs. Anim. Behav. 37: 508-510.

FitzGibbon, C. D. 1993. Antipredator strategies of female Thomson's gazelles with hidden fawns. J. Mamm. 74:758-762.

FitzGibbon, C. D. & Lazarus, J. 1995. Anti-predator behaviour of Serengeti ungulates: individual differences and population consequences. In: Serengeti II: Research, Management and Conservation of an Ecosystem. (Ed. by A. R. E. Sinclair & P. Arcese), pp. 274-296, Chicago: University of Chicago Press.

Frid, A. 1995. Dall's sheep of the Killermun Lake region: ecological and behavioural data in relation to mineral exploration. Yukon Fish and Wildlife consultant report TRC-95-2. (Available from Jean Carey, sheep and goat biologist, Department of Renewable Resources, Whitehorse, Yukon. email jcarey@yknet.yk.ca).

Frid, A. 1996. Hypotheses and preliminary experimental designs for investigating impacts of helicopter disturbance on Dall's sheep. Unpublished report to the Yukon Fish and Wildlife Branch. (Available from Jean Carey, Department of Renewable Resources, Whitehorse, Yukon. email jcarey@yknet.yk.ca).

Frid, A. 1997. Vigilance by female Dall's sheep: interactions between predation risk factors. Anim. Behav. (In press).

Hastings, H.M., & G. Sugihara. 1993. Fractals: A user's guide for the natural sciences. Oxford University Press, Oxford, UK.

Ilius, A. W. & FitzGibbon, C. 1994. Costs of vigilance in foraging ungulates. Anim. Behav., 47, 481-484.

Kleinbaum, D. G. & L. L. Kupper. 1978. Applied Regression Analysis and other Multivariable Methods. Boston: Duxbury Press.

Kramer, M., & W. R. Bell. 1996. Comments on predictabilility and overdiffercing in vigilance sequences. Anim. Beh. 51:1175-1178.

Krause, J. & J. G. J. Godin. 1996. Influence of prey foraging posture on flight behavior and predation risk: predators take advantage of unwary prey. Behav. Ecol. 7: 264-271.

Krebs, J. R. & Kacelnik, A. 1992. Decision making. In: Behavioural Ecology. (Ed by J.R. Krebs & N.B.Davies), pp. 105-137, Oxford: Blackwell Scientific Publications.

Lazarus, J. & Symonds, M. 1992. Contrasting effects of protective and obstructive cover on avian vigilance. Anim. Behav., 43, 519-521.

Lima, S. L. & Dill, L.M.1990. Behavioral decisions made under the risk of predation: a review and prospectus. Can. J. Zool. 68, 619-640.

Loudon, A. S. I. 1985. Lactation and neonatal survival of animals. Symposia of the Zoological Society f London 54:183-207.

Machlis, L., P. Dodd, W. D., &. Fentress, J. C.. 1985. The pooling fallacy: problems arising when individuals contribute more than one observation to the data set. Zeit. Tierpsych. 68, 201-214.

Martin, P. & Bateson, P. 1993. Measuring Behaviour. Cambridge: Cambridge University Press.

MathSoft Inc. 1994. Mathcad Plus 5.0 for Windows: User's Guide. MathSoft Inc., Cambridge, MA.

McCullagh, P. & Nelder, J. A. 1983. Generalized Linear Models. London: Chapman and Hall.

McNamara, J. M., & A. I. Houston. 1987. Starvation and predation as factors limiting population size. Ecology 68:1515-19.

Miller, M. W., N. Thompson Hobbs, & M. C. Sousa. 1991. Detecting stress responses in Rocky Mountain bighorn sheep (*Ovis canadensis canadensis*): reliability of cortisol concentrations in urine and feces. Can. J. Zool. 69:15-24.

Quenette, P. Y. & J. P. Desportes. 1992. Temporal and sequential structure of vigilance behavior of wild boars (*Sus scrofa*). J. Mamm. 73:535-540.

Risenhoover, K. L., & Bailey, J.A.. 1985. Foraging ecology of bighorn sheep: implications for habitat management. J. Wildl. Mgmt., 49, 797-804.

Roberts, G. 1994. When to scan: an analysis of predictability in vigilance sequences using autoregression models. Anim. Behav. 48:579-585.

Roberts, G. 1996a. Why individual vigilance declines as group size increases. Anim. Beh. 51:1077-1086.

Roberts, G. 1996b. Testing for patterns in sequences of vigilance behaviour. Anim. Beh. 51:1179-1182.

Shackleton, D. M. & J. Haywood. 1984. Early mother-young interactions in California bighorn sheep, *Ovis canadensis californiana*. Can J. Zool. 63:866-875.

Stockwell, C. A., G. C.Bateman, & J. Berger.1991. Conflicts in National Parks: a case study of helicopters and bighorn sheep time budgets at the Grand Canyon. Biol. Conserv. 56: 317-328.

Thomas, L., & F. Juanes. 1996. The importance of statistical power analysis: an example from *Animal Behaviour*. Anim. Beh. 52:856-859.

Wilkinson, L. 1990. SYSTAT: The System for Statistics. Evanston, U.S.A.: Systat, Inc.

Zar, J. H. 1984. Biostatistical Analysis. 2nd edn. New York: Prentice Hall.

APPENDIX A. EFFECT OF SAMPLE DURATION ON VARIABILITY EXPLAINED BY REDUCED REGRESSION MODELS

This example is based on data from Frid (1997). Table A1 shows the reduced regression model estimating vigilance responses (arcsin square-root of % time vigilant) of adult female Dall's sheep based on the original focal animals samples, which had durations of 13-15 min (as reported in Frid 1997). Table A2 shows results of the same analysis on the same data, except that samples were truncated to 6 min. Notice that the reduced regression model is essentially the same, but truncating sample durations caused the R^2 to drop from 0.24 to 0.18, and the standard error of estimate (SEE) to increase from 0.11 to 0.16. Prior to being reduced, independent variables in both models were group size, distance to cliffs, distance to obstructive cover, and the second and third order interactions of these variables.

Table A1: Reduced model based on original samples from Frid (1997) with durations of 13-15 min.

Variable (log-transformed)	Regression coefficient	Standardized regression coefficient	Tolerance	P
Constant	0.12			0.008
Distance to cliffs	0.12	0.54	0.59	< 0.001
(Group size) x (Distance to cliffs)	-0.056	-0.61	0.59	< 0.001

ANOVA summary: $R^2 = 0.24$, F = 16.21, DF = 2, 102, P < 0.001, SEE = 0.11

Table A2: Reduced model based on samples from Frid (1997) that were truncated down to 6 min durations.

Variable (log-transformed)	Regression coefficient	Standardized regression coefficient	Tolerance	P
Constant	0.073			0.22
Distance to cliffs	0.14	0.46	0.58	< 0.001
(Group size) x (Distance to cliffs)	-0.067	-0.53	0.58	< 0.001

ANOVA summary: ($R^2 = 0.18$, F = 10.98, DF = 2, 102, P < 0.001, SEE = 0.16.)

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