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Fractal analysis of animal behaviour as an indicator of animal welfare

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Abstract

Animal welfare assessment commonly involves behavioural and physiological measurements. Physiological measures have become increasingly sophisticated over the years, while behavioural measurements, for example duration or frequency, have changed little. Although these measures can undoubtedly contribute to our assessment of an animal's welfare status, a more complex analysis of behavioural sequences could potentially reveal additional and valuable information. One emerging methodology that could provide such information is fractal analysis,, which calculates measures of complexity in continuous time series. Its previous application in medical physiology suggested that it could reveal 'hidden' information in a dataset beyond that identified by traditional analyses. Consequently, we asked if fractal analysis of behaviour might be a useful non-invasive measure of acute and chronic stress in laying hens and in pigs. Herein, we outline our work and briefly review some previous applications of fractal analysis to animal behaviour patterns. We successfully measured novel aspects of complexity in the behavioural organisation of hens and pigs and found that these were stress-sensitive in some circumstances. Although data collection is time consuming, the benefit of fractal analysis is that it can be applied to simple behavioural transitions, thereby reducing subjective interpretation to a minimum. Collectively, the work to date suggests that fractal analysis — by providing a novel measure of behavioural organisation — could have a role in animal welfare assessment. As a method for extracting extra information from behavioural data, fractal analysis should be more widely examined in animal welfare science.

Keywords: *animal welfare, behaviour, chicken, fractal analysis, pig, stress*

Introduction

The analysis of animal behaviour patterns has a central role in the assessment of animal welfare (Mench & Mason 1997). However, with a few notable exceptions (eg consumer demand [Mason *et al* 2001] and qualitative behavioural assessment [Wemelsfelder *et al* 2001]), the methods and analyses used in behavioural research have changed little over the years. The majority of behavioural studies still rely on statistical comparisons of mean duration or frequency. Although such measures undoubtedly contribute to welfare assessment, it may be possible to develop new analyses that allow the extraction of additional information from the behavioural data at our disposal. Fractal analysis is one methodology that might prove useful in this respect.

In this paper we briefly review some studies where fractal analyses have been applied to behaviour patterns. We also include examples of our own research, where we examined the value of fractal analysis as an indicator of stress in laying hens and in pigs. Our primary aim is to illustrate the sort of information that fractal analysis methods can reveal.

What is fractal analysis?

Many structures and processes in nature exhibit an irregularity and complexity that can be hard to measure. One mathematical technique used to describe complexity in nature is called fractal analysis (Mandelbrot 1977). Since its development, fractal analysis has been used to measure the degree of irregularity in a wide range of complex phenomena. The classic example is Mandelbrot's (1967) analysis of the irregularity of the British coastline. Although the original fractal methods were developed for such geometric analysis, fractal analysis can be used to measure complexity either temporally or spatially.

Mathematically, fractal analysis measures the degree of irregularity using the concept of scaling. The degree of scaling depends on how the measured size of an object changes as the scale of measurement changes. For example, the measured length of a highly irregular coastline will be larger if the measurement scale is changed from kilometres to metres. However, in very regular coastlines there is little change in the measured value as the measurement scale becomes more precise. In fractal structures the relationship between a measured property (P) (eg length) and measurement resolution (r) is defined as a power law: $P = kr^d$, where k is a constant (Avnir *et al* 1998). The exponent of the power law, d, relates to the degree of scaling. Different exponents indicate alterations in the scaling relationship and these exponent values are therefore used as a measure of irregularity in the analysed structure or process.

The ability of fractal mathematics to describe complex structure or dynamics means that changes in the organisation of the phenomenon in question can be identified. One of the most common applications of fractal analysis in biology has been in human medical physiology (Bassingthwaighte *et al* 1994), where the potential diagnostic value of fractal analysis is being increasingly recognised. A common finding is that temporal patterns (eg heart rate fluctuations) become more regular and predictable with age or disease (Goldberger *et al* 2002). In medicine and in many other fields it is therefore becoming recognised that fractal analysis can reveal useful information about temporal or spatial complexity. Although summary measures remain important, analysing temporal structure in a data set can provide valuable insights into the underlying process, revealing information that is both distinct and informative (Paulus & Braff 2003).

Although the original fractal methods were developed for geometric analysis, fractal analysis can be used to measure complexity either temporally or spatially. The ability of fractal mathematics to describe complex structure or dynamics means that changes in the organisation of the phenomenon in question can be identified. Fractal analysis embraces a variety of techniques, all of which centre around the presence of power-law scaling either in sequential or statistical properties of data.

Previous fractal analyses of animal behaviour

Fractal analysis has been applied to animal behaviour data in many ways and in numerous different species. Here, we briefly review some particularly pertinent studies.

Fractal analysis has been applied to the movement patterns of many species. In these analyses a fractal exponent is calculated on the basis of the complexity of movement: ranging from animals that move in a straight line to animals that have such a complex movement pattern that they cover all of the possible ground in any given area. The studies most relevant to animal welfare research are those where Paulus and Geyer (1991, 1993) assessed rodent movement patterns in a small arena by calculating a scaling exponent relating total distance travelled to the scale of the measurement. This measure of pattern complexity is independent of total locomotor activity (Paulus *et al* 1999a), and when the method was applied to the behaviour patterns of different rat strains, Paulus and co-workers (1998b) found that different strains had significantly different fractal movement patterns but the same total level of activity. Thus, the pattern and amount of locomotor activity provide different dimensions for describing behaviour. The complexity of movement patterns is affected by rearing conditions (Paulus *et al* 1998a) and various drugs (Paulus & Geyer 1991, 1993). Mice rendered hyperdopaminergic by gene knockout show less complex 'perseverative' movement patterns (Ralph-Williams *et al* 2003).

Fractal analysis has also been applied to assess the complexity of behaviour patterns over time. Motohashi and colleagues (1993) measured the fractal structure of rat locomotor activity over time using a power spectral analysis method. They found that the fractal organisation of the rats' locomotor behaviour was significantly altered by intra-peritoneal injection of the toxic solvent, tetrachloroethylene. Alados and various co-workers have investigated the potential use of fractal analysis as an indicator of animal well-being. For example, Alados and co-workers (1996) found a reduction in the complexity of the pattern of vigilance and feeding behaviours in Spanish ibex (*Capra pyrenaica*) as a result of pregnancy or parasitism. These authors also used a spectral analysis method, as well as other fractal methods. What is interesting about this study is that standard behavioural measures did not differ between pregnant or parasitised animals and controls. For example, pregnant animals spent just as long feeding and performed the same number of head-lifts as non-pregnant animals, yet the temporal pattern of these behaviours was significantly altered.

Other studies have shown lowered complexity in the reproductive behaviour of fathead minnows (*Pimephales promelas*) exposed to lead (Alados & Weber 1999), and in the social behaviour of diseased chimpanzees (*Pan troglodytes schweinfurthii*) (Alados & Huffman 2000). Specifically, Alados and Weber (1999) found that the pattern of certain reproductive behaviours became more structured (less complex) in male minnows exposed to lead before sexual maturity, but not in those exposed after, and Alados and Huffman (2000) found that behavioural complexity was reduced during disease in female chimpanzees, whereas it was unaffected by health status in males.

Fractal analysis in animal welfare assessment

The notion that fractal analysis could potentially provide information about behavioural organisation beyond that revealed by standard analyses of frequency and duration prompted us to investigate its value in welfare assessment. We chose the fractal technique of Detrended Fluctuation Analysis (DFA), which has previously been applied to behaviour patterns (Alados & Weber 1999; Alados & Huffman 2000). DFA was originally developed to assess complexity in DNA sequences (Peng *et al* 1995) and has since been applied in a range of contexts. When applied to behavioural data, DFA measures the randomness of the pattern of fluctuation back and forth between two behavioural states in terms of long-range autocorrelation. The presence of long-range autocorrelation indicates that the value of a particular variable depends not only on the immediately preceding values, but also on values much earlier in the sequence (ie the sequence has 'memory'). As the long-range autocorrelation of the data increases, the patterns become more structured and less complex. Conversely, fluctuations of higher complexity are those in which the switching is closer to a random pattern, with little or no relationship between current and previous states. Our first experiment aimed to determine whether the fractal organisation of vigilance behaviour in juvenile laying hens was altered by exposure to mild acute stressors (Rutherford *et al* 2003). Each bird's behaviour was recorded for just over 51 min

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using focal observations from video recordings. We compared the bird's behaviour when they were: 1) undisturbed in their home pen (n = 24); 2) placed in a novel arena (n = 23); 3) returned to their home pen following blood withdrawal (n = 16); and 4) returned to their home pen following a 5 min period of mechanical restraint and subsequent blood withdrawal (n = 17).

DFA revealed that the pattern of fluctuation between vigilant and non-vigilant behaviours showed long-range autocorrelation. Thus, although unpredictable from moment to moment, vigilance had an underlying structure, in that the current vigilance behaviour of an animal was influenced by its past sequence of vigilance. The pattern of vigilance was unchanged following blood sampling alone, but became more random during exposure to a novel arena or after blood sampling plus restraint, whereas the total time spent vigilant was elevated only in the novel arena. Hence, vigilance behaviour was qualitatively but not quantitatively altered after blood sampling plus restraint. Additionally, the fractal measure of vigilance organisation did not correlate with the total amount of vigilance shown over the observation period. These results confirm that fractal analysis can provide a measure of the 'temporal architecture' (Paulus & Braff 2003) of behaviour that is independent of the total quantity of that behaviour and is altered following exposure to an acute stressor.

Previous applications of fractal analysis to behavioural patterns (Motohashi *et al* 1993; Alados *et al* 1996; Alados & Weber 1999; Alados & Huffman 2000) have found reduced behavioural complexity under conditions that chronically challenge the animal. In contrast, we found an increased behavioural complexity under short-term fear-inducing conditions, and an associated mild distress in the animals. This difference matches intuitive expectations as to the effects of chronic and acute challenges on behaviour (eg acute challenges cause a more active response while chronic challenges inhibit behaviour). The key point is that behavioural complexity can potentially increase or decrease in response to different types of challenge.

We have also applied DFA to behaviour in pigs (Rutherford *et al* unpublished). Here, ten growing pigs were individually exposed to a chronic stress treatment involving repeated aggressive interactions with an unfamiliar larger pig and additional environmental stressors such as wetting or removal of the substrate (straw), or unavoidable airflow. Video recordings were made of the behaviour of test pigs and littermate controls over two 24 h periods (before and after the stressor treatment). We also measured salivary cortisol over 24 hours, before and after the stressor treatment.

We applied DFA to various different behavioural categories, such as: postural activity, behavioural activity (all periods when the animals were neither asleep nor idling/dozing), feeding, interacting with the environment and social behaviour. Preliminary analysis showed that following the stressor period the fluctuating pattern of postural activity (ie the changes back and forth between standing/walking and sitting/lying) was significantly more structured (less random) in test pigs compared to controls. No detectable differences were found in the total amount of activity or in its circadian pattern. Circulating cortisol levels were also higher in test pigs, compared to controls, over the 24 h period following stress exposure. For all pigs (ie test and control pigs), cortisol concentration was correlated with the pattern of postural activity, such that pigs with higher cortisol values had a more structured behavioural pattern.

Discussion

Fractal analysis is used to describe and measure complexity in a variety of different fields and is thought to reveal otherwise hidden information about organisational properties of complex systems (Peng *et al* 2000). These features are rarely considered in behavioural research and it is this fact that makes fractal analysis a potentially useful methodology. In many cases, fractal analysis enables assessment of when complex organisational properties change or how they compare in different situations.

We successfully measured a novel aspect of behavioural complexity both in laying hens and in pigs. We also showed that fractal analysis provides a measure of behavioural organisation that is independent of the total amount of behaviour and that these measures can alter under stress when standard measures do not. Collectively, our findings tentatively suggest that fractal analysis could play a useful role in welfare assessment. However, if fractal analyses, such as the DFA method used in our work or any of the other fractal methods, are to prove useful, more detailed examination of them will be necessary. Fractal measures need to be stringently validated against currently available indices of poor welfare, such as physiological markers of stress, and metabolic and immunologic parameters. The degree to which fractal analysis reveals hidden information must also be established: where standard measures of behaviour are sensitive to stress it is questionable whether fractal analysis would provide any extra useful information.

Furthermore, there will probably be factors that cause fractal behavioural complexity to alter, yet have no impact on an animal's state of well-being. It is also possible, indeed likely, that fractal complexity could alter in situations that are neutral or positive from a welfare perspective, just as some physiological indicators, such as glucocorticoids, do (Dawkins 1999). Rushen (2003) recently noted that many parameters (behavioural and other parameters) have been used as welfare indicators despite a poor understanding of their biological basis. It is important that we do not fall into this trap with fractal analysis. It could be said that fractal analysis provides a fine-scale tool with which to describe very subtle alterations in behaviour. Alternatively, a critic could argue that the alterations in behaviour are so small as to be meaningless. It is this point (which may well have some basis) that calls for the careful validation of fractal analysis. Hopefully, this will come from its wider use in the applied ethology community.

How then might fractal analysis be used in welfare research? In acute stress situations it could provide more precise quantification of the lasting impact of an acute challenge. The ability of animals to adapt to acute challenge is an important determinant of welfare, and one that may be altered when environmental conditions and/or the background genome lead to exaggerated fearfulness, anxiety or hyper-excitability (Jones 1996; Rosen & Schulkin 1998; Boissy *et al* 2001). In this sense, altered fractal complexity could be used in welfare research as an indicator variable (Fayers & Hand 2002).

Alternatively, if poor welfare results from lowered behavioural complexity, reduced behavioural complexity during chronic stress could actually be viewed as a causal variable (Fayers & Hand 2002). Physiological studies suggest that complexity is healthy as it allows body systems to readily adapt to challenge, and that a loss of complexity in biological functioning, with age or disease, results in a decreased ability to cope (Goldberger *et al* 2002). Alados and Weber (1999) also suggest that a degree of complexity in behaviour patterns is beneficial in dealing with events in the environment, and that decreased behavioural complexity could leave animals vulnerable to challenge. It remains to be established whether this is the case. In this context, the influence of environmental rearing conditions on the fractal complexity of behaviour patterns needs to be established. Inglis (2000) suggests that, in a heterogeneous changing environment, behavioural variability will be favoured over regularity because animals can never be absolutely certain that the cognitive expectations they have formed about the environment will be entirely correct. A corollary of this is that behavioural variability may decrease when animals are kept in barren environments.

Previous work (Alados *et al* 1996; Alados & Weber 1999; Alados & Huffman 2000) has suggested that reduced behavioural complexity might correlate with states of increased allostatic load (described as "a measure of the cumulative physiological burden exacted on the body through attempts to adapt to life's demands" [Seeman *et al* 2001]; see also McEwen & Wingfield 2003), or with pre-pathological/sub-clinical stress states (Moberg 2000), where an animal continues to function ostensibly as normal, yet is impaired and vulnerable to further challenge. Further work is needed to investigate this possibility, but fractal analysis potentially provides a tool for identifying subtle alterations that may well occur during states of sub-clinical stress or in the early stages of disease. Behavioural measures are under-utilised in studies of disease processes and their treatment or prevention. Many studies would benefit from a sensitive and non-invasive measure of effect onset or severity. Similarly, studies of conditions that may be chronically painful or uncomfortable, such as mastitis in cattle, or sheep scab, could benefit from the use of a sensitive behavioural assay.

Spatial fractal analysis (eg Paulus & Geyer 1991, 1993) might provide a useful additional measure of open-field behaviour, beyond those currently used. Fractal analysis could also play a role in the investigation of abnormal behaviours. For example, might a decrease in behavioural complexity be a precursor to stereotypic behaviour, or, more generally, might fractal analysis provide novel measurements of stereotypy? For example, using a fractal measure, Paulus and co-workers (1999b) found that, compared to controls, schizophrenic humans showed a larger and longer-lasting dependence of current behaviour on previous behaviour. Behavioural tests relating to such perseverance in behaviour are being investigated in animal behaviour studies relating to stereotypic behaviour (eg Garner & Mason 2002).

Animal welfare implications

We highlighted, in the introduction, the importance of behavioural assessment in animal welfare research. Fractal analysis can add to current analyses by revealing previously 'hidden' information about behavioural organisation. Calculation of a fractal exponent as a description of behavioural organisation provides additional information above and beyond that provided by simpler analyses of mean frequencies and durations of behaviour.

A small number of studies, including our own, suggest that fractal analysis can reveal alterations in behaviour relating to well-being when standard measures do not. Although the significance of this for welfare assessment remains to be fully understood, the ability to extract more information from our data can never be detrimental. Indeed, it is more likely that by providing the opportunity to assess stress in a non-invasive way, and using data that are already routinely collected, fractal analysis could add to the range of tools used within welfare assessment. We have speculated upon areas in which fractal analysis could prove useful and would hope that fractal measures will be more widely investigated as a welfare assessment tool.

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