

Morphometric Grayscale Texture Analysis using Foot Patterns

Dan Ashlock

Iowa State University
Department of Mathematics
Bioinformatics and Computational
Biology
danwell@iastate.edu

Dean C. Adams

Iowa State University
Department of Ecology, Evolution,
and Organismal Biology
Department of Statistics
Bioinformatics and Computational
Biology
dcadams@iastate.edu

David Doty

Iowa State University
Department of Mathematics
Bioinformatics and Computational
Biology
ddoty@iastate.edu

Abstract- The field of quantitative morphology has long been important in biological investigations. Various elements of an organism's morphology, such as size and shape, are easily quantified, and standard methods for the analysis of these components exist (i.e., *geometric morphometrics*). However, methods for reliably quantifying textures and patterns are currently lacking. In this paper we propose a technique for quantifying grayscale images of biological textures and patterns. With our method, the textural properties of an image are represented as a *foot pattern* of 2-dimensional Cartesian coordinates, obtained via an evolutionary algorithm that minimizes pattern entropy. The pixels of the foot pattern are then assigned labels using one of two techniques: complete enumeration, or by minimizing the differences between sets of landmarks (using a heuristic search for the optimal assignment). The labeled landmark coordinates are then treated as input data for standard quantitative morphometric analysis. With this approach we were able to statistically distinguish between foot patterns generated from two different textural images drawn from the backs of salamanders. Thus, morphological textures and patterns may be quantified, and sets of textures statistically compared.

1 Introduction

For centuries biologists have compared organisms based on their anatomical features. These studies were historically descriptive, where differences of particular morphological components were described qualitatively (e.g., species A has a longer neck and is more slender than species B). As biology became increasingly quantitative however, so too did morphological analysis. Morphological studies soon included quantitative data for one or more measurable traits, which were

statistically summarized and compared among species. Thus, by the mid-twentieth century, quantitative description of morphological shape was combined with statistical analysis describing patterns of shape variation within and among groups, and the modern field of morphometrics began [1].

The morphology of a biological structure can be thought of as a combination of factors, including: size, shape, color, color patterns, and texture [2]. Size and shape describe the geometric properties of an object, while color, color patterns, and texture describe other structural properties. Using the tools of *geometric morphometrics*, size and shape can be reliably quantified, and patterns of shape variation and its covariation with other variables documented. In stark contrast, the rich information contained in biological textures and patterns remains largely unexploited.

Textures and color patterns are extremely important components of an organism's morphology. External surface texture is used by some organisms to obtain moisture from early morning fog, while other organisms use color patterns to enhance their survival by blending in with their environment (crypsis), or by mimicking other prey items that their predators avoid. Several attempts have been made to compare biological textures and patterns [3, 4], but quantifying them in a consistent and repeatable fashion remains difficult. This has resulted in the use of ad hoc approaches to quantify selected components of an organism's texture or color pattern. Clearly, given the importance of morphological textures and patterns on an organism's survival, developing a repeatable means of quantifying these aspects of morphology is desirable.

In this paper we propose a new approach to extracting texture and pattern information from digital images, so these features may be quantified more accurately. The well-developed theory of shape analysis and its quantification procedure serve as the template on which we base the analysis of biological textures. Our approach extends existing texture classifi-

cation techniques using evolutionary algorithms, and combines them with geometric morphometric methodology. We describe our method below, and illustrate its use on a simple data set by quantitatively comparing the textural images from *Gyrinophilus porphyriticus* salamanders from two different populations.

2 Methods

2.1 Texture Quantification

For our approach, we quantified textures by extending previous methods of texture classification that use *foot patterns* optimized by evolutionary algorithms [5], and combined this with methods for the analysis of biological shape (see below). In this method, a foot pattern is defined as a $n \times n$ window containing a set of k pixels (feet). Each $n-k$ foot pattern may be represented as a list of pixel positions within the window of the foot pattern. How well a foot pattern matches an image is defined in terms of entropy [6]. A foot pattern with low entropy has a better ‘fit’ to the image than a foot pattern with high entropy. To calculate a foot pattern’s entropy, the $n \times n$ window is scanned along the image, and at each position, the pattern of black and white pixels under the k pixels of the foot pattern are observed (this pattern is called a *footprint*). For a given foot pattern, there are g^k possible footprints, where g is the number of gray values in the image. The number of times each footprint, ϕ , occurs is counted, generating an event histogram of footprints and their frequency. This is normalized by the total number of possible ϕ , providing an empirical probability distribution p_ϕ [5]. The entropy (in bits) of that foot pattern X relative to image Y is then calculated as the entropy $E(X, Y)$ of the empirical probability distribution:

$$E(X, Y) = - \sum p_\phi * \log_2(p_\phi)$$

The goal is to find the foot pattern with the lowest entropy (and hence best fit) relative to a particular image. Unfortunately, finding the optimal foot pattern is difficult, as an exhaustive search is computationally unfeasible. For $k = 7$ pixels in a 10×10 window, $\sim 6.40 * 10^{14}$ calculations are required to find the optimal foot pattern for a single black-and-white image. Therefore, a heuristic search procedure using an evolutionary algorithm is implemented [7]. For our procedure (see figure 1), we used a generalization of the discrete $n-k$ foot patterns that allowed the x, y coordinates of each foot to be coded as continuous, real numbers. To generate continuous (k, r, s) foot patterns, k real-valued pixels (x, y) were chosen inside a circle of diameter r , with a minimal pairwise distance (s) between points,

where $s \geq 1.5$, to prevent two pixels from occupying the same location.

In implementing the algorithm an initial population of 400 continuous (k, r, s) foot patterns was created, each consisting of k pairs of real numbers representing the x, y coordinates of each foot. The foot patterns were then improved by running a steady state evolutionary algorithm using single tournament selection of size four. For any foot pattern the entropy relative to the image for that foot pattern was computed. Four foot patterns were randomly selected to form a tournament. The two foot patterns with the lowest entropy were blended with uniform crossover to produce new foot patterns, which replaced the worst two. If a crossover produced pairs of points closer than 1.5 pixels the results were deleted and a new crossover was performed until all pairs of points were at distance at least 1.5. Random mutations were performed in the following fashion. Either one of the points in the foot patterns was moved or the entire foot pattern was rotated. If a point (foot) was moved its proximity to other feet was checked for a minimum distance 1.5. Movement was repeated until the minimum distance requirement was satisfied. Rotation was by a random angle in the range $[-0.1, 0.1]$ radians, selected uniformly at random. This procedure was repeated for 40,000 mating events. With this approach, entropy minimization proceeded over a sampling of all possible foot patterns, and all possible foot pattern rotations with respect to the image. As a final step, the optimal foot pattern was rotated in small increments about its axis, to determine if any fine-scale rotations improved its fit to the image.

2.2 Grayscale Textures

Texture analysis methods using foot patterns were previously only implemented for black and white images. When used on gray-scale images, the method suffers from a combinatorial explosion of possible footprints ϕ used to generate the histogram, causing an entropy surface that is effectively flat. Several possible generalizations of the approach for gray-scale images are possible (see e.g., [8]). One solution is to convert the grayscale image to black-and-white, by binning the shades of gray into two bins: high and low. Alternatively, one could decompose the gray-scale image into multiple black and white images, which are then treated in parallel, using a single foot pattern as before. Unfortunately, the most logical procedure (pulling bits off the gray-scale values sequentially) is ineffective, because these bits, in the order they appear in a 256-shade (8 bit) gray picture, yield sparse, large-scale, and random images in at least some of the bit-layers. The reason is that the local ran-

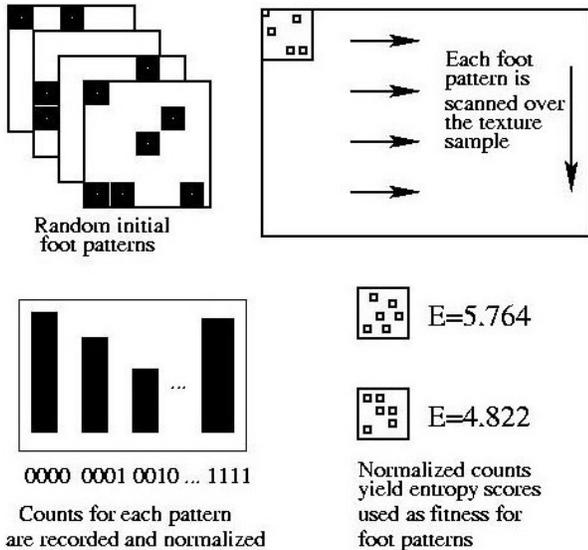


Figure 1: Graphical illustration of the protocol for identifying the optimal foot pattern for a given texture using a genetic algorithm. Each foot pattern is scanned over the image, generating a histogram of footprints. The foot pattern yielding the lowest-entropy histogram is considered the best quantitative description of the image.

domness characterizing a texture is not apparent in the individual bits of the gray-scale representation.

Another solution is to use a clustering algorithm, such as k-means, on the footprints, and treat any footprint in the same cluster as the same. With k-means clustering, we may set the number of clusters, and so we could guarantee that the number of types of footprints ϕ counted in the event histogram is less than the total number of footprints counted when scanning the image. However, this does discard some potentially useful information about the texture of the image, different sets of pixel shades are treated as being the same. Furthermore, running this procedure after each foot pattern is scanned over the image would be extremely time-consuming.

Instead, the solution we propose is one that uses incremental entropy. The key observation is that if we use a small number of feet then the combinatorial explosion does not overwhelm the search technique. Using a small number of feet, say two, creates a different problem: the foot patterns are too simple to distinguish between different textures. For any given foot pattern we compute the incremental entropy as follows. A *prefix* of a foot pattern is an initial set of feet. The feet are ordered within the data structure and this is the ordering used to compute the prefixes. For all the prefixes with

2 or more feet, and for the full foot pattern, the entropy is computed in the same manner as for foot patterns in general. These entropies are summed to obtain the incremental entropy. The incremental entropy is used as the fitness function in evolutionary algorithms searching for foot patterns on gray scale textures.

It is worth examining the incremental entropy from the point of view of fitness landscapes. The entropy for a full foot pattern with enough feet for later use in classification (and possibly synthesis) of textures forms a fitness landscape that is a vast plane covered with small-scale noise and possessing a hidden valley here and there. The prefixes of a foot pattern have a much simpler landscape in which the valleys are not so hidden - but they are also much larger and contain less information about the texture. Summing entropy across all prefixes superimposes these landscapes. Optimization takes place preferentially in the prefixes permitting the algorithm to successively place the feet of the foot pattern one after the other during the course of evolution. This technique does not appear to have immediate generalizations to other optimization problems but it does permit the location of low entropy foot patterns in spite of the combinatorial explosion of footprints caused by attempting to work in grayscale.

2.3 Landmark Assignment

Using the above procedure a low-entropy foot pattern is identified for each image, consisting of the x, y coordinates of k unlabeled feet. To use these as a set of 'texture variables' for morphometric analysis however, we must first identify correspondence between pixels in different foot patterns. Because the only information available is the set of x, y coordinates of the feet (hereafter called *landmarks*), we use the relative positions of the landmarks themselves as a means of assigning their correspondence to one another. We implemented two methods for comparison.

The first method (*Complete Enumeration*) is based on the method described by Dryden and Mardia [9], which minimizes the differences between objects under rigid transformations to identify the optimal labeling of their landmarks. First, positional differences were eliminated by translating each object so that its centroid was located at the origin (i.e., mean centering). Next, each object was scaled to unit size, where size was defined as *centroid size*: the square root of the sum of squared distances from each landmark to the centroid [10]. The correspondence of landmarks between two objects F and F' was then found by first assigning labels arbitrarily, and then optimally rotating one configuration on the second using a least squares superim-

position [11]. Fitness was defined as the square root of the sum of squared differences between corresponding landmarks, or Euclidean distance:

$$d(F, F') = \sqrt{\sum (x - x')^2 + \sum (y - y')^2}$$

This process was repeated for all possible landmark labellings and each time the distance was calculated. The landmark assignment that yielded the minimal distance:

$$\min [d(F, F')]$$

was considered to be the correct landmark assignment. For more than two objects, all objects were superimposed relative to one object in the data set, and its landmark assignments were used as the global assignment for all specimens.

A critical shortcoming with the *Complete Enumeration* method is that as the number of landmarks increases, assessing all possible $k!$ landmark assignments quickly becomes computationally impractical. The second approach (*Heuristic Assignment*) uses heuristic search to provide a solution to the landmark labeling problem that can be found in polynomial time. This method uses a variant of the Hungarian algorithm [12]. Typically, this linear programming method is used to solve the ‘‘assignment’’ problem, where a set of n jobs are assigned to n workers in a 1-to-1 mapping, so as to minimize the total cost of the assignment (as specified by the n by n worker-job matrix). In our implementation, we assigned landmarks in one foot pattern to landmarks in a second pattern so as to minimize the total squared Euclidean distance between them. First, we mean-centered each foot pattern and scaled them to unit centroid-size. For two foot patterns F and F' we then calculated the k by k matrix of squared Euclidean distances between landmarks. This matrix was subjected to the Hungarian algorithm, and the matching which minimized the sum of squared Euclidean distances between landmarks was treated as the preliminary labeling of the landmarks:

$$\min [d^2(F, F')]$$

Finally, to eliminate differences in rigid rotation between F and F' , we rotated F in 30 equal degree increments, each time finding the optimal landmark assignment using the above procedure. The optimal labeling of landmarks in F and F' was then defined as the labeling from the rotation with the minimal sum of squared Euclidean distances between landmarks:

$$\arg \min [d^2(F, F')]$$

One complication with the procedure described above is that it only works for two objects. For more

than two foot patterns, a generalization of this approach must be used that identifies the *optimal multi-dimensional assignment* of landmarks. The problem is to find, for each foot pattern $F \in T$, a labeling to every other foot pattern $F' \in T$, such that all the labellings are *consistent* with each other. Because the multi-dimensional assignment problem is NP-complete [13], we used a polynomial time algorithm that approximates the optimal multi-dimensional assignment within a factor of 2 [14]. First, we calculated the optimal matching and minimal squared Euclidean distance between F and all other $F_i \in T$:

$$\min [d^2(F, F_i)]$$

Next, the sum of these minimal squared Euclidean distances between F and all other $F_i \in T$ was calculated for all F . The F with the minimal sum of squared minimal distances was treated as the best approximation of the *center star*, F^c , which is a measure of central tendency for the set T (analogous to its mean). To find the multi-dimensional landmark assignment, each foot pattern in T was then matched to F^c using the Hungarian algorithm, and the labeling imposed by F^c was treated as the optimal labeling of landmarks for each foot pattern. Because all foot patterns in T were matched to F^c , upon completion of this procedure they were also optimally labeled with respect to each other. To compare foot patterns from two different textures, the center stars F^{c1} and F^{c2} were found for the two sets of foot patterns, and the optimal matching between them was determined. Landmark assignments for all specimens were then determined by matching each foot pattern to its respective center star. This procedure can be extended to more than two groups in a similar fashion.

2.4 Morphometric Analysis

The minimum entropy foot pattern is the best available quantitative representation of the textural information of an image. To compare these quantitatively, the k labeled pixels for each foot pattern were treated as primary data for a geometric morphometric analysis. In geometric morphometrics, a set of two- or three-dimensional coordinates of corresponding points are used to represent the shape of a morphological structure. Variation in digitizing location, orientation, and scale is removed from these values by translating each object to a common location, scaling to unit size, and optimally rotating using a least squares (Procrustes) superimposition [11]. The aligned objects are then projected to a Euclidean space, where similar objects are close together and dissimilar objects are far

apart. Scores on the axes of this space represent a set of shape variables, which may then be used in conventional multivariate statistical analysis (e.g., [15], [16]). For the quantitative analysis of texture, the foot patterns were aligned using Procrustes superimposition, were then projected to a Euclidean space, and scores on the axes of this space were treated as “texture variables.” These scores were then treated as multivariate data for statistical assessment of within and between group texture variation.

3 Example Application

3.1 Data Collection and Analysis

To assess the efficacy of our approach, we quantified the textural information present in images of the backs of two *Gyrinophilus porphyriticus* salamanders, one from each of two adjacent populations in the Cowee Mountains in North Carolina (figure 2). The images were converted to 8-level normalized grayscale to eliminate light level variation and noise-level variations in shade. To account for grayscale, the foot-pattern location genetic algorithm used the successive pixel adding technique described in section 2.2, where incoming vectors of grayscale data were placed in a tree with information for each pixel stored on one level of the tree (a TRIE data structure). The entropy for the first k pixels, $k = 2-7$ was computed and summed. This has the effect of conditioning evolution on the level of the tree where variation currently appears, with optimization of the first i pixels blending smoothly into optimization of the first $i+1$ pixels as evolution progresses, $i = 2-6$. For each image, the optimal foot pattern was obtained for 100 runs each, generating two populations of 100 foot patterns. Landmarks were then assigned labels using both of the methods described in section 2.3 (Complete Enumeration and Heuristic Assignment). Labeled landmark configurations (i.e. foot patterns) were then treated as data for a morphometric analysis, where they were superimposed, and a set of texture variables generated. These were then used in multivariate statistical analysis. To distinguish between the groups we performed multivariate analysis of variance (MANOVA) and discriminant function analysis (DFA). We also described patterns of variation in foot patterns using principal components analysis (PCA). Finally, to directly compare results between landmark assignment methods we calculated the matrix of Euclidean distances between foot patterns in each data set for each method, and compared these using a Mantel test of matrix correlation.

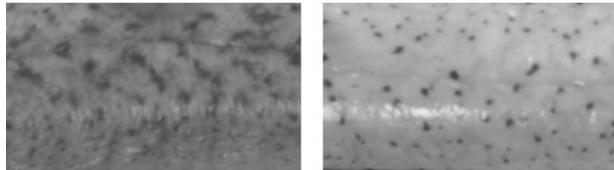


Figure 2: Images from a portion of the backs of two *Gyrinophilus porphyriticus* salamanders. The left image is from a salamander from the Cowee North population, and the right image is from a salamander from the Cowee South population.

3.2 Results

For both methods of assigning labels to landmarks, foot patterns generated from the two images were distinguishable from one another. Using MANOVA, we found numerically identical results from the Complete Enumeration and the Heuristic Assignment methods: (Wilks’ $\Lambda = 0.706$, $F = 7.867$, $P \ll 0.0001$). When analyzed using DFA, the majority of foot patterns were correctly classified to their population: Complete Enumeration (72%) and Heuristic Assignment (72%). This showed that the textural information in the two images was distinct, and that the foot pattern approach was capable of quantifying this information.

Using PCA, 42.91% of the variation in texture variables was explained in the first two PC axes for both the Complete Enumeration and the Heuristic Assignment methods. When the texture data space was viewed along these PC axes, foot patterns from the two groups were ordered along PC1, with minimal entropy foot patterns from Cowee North located towards one end of PC1, and minimal entropy foot patterns from Cowee South located towards the other end of PC1. There was considerable overlap between the two populations in the center of PC1 however, which likely corresponds to foot patterns which were misclassified in the DFA. (figure 3).

Using a Mantel test, we found a perfect correlation between distances generated from the Complete Enumeration and the Heuristic Assignment methods ($r = 1.0$). This demonstrated that, for this data set, the textural information captured by each method was identical, and that the assignment of landmark labels was consistent between the two methods.

4 Conclusions and Future Work

4.1 Conclusions

In this paper we proposed a technique for quantifying grayscale images of biological textures and pat-

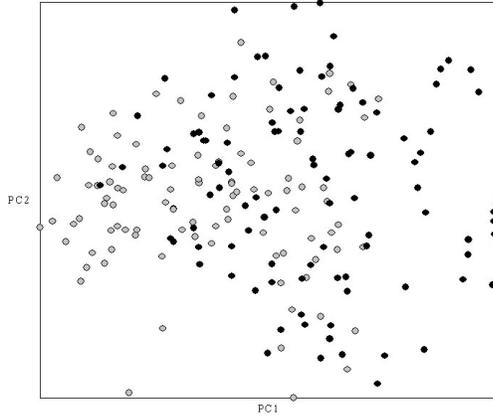


Figure 3: Results from principal components analysis of texture variables from 100 minimum entropy foot patterns from each of two images. Only data from the Heuristic Assignment method are displayed, as results from the Complete Enumeration method were numerically identical. Cowee North = gray, Cowee South = black.

terns using foot patterns optimized by evolutionary algorithms. The pixels of each foot pattern were assigned labels using one of two methods (Complete Enumeration and Heuristic Assignment), and sets of labeled foot patterns were then treated as input data for standard quantitative morphometric analysis. We demonstrated our approach by comparing 100 low entropy foot patterns generated from each of two morphological textures from the backs of *Gyrinophilus porphyriticus* salamanders.

Statistical results from MANOVA, PCA, and DFA were numerically identical for data obtained by the Complete Enumeration and Heuristic Assignment methods. Therefore, for this data example, the heuristic landmark assignment based on the Hungarian algorithm is able to identify the correct labeling of pixels in the foot pattern. This finding is significant, as the Heuristic Assignment method is considerably faster than Complete Enumeration (about 60 times faster for 7 feet!). As these algorithms are of factorial and polynomial time respectively, the difference in performance greatly increases as more images are examined, or as the number of pixels used the foot pattern is increased. Therefore, we anticipate that the Heuristic Assignment approach will be useful for most practical applications.

The evolutionary algorithm produced foot patterns that could be used to successfully statistically distinguished the two populations of salamanders. Examining figure 3 it is clear that the two groups of salamanders are distinguished in a manner involving more than

the first two principal components. The evolutionary algorithm is thus finding relatively subtle features that distinguish the salamander’s skin patterns.

We also found significant differences between sets of foot patterns generated from the two images. From this we conclude that our method of quantifying textural information using optimized foot patterns is capable of distinguishing between biological textures, and shows its great potential to extract biologically sensible characters from arbitrary biological texture data. This result is also biologically interesting, as previous work has shown that these two salamander populations are genetically isolated [17], and are morphologically distinct [18]. This differentiation occurs in spite of the fact that the two populations are adjacent (parapatric) and have a narrow contact zone in the Cowee mountains of North Carolina, and lends further support to the suggestion that they are no longer following the same evolutionary trajectory.

4.2 Future Work

Several aspects of our texture quantification protocol require additional investigation. First, the incremental entropy technique permits us to find foot patterns for 8 level grayscale, a substantial advance over the previous black-and-white work. However, the algorithm should be tested with larger numbers of gray values. The algorithm may already be practical at finer resolutions of gray, or it may require additional tinkering. Additional exploration of the characteristics of the fitness landscape for incremental entropy should also be made. Our nested valleys view is consistent with the behavior of the algorithm on the current test case, but is at best a hypothesis at present.

Finally, the original black-and-white foot pattern work permitted not only texture classification but also high fidelity synthesis of many textures. Such synthesis is quite valuable in biology, virtual reality, and other fields and is the focus of ongoing research. Synthesis techniques that permit morphing between textures would be especially valuable for exploring biological questions concerning natural history and evolution.

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