A MORPHOMETRIC APPROACH FOR THE ANALYSIS OF BODY SHAPE IN BLUEFIN TUNA: PRELIMINARY RESULTS

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SUMMARY

Geometric morphometrics was used to explore body shape morphology in 120 bluefin tuna specimens captured in a traditional trap in the western Mediterranean during the years 2008 and 2009. The shape of each individual was captured by recording the 2-D coordinates of 10 morphological pointing landmarks. We applied a general procustes analysis (GPA) in order to eliminate any morphological variations resulting from size, position or orientation of specimens. We then used the TPS (thin-plate spline) method to provide a graphical representation of shape and to compare the two sets of data. Preliminary results highlight some shape differences between the two groups and we discuss their possible sources.

RÉSUMÉ

La morphométrique géométrique a été employée pour explorer la morphologie de la forme corporelle de 120 spécimens de thon rouge capturés dans une madrague traditionnelle dans la Méditerranée occidentale en 2008 et 2009. La forme de chaque spécimen a été consignée en enregistrant les coordonnées bidimensionnelles de 10 points de repère morphologiques. Nous avons appliqué une analyse procustes générale (GPA) en vue d'éliminer toute variation morphologique résultant de la taille, de la position ou de l'orientation des spécimens. Nous avons ensuite utilisé la méthode TPS (plaque mince flexible) afin de fournir une représentation graphique de la forme et comparer les deux jeux de données. Les résultats préliminaires soulignent certaines différences de forme entre les deux groupes et le document examine leurs possibles origines.

RESUMEN

Se ha utilizado la morfometría geométrica para explorar la morfología de la forma del cuerpo de 120 atunes rojos capturados en una almadraba tradicional en el Mediterráneo occidental durante los años 2008 y 2009. La forma de cada individuo se obtuvo registrando las coordenadas bidimensionales de diez puntos morfométricos ("landmarks") prominentes desde el punto de vista morfológico. Se aplicó un Análisis Procrustes General (GPA) para eliminar cualquier variación morfológica derivada de la talla, posición u orientación de los ejemplares. A continuación se utilizó el método de TPS (delgada lámina deformada) para proporcionar una representación gráfica de la forma y comparar los dos conjuntos de datos. Los resultados preliminares muestran algunas diferencias en la forma entre los dos grupos, y en el documento se debaten las posibles fuentes que originan estas diferencias.

KEYWORDS

Bluefin tuna, body shape, geometric morphometrics, Mediterranean Sea, trap fishery

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1. Introduction

Phenotypic plasticity in morphometric traits may often be adaptive reflecting the effects of anthropogenic impacts or prey-predator processes in the ecological niches of a population (Robinson and Parsons, 2002). This phenomenon has been observed by applying geometric morphometrics to the Mediterranean horse mackerel (*Trachurus mediterraneus*) (Turan, 2004), and there are also well-studied examples among the species *Oncorhynchus*. Geometric morphometric analysis reveals differences in body shape between the Atlantic and Mediterranean lineage and hybrids of the brown trout (*Salmo trutta*)(Monet et al., 2006) and to allow to discriminate populations of Mugilidae by analyzing fish scales (Ibanez *et al.*, 2007).

Such techniques are frequently combined with genetic analysis in order to better understand phenotype-genotype relationships.

The Atlantic bluefin tuna, *Thunnus thynnus*, is known for its well-defined adaptability to specific environmental conditions. Study of physiological features such as size at reproductive maturity, the beginning and end of the spawning period, growth rate together with recent otolith chemistry studies (Rooker *et al.*, 2007, 2008) and retrospective analysis on population structure (Fromentin, 2009), have strengthened the hypothesis of ecotypes also referred to as "patchy or meta-population" types.

Trap fisherman throughout the Mediterranean Sea claim that they can determine the difference between a bluefin from the Atlantic Ocean and those that are resident in the Mediterranean Sea. In order to verify the hypothesis "oceans vs. resident" a comprehensive investigation, by means of a genetic and morphometric approach, started in 2008.

Here, we use a geometric morphometrics approach (Bookstein, 1991) to explore body shape variation among two data sets of specimens collected at a trap site in south western Sardinia (western Mediterranean).

2. Material and methods

The first sample (S1) consists of 92 bluefin chosen from the specimens caught during 2008 at the trap "Isola Piana" (south western Sardinia, Italy). The second sample (S2) consists of 67 bluefin caught during 2009 at the same location.

2.1 Coordinate acquisition

Each tuna was photographed on its left side, immediately after being taken out of the death chamber or "camera della morte". A Canon PowerShot G9 (12,1 megapixel, sensor 1/1,7", optical zoom 6x) was used and the same light exposure employed throughout. The digital photographs were then processed using tpsDig 2.10 software (Rohlf, 2006) for landmark acquisition. 10 Landmarks (**Figure 1**) were identified by conventional rules on biological homology (spatial congruence, ontogenetic and phylogenetic) (Bookstein, 1991). According to Bookstein (1991) landmarks are classifiable as first category points (suture points and tissue boundaries) and secondary points (drop points or highest warp points).

Their position was related to systems of coordinates (the x and y coordinates) which were useful for transformation.

2.2 Transformation and multivariate analyses

Each set of co-ordinates were submitted separately to a generalized procrustes analysis (GPA) available in the tpsRelw software (Rohlf, 2006). This procedure translated, rotated and scaled the original configurations in order to achieve the best superimposition of all shapes. The size of each specimen is represented by the "centroid size", a measure that is able to estimate the size in all directions in a body better than is possible by using univariate measures such as maximum length. After this superimposition, the software breaks down the morphological difference into a series of non-uniform components, described as partial warps. The scores of the specimens on the partial warp axes constituted the shape variables that were used in the subsequent statistical analyses (Rohlf 1999). The software was used to introduce shape variables into a Principal Component Analysis (PCA), and to visualize the warping associated with the various principal components (PCs). These components represent relative warps in the context of a TPS (thin-plate spline) approach (Bookstein 1991). PCAs can identify any regularity within the sample. In a morphometric analysis, these regularities correspond to simultaneous

displacements of anatomical points that are often observed in the specimens. A value is assigned to each relative warp and is expressed as a percentage, reflecting the proportion of the variation accounted for by this component. PCA automatically classifies the relative warps in decreasing order of their specific values. The greatest variations, generally attributable to biological factors, occur in the first few relative warps. The morphological warps associated with each component are visualized by observing the conformations corresponding to the points located at the ends of the axes. The changes in shape are illustrated by a potentially warpable grid, which represents the warps corresponding to a consensus (an average individual).

3. Results

Figure 1 shows the 10 landmarks chosen for the analysis. Row data of landmarks before and after GPA standardization are shown in **Figure 2**.

Variable shape output of the Tpsrelw application are shown in **Figure 3.** In the PCA the projections onto the first and second principal components are reported. The first component of the model shows the differences in body shape that are linked to the two data sets and it accounts for 40% of the body shape variation. The second component accounts for 21%. According to the amount of a unit of bending energy (in a positive and negative direction), the distinctive feature of body shape in the pooled specimens of 2008 are associated with a relative ventral expansion in the anterior-posterior shape. In contrast, the pooled specimens of 2009 are associated with a relative curvature of the dorsal-ventral shape. The body shape differences from the overall mean or consensus configuration are presented in **Figure 4**. The observation of the extreme warps associated with the first component are reported in **Figure 4AB**, and second component is shown in **Figure 4CD**. Analysis of overall data (2008-2009 pooled) separated by sex did not reveal any sexual dimorphism (**Figure 5**).

4. Conclusion

This preliminary contribution made by geometric morphometrics has shown small differences in the body shape of bluefin specimens collected over two different years. These differences should be further investigated in order to clarify their source. We hypothesis a) potential errors in the acquisition of landmarks from the images or b) genuine differences due to the diverse biological conditions in the two groups of bluefin sampled. According to the deformation grid obtained, the ventral expansion recorded in the shape of bluefin from the data set of 2008 could be related to the imminent reproductive phase of these groups of fish, dissimilar from the other group. Although these results are questionable, understanding body shape and quantitative deformation, could help to explain some of the mechanism during the different life phases of the bluefin. This exploratory study is still in progress because it forms part of a comprehensive approach to population discrimination by means of genetic analysis. Finally, major improvements needed include: increasing the samples of bluefin by varying locations and extending periods, increasing the number of landmarks and developing a protocol on 3-D landmark acquisition and processing.

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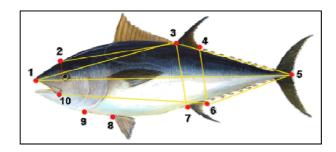


Figure 1. Truss network: landmarks and geometric distances used for the analysis.

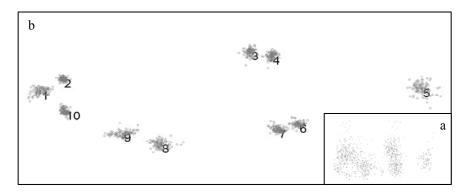


Figure 2. Landmark configuration of raw data before (a) and after standardization (b).

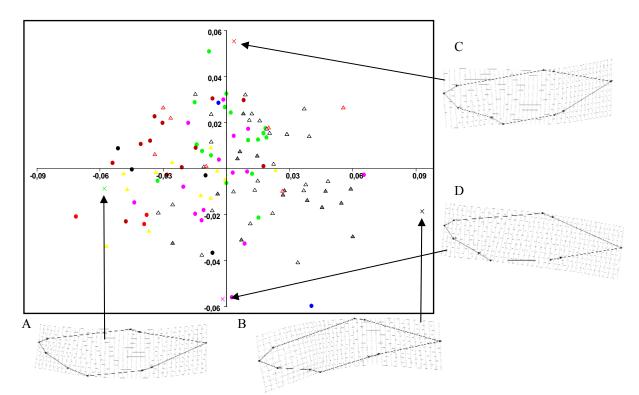
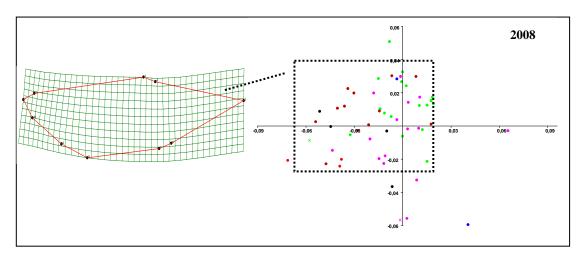


Figure 3. Projection onto the first (PC1) and second principal components (PC2) of the 120 bluefin tuna analysed. Symbols refer to individuals caught in 2008 (\bullet) and 2009 (\blacktriangle) and (AB) extreme warps associated with the first principal component (PC1); and (CD) extreme warps associated with the second principal component (PC2).



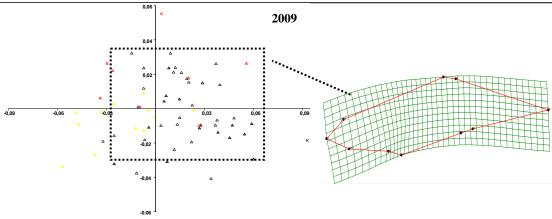


Figure 4. Extreme warps associated with PC1 for each data set (2008-2009).

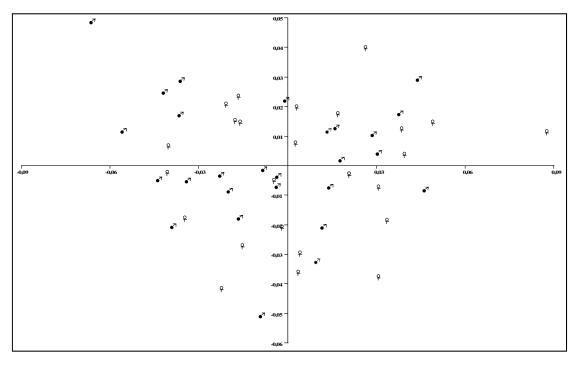


Figure 5. Projection onto the first (PC1) and second principal components (PC2) of the bluefin separated by sex (pooled data).