

Procrustes Analysis of Muscle Fascicle Shapes Based on DTI Fibre Tracking

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Abstract. Diffusion Tensor Imaging (DTI) is a technique developed from Magnetic Resonance Imaging (MRI), which uses a mathematical form diffusion tensor to measure the movement of water molecules in biological tissues in *vivo*. By performing fibre tracking using diffusion tensor data, we can study the micro-structure of biological tissues in a non-invasive way. Skeletal muscle plays a significant role in force and power generation that contribute to maintaining body postures and to controlling its movements. DTI fibre tracking may re-construct the skeletal muscle in a fascicle level. Procrustes analysis is a landmark-based method for studying the shapes of objects. In this paper, we explore using Generalised Procrustes Analysis to study the fascicle shapes that we have collected in medial gastrocnemius muscles from 6 healthy adults by using DTI technology. This is an innovated attempt of using Procrustes analysis to find the trend of the changes of fascicle shape when foot is in plantarflexion and dorsiflexion, by clustering method.

Keywords: Procrustes \cdot Shape \cdot Analysis \cdot DTI \cdot Skeletal muscle \cdot Fascicle

1 Introduction

Shape analysis has been introduced in scientific research for a long time and has been widely used. For example, animal skulls research in biology; postcode recognition in image analysis; steroid molecules in chemistry; MRI and cortical surface shape in medicine; DNA molecules in pharmacy; protein matching in bioinformatics; sand grains in particle science; human movement data in physiotherapy; electrophoretic gels in genetics; Central Place Theory in geography; alignments of standing stones in archaeology and so on [1-7].

As an innovative exploration of applying shape analysis on biological tissues, based on our previous research [8], which uses DTI technology to reconstruct the skeletal muscle of human beings, we try to find the hidden information between foot positions and the muscle fascicle shapes.

1.1 DTI and Fibre Tracking

Diffusion Tensor Imaging (DTI) is a technology which uses Diffusion-Weighted Magnetic Resonance Imaging (DWI or DW-MRI) data to describe the movement of water molecules in biological tissues. The movement of water molecules is not free, but is restricted by structures and objects in the tissue such as macromolecules, fibre walls and membranes [9-12]. Therefore, by analysing the movement of water molecules in images, we have the chance to study detailed structures in body tissues, in *vivo*, in a non-invasive way.

DTI provides information on the extent of diffusion anisotropy and its orientation, which can be used to generate a 3D vector field. Generally, at each voxel in DTI, the eigenvectors represent the diffusion directions, and the diffusion strength are proportional to the square roots of eigenvalues associating to the eigenvectors. By studying the connectivities along the vectors within the vector field, we are able to perform fibre tracking and reconstruct fibre tissues from within the human body.

1.2 Skeletal Muscle

Skeletal muscle is one of the most important tissues of the human body, playing a significant role in force and power generation that contribute to maintaining body postures and to controlling its movements [13]. By applying fibre tracking technology on skeletal muscle, we are able to explore its inner structure. Although the tracked fibres cannot be recognized as exactly the same as the muscle fascicles that the tracked fibres follow the orientation of muscle fascicles and therefore we are able to use these fibres as the equivalent of a bundle of muscle fascicles [14]. Figure 1 shows the results of performing fibre tracking within the medial gastrocnemius (MG) of a human calf.

1.3 Shape Analysis

Muscle fascicles can be reconstructed to 3D curves using DTI fibre tracking methods. A fascicle shape can be described by marking a serials points along the contour of a fascicle curve. Specifically a fascicle shape is a collection of k landmarks (points of interest) in an *m*-dimensional space (m = 3 in 3D space). In statistical shape analysis, Procrustes analysis is a landmark-based method for studying the shapes of objects [15]. The principal implementation of Procrustes analysis is to align different shapes in space using translation, rotation, scaling and reflection, then study their relationships. For example, to study the similarity of shapes is of main interest [1, 16]. Thus, we would like to explore a method that uses the Procrustes shape analysis to study the muscle fascicles data that we have collected in the human calf.

The aim of our research is to apply statistical shape analysis methods to quantitatively estimate the mean fascicle shapes and to explore the variation of fascicle shapes, and therefore find the connection between fascicle shapes and muscle states.



Fig. 1. (a) MG boundary (red) with corresponding MRI. (b) Tracked fascicles (green) within MG with corresponding MRI. (Color figure online)



Fig. 2. Foot plantarflexion and dorsiflexion. In our study, the ankle joint angles are about 67.5° and 107.3° , respectively. Image from www.pinterest.com

2 Procrustes Shape Analysis

The basic stratagem of the Procrustes shape analysis is to align the involved shapes as close as possible by transforming, rotating, scaling or reflecting and then to measure the distance between the shapes. The very first procedure of comparing the shapes of two or multiple objects is called superimpose. This procedure, also known as Procrustes superimposition (PS), is performed by applying optimally translating, rotating and scaling to the involved objects. In 2D or 3D space, the location, orientation, or size of the objects can be adjusted freely in order to make them to fit each other as close as possible. The objective of the operation is to minimize the measurement of the shape difference, which is called Procrustes distance between the objects. Full PS allows translation, rotation and scaling operation. Partial PS means the sizes of the objects keep unchanged. As an extra option, for both full PS and partial PS, there is an operation called 'reflection', which is able to produce a perfect superimposition for the objects with symmetric property (e.g. left ear and right ear, or left hand and right hand, in most of the cases).

Figure 3 shows the three steps to superimpose two object. By labeling four corresponding landmarks, A, B, C, D, and A', B', C', D', on both objects, respectively, we are able to see the procedure of Procrustes superimposition step by step.

2.1 Shape

Procrustes analysis is based on shapes, which exist everywhere and are used to describe the appearance of objects. **Shape** is all the geometrical information that remains when location, scale and rotational effects are removed from an object [1]. A shape is invariant to the Euclidean similarity transformations, which includes translation, scaling and rotation. Two objects can be recognized as in same shape if they match each other after translating, scaling and rotating.



Fig. 3. The three transformation steps of an ordinary Procrustes shape fit for two configurations of landmarks. (a) Scaling of both configurations to the same size; (b) Transposition to the same position of the center point of gravity; (c) Rotation to the orientation that provides the minimum sum of squared distances between corresponding landmarks. Original image based on https://en.wikipedia.org/wiki/Procrustes_analysis

2.2 Landmark and Configuration

Landmarks, a finite number of points selected along the outline of an object, are used to define a shape. A **configuration** is the set of landmarks on a particular object. The **configuration matrix** X is a $k \times m$ matrix of Cartesian coordinates of the k landmarks in m dimensions. The **configuration space** is the space of all landmark coordinates [1].

2.3 Pre-shape

To compute a distance between shapes it is necessary to standardise for size. In this study, we use the centered pre-shape Z_C which is defined by

$$\boldsymbol{Z}_{C} = \boldsymbol{C}\boldsymbol{X} / \parallel \boldsymbol{C}\boldsymbol{X} \parallel, \tag{1}$$

where

$$\boldsymbol{C} = \boldsymbol{I}_k - \frac{1}{k} \boldsymbol{1}_k \boldsymbol{1}_k^T \tag{2}$$

is the centering matrix, I_k is the $k \times k$ identity matrix (diagonal matrix with ones on the diagonal), $\mathbf{1}_k$ is the $k \times 1$ vector of ones and $|| \mathbf{A} || = \sqrt{trace(\mathbf{A}^T \mathbf{A})}$ is the Euclidean norm.

2.4 Procrustes Distances

Procrustes distance is a method to describe the similarity of different shape objects. Consider two configuration matrices from k points in m dimensions X_1 and X_2 with pre-shapes Z_1 and Z_2 . We minimize over rotations and scale to find the closest Euclidean distance between Z_1 and Z_2 . There are two types of Procrustes distances. The full Procrustes distance between two shapes considers translating, rotating, as well as scaling for superimposition. The partial Procrustes distance applies translating and rotating but not scaling for superimposition. The full Procrustes distance between X_1 and X_2 is

$$d_F(\boldsymbol{X}_1, \boldsymbol{X}_2) = \inf_{\boldsymbol{\Gamma}, \boldsymbol{\beta}, \boldsymbol{\gamma}} \| \boldsymbol{Z}_2 - \boldsymbol{\beta} \boldsymbol{Z}_1 \boldsymbol{\Gamma} - \boldsymbol{1}_k \boldsymbol{\gamma}^T \|,$$
(3)

where the real value $\beta > 0$ is the scale parameter, $\boldsymbol{\Gamma}$ is an $m \times m$ rotation matrix (in the special orthogonal group SO(m)), and the real vector $\boldsymbol{\gamma}$ is an $m \times 1$ location vector. The partial Procrustes distance between \boldsymbol{X}_1 and \boldsymbol{X}_2 is

$$d_P(\boldsymbol{X}_1, \boldsymbol{X}_2) = \inf_{\boldsymbol{\Gamma}, \boldsymbol{\gamma}} \parallel \boldsymbol{Z}_2 - \boldsymbol{Z}_1 \boldsymbol{\Gamma} - \boldsymbol{1}_k \boldsymbol{\gamma}^T \parallel.$$
(4)

2.5 Ordinary and Generalised Procrustes Analysis

Procrustes shape analysis aims to match different configurations with similarity transformations to be as close as possible according to Euclidean distance by using the least-squares algorithm. Technically, Procrustes analysis can be further classified into two types, which are Ordinary Procrustes Analysis (OPA) and Generalized Procrustes Analysis (GPA) [1].

OPA is an operation that compares two shapes, or compares a set of shapes with a selected reference shape. The involved shapes need to perform superimposition, which includes removing the translational, rotational and uniform scaling components in order to transform them into a member of an equivalent class which can be used for comparison [17]. The full OPA solution to the minimisation of Eq. 3 is given by $(\hat{\gamma}, \hat{\beta}, \hat{\Gamma})$ where

$$\hat{\boldsymbol{\gamma}} = 0 \tag{5}$$

$$\hat{\boldsymbol{\Gamma}} = \boldsymbol{U}\boldsymbol{V}^T \tag{6}$$

where

$$\boldsymbol{X}_{2}^{T}\boldsymbol{X}_{1} = \| \boldsymbol{X}_{1} \| \| \boldsymbol{X}_{2} \| \boldsymbol{V}\boldsymbol{\Lambda}\boldsymbol{U}^{T}$$

$$\tag{7}$$

with $\boldsymbol{\Lambda}$ a diagonal $m \times m$ matrix of positive elements, \boldsymbol{U} and \boldsymbol{V} are from the singular value decomposition of $\boldsymbol{X}_2^T \boldsymbol{X}_1$. Furthermore,

$$\hat{\beta} = \frac{trace(\boldsymbol{X}_{2}^{T}\boldsymbol{X}_{1}\hat{\boldsymbol{\Gamma}})}{trace(\boldsymbol{X}_{1}^{T}\boldsymbol{X}_{1})}.$$
(8)

The partial OPA has the same solution for the location vector and the rotation matrix as in the full OPA.

GPA applies Procrustes analysis to optimally superimpose more than two objects, instead of superimposing them to an arbitrarily selected shape. GPA is able to generate the mean shape of a set shapes automatically by using an iterative approach. OPA and GPA perform the same superimposition operations. The difference between them is the method to determine the reference orientation for the objects, which is arbitrarily selected in OPA but is optimally determined in GPA. When only two shapes are compared, GPA is equivalent to OPA [1,18]. Both OPA and GPA have the options of full and partial sub-types. Their difference is whether a scaling operation is performed.

3 Procrustes Methods for Fascicle Shape Analysis

Muscle fascicles are equivalent to 3D spatial curves. As we have known, a tracked fascicle is composed of a certain number of consecutive voxels, which can be recognized as landmarks along the fascicle shape. To demonstrate the idea of Procrustes analysis, we will carry out experiments using artificial 3D curves.

3.1 Landmark Construction for Fascicle Shapes Using Interpolation

We need to define a configuration matrix for a fascicle shape. The configuration matrix is a $k \times m$ matrix of the k landmarks in m dimensions (in our case m = 3). Also, the fascicle shapes need to have the same landmarks (same k value for all fascicle shapes) before performing Procrustes analysis. In this study, we interpolate fascicles which are shorter than the longest fascicle.

Refer to Fig. 4(a). Consider there are three fascicles with different length in muscle. The diamonds on fascicles indicate the original position of voxels, and the fascicles are in the length of 3(red), 5(green) and 7(blue), respectively. Since they are of different lengths, we interpolate the fascicles, for instance, to 20 landmarks. The circle marks on each fascicle show the results of interpolation. By collecting the coordinates of all landmarks on fascicles and storing them into configuration space, which is a $20 \times 3 \times 3$ matrix (k = 20, m = 3, shape = 3), we are able to do Procrustes analysis on those artificial curves.

3.2 Example of OPA

Let's perform ordinary Procrustes analysis first. In Fig. 4(a), we choose Fascicle 1 (red) and Fascicle 3 (blue), where Fascicle 3 works as a reference shape. To match Fascicle 1 and 3, all three transformations (translation, rotation and scale) are carried out. The function matches involved configurations by least squares. Figure 4(b) and (c) show the visualized result of full and partial OPA. After performing full OPA, we can get an estimated rotation matrix

$$\hat{\boldsymbol{\Gamma}} = \begin{pmatrix} 1 & 0 & 0\\ 0 & 0.8618 & -0.5072\\ 0 & 0.5072 & 0.8618 \end{pmatrix}$$
(9)

and an estimated scale factor of $\hat{\beta} = 2.8326$. The centered configuration of Fascicle 3 (blue) and its matched configuration of Fascicle 1 (red) are rendered by 'square' marks and '*' marks at position (b) in the figure, respectively. The black cross is the gravity center (origin) of the superimposed configurations.

Look at the image, compared with the original shapes, the reference shape Fascicle 3 (blue) has no shape change but its location is aligned to the origin according to its gravity center. At the same time, the Fascicle 1 (red) has been translated (matching the origin), rotated (applying matrix $\hat{\boldsymbol{\Gamma}}$) and scaled (applying scale factor $\hat{\boldsymbol{\beta}}$) in order to match with the reference shape.



Fig. 4. (a) Example fascicles for performing OPA between Fascicle 1 (red) and Fascicle 3 (blue), where Fascicle 3 (blue) is the reference shape. (b) Re-allocated reference shape (blue) and its matched shape of Fascicle 1 (red) by using full OPA. (c) Re-allocated reference shape (blue) and its matched shape of Fascicle 1 (red) by using partial OPA. The black cross is the common gravity center of them. (Color figure online)

As a comparison, after performing partial OPA, the estimated rotation matrix

$$\hat{\boldsymbol{\Gamma}} = \begin{pmatrix} 1 & 0 & 0\\ 0 & 0.8618 & -0.5072\\ 0 & 0.5072 & 0.8618 \end{pmatrix}$$
(10)

is exactly the same as the $\hat{\boldsymbol{\Gamma}}$ in full OPA. Since there is no scale changing required, the scale factor $\hat{\boldsymbol{\beta}} = 1$. Similarly, the centered configuration of fascicle 3 (blue) and its matched configuration of fascicle 1 (red) are rendered at position (c) in the figure and the black cross is the gravity center (origin) of the superimposed configurations.

From the parameters and image, we can see that in partial OPA, the only difference is that there is no scale factor applied to the configuration (red) which is to be matched with the reference shape (blue).

3.3 Example of GPA

Essentially, GPA performs a series of OPA operations iteratively to find the optimized superimposition as the reference for all involved shapes; we call this the mean shape.

Please refer to Fig. 5, which shows the visualized results of performing a full GPA and a partial GPA, respectively. Similar to above figures, the original fascicles are drawn at left side (a). The black fascicles at the right side, (b) and (c), with square marks, are the mean shapes and the cross signs are the gravity centers. It is notable that mean shapes generated by full and partial GPAs have just a little difference.



Fig. 5. Performing GPA on the 3 example fascicles at left side (a). The 2 curves on right side are the mean shapes of the example fascicles by using full GPA (b) and partial GPA (c), respectively. Black crosses are the gravity centers of the mean shapes. (Color figure online)

3.4 Example of Procrustes Distance

Once the mean fascicle shape is obtained using the GPA method, we are able to calculate the Procrustes distance d_P between the original example fascicles and their mean shape. Table 1 lists the results based on the GPA example in Fig. 5. It is clear that in full GPA, the mean shape is closest to Fascicle 2 (green) and is very far from Fascicle 1 (red). On the other hand, in partial GPA, the mean shape is most similar to Fascicle 3 (blue) but very different from Fascicle 1 (red).

Table 1. Procrustes distances between the three original fascicles and their mean shapein Fig. 5.

	Fascicle 1 (red)	Fascicle 2 (green)	Fascicle 3 (blue)
Full	0.1211	0.0731	0.0856
Partial	0.1519	0.0642	0.0569

4 Real Application: Mean Fascicle Shapes in Medial Gastrocnemius

In DTI skeletal muscle studies, the number of fascicles which can be tracked is variable, which is determined by the tracking algorithm and parameters chosen. It is also affected significantly by the settings of the MRI machine, as well as by the position and posture of the scanned object. No matter what different results are obtained, the number of fascicles is normally in the thousands or more. In this scenario, we are more interested in the overall appearance of the fascicles than in their one to one relationship. In this context, we would like to perform Generalized Procrustes Analysis in our muscle fascicle research, that is to find the mean shape of the fascicles in a muscle group, and then study the relationship between individual fascicles and this optimal superimposition.

4.1 Data Preparation

The raw DTI data is provided by Dr Bart Bolsterlee [19, 20] at Neuroscience Research Australia (NeuRA). The DTI is scanned from the medial gastrocnemius (MG) in the left calf of 6 healthy adults at 2 different ankle positions, respectively. The ankle position is determined by ankle joint angle, which is the angle between the tibia and the sole of the foot. Please refer to Fig. 2. Assuming 90° is a neutral ankle joint orientation, angle values below and above 90° indicate plantarflexion (foot down) and dorsiflexion (foot up), respectively.

After performing fibre tracking [8] based on the DTI data we are able to reconstruct the fascicles within the MG. As above explained, Procrustes analysis can only accept data of the same dimensionality. However, the fascicles obtained by DTI tracking are of different lengths. Therefore, we need to perform data interpolation for the fascicles before Procrustes analysis, as introduced in Sect. 3.1. For these muscle fascicles, the actual solution is finding the longest fascicle first, then interpolating the rest of the fascicles individually in order to reach the identical data points of the longest one. For example, a muscle group has 3000 fascicles in 3D space and the length of the longest fascicle is 50. After interpolation, we will have a $50 \times 3 \times 3000$ data matrix available for Procrustes analysis.

By applying Generalised Procrustes Analysis to all fascicles in the MG, we will find the overall mean fascicle shape of the corresponding muscle. In the meanwhile, the Procrustes distances between each fascicle and the mean shape are calculated as well. As we have performed both full and partial GPA, a total of $6 \times 2 \times 2 = 24$ groups of data are collected. Each data group includes a mean fascicle shape matrix and a distances matrix which indicate the similarity between individual fascicle and the mean shape.



Fig. 6. Coronal (front-back) view of selected MG fascicle shapes from a participant at foot-down position). (a) is the mean fascicle shape in MG obtained using GPA. (b) to (e) are the fascicles with 25th, 50th, 75th and 100th percentile Procrustes distances from the mean shape. (Color figure online)

4.2 Results Analysis

We would like to treat the mean shape of the fascicles in a MG as the overall fascicle shape of this particular muscle. Figure 6 demonstrates the variability of fascicle shapes in one participant's MG at one foot angle. The left (red) is the mean fascicle shape which has a (nearly) straight line shape. The second left (b) is the fascicle with the 25th percentile Procrustes distance from the mean (red). It is clear that the 25th percentile fascicle is similar to the mean fascicle with a slight difference in orientation. The third left (c) is the fascicle with the median percentile Procrustes distance from the mean and has a bump in the middle. The fourth left (d) is the fascicle with 75th percentile Procrustes distance which is longer than the mean fascicle and are more curly. The very right (e) is the fascicle with the largest Procrustes distance from the mean fascicle. This fascicle has a hook shape and is the most different one to the mean shape in the MG region.

In order to observe the shape changes of muscle fascicles when the foot is at different positions, we separate the muscle fascicles into two groups by using k-means clustering algorithm, where K = 2. Clustering is based on the Procrustes distance, or say shape similarity, between each fascicle and the mean shape. One of the groups includes those fascicles which are more similar (nearer in distance) to the mean shape, while the other group is the fascicles with relatively lower similarity (farther in distance). Here we classify the two groups as 'Group Near' and 'Group Far'.

Figure 7 and Fig. 8 show the visualized results we have obtained for one participant. Figure 7 shows the results of using partial GPA and Fig. 8 shows the full GPA results. In each figure, the columns are different foot positions (left: foot down, right: foot up). To make the figures clearer, we render the fascicles in 'Group Near' in black and the fascicles in 'Group Far' in white. As an initial visual impression, we can see that, compared to the foot is at the 'down' position (left column), when the foot is at the 'up' position (right column), the fascicles in 'Group Near', which are in black, are evidently more than those in 'Group Far'. This phenomenon is applied to all participants.

Partial GPA		Obj 1	Obj 2	Obj 3	Obj 4	Obj 5	Obj 6
Foot down	Distance to mean shape	0.0505	0.0532	0.0524	0.0527	0.0524	0.0504
	Fascicles in group near	41.61%	44.67%	43.80%	51.66%	51.88%	46.23%
Foot up	Distance to mean shape	0.0501	0.0519	0.0504	0.0510	0.0509	0.0503
	Fascicles in group near	55.78%	53.25%	54.17%	55.02%	55.65%	56.30%

Table 2. Partial GPA statistics for the muscle fascicles when foot is at 2 positions.

Full GPA		Obj 1	Obj 2	Obj 3	Obj 4	Obj 5	Obj 6
Foot down	Distance to mean shape	0.0506	0.0532	0.0525	0.0527	0.0524	0.0505
	Fascicles in group near	42.26%	44.08%	43.58%	50.34%	52.04%	47.88%
Foot up	Distance to mean shape	0.0501	0.0519	0.0504	0.0510	0.0509	0.0503
	Fascicles in group near	56.13%	53.40%	55.08%	55.28%	55.29%	55.16%

Table 3. Full GPA statistics for the muscle fascicles when foot is at 2 positions.

Numerically, for each participant, we calculate the mean distance from all fascicles in the muscle to their mean shape, and the percentage of the fascicles in the 'Group Near'. The values are listed in the tables. Table 2 corresponds to Fig. 7 and Table 3 corresponds to Fig. 8.



Fig. 7. Figure shows the percentage of the fascicles which are close to the mean shape. Images show 1 participant \times 2 positions, using partial GPA and k-means (K = 2) clustering algorithm.



Fig. 8. Figure shows the percentage of the fascicles which are close to the mean shape. Images show 1 participant \times 2 positions, using full GPA and k-means (K = 2) clustering algorithm.



Fig. 9. Fascicle shape similarity percentage chart while the foot is in different positions, using partial GPA (a) and full GPA (b), respectively. In both plots, blue (on the left): foot at 'down' position; red (on the right): foot at 'up' position. (Color figure online)

From the values in the tables, we can tell that for all participants, when the foot is at the 'up' position, the overall mean Procrustes distance (shape similarity) between fascicles and the mean shape is smaller than that when the foot is at the 'down' position. Meanwhile, the percentage of fascicles in 'Group Near' is higher when the foot is at the 'up' position. We therefore can



Fig. 10. Boxplots show the distribution of Procrustes distances between each fascicle and the mean shape, using partial GPA (a) and full GPA (b), respectively. Boxes in each plot present 6 participants \times 2 foot positions. In both plots, the distances range between 25% and 75% percentile are narrower at the 'up' position.

conclude that when the foot is being raised up, the MG muscles stretch longer, and the fascicles' shapes in the muscle are becoming more similar to each other. In order to give an intuitive understanding for the information in above tables, the percentages of fascicle similarity for all participants with the foot at both positions are visualized by bar charts in Fig. 9a and Fig. 9b. Furthermore, we verify the collected data using boxplots. Figure 10a and Fig. 10b show the Procrustes distance distribution between each fascicle and the mean shape. The boxplots are based on partial GPA and full GPA, respectively. Each plot presents 6 participants \times 2 foot positions. In these plots we can see that the range of distances between 25% percentile and 75% percentile for each participant is narrower when the foot is at the 'up' position. This observation supports the conclusion we have mentioned above. We notice in partial and full GPA, the changes of data distribution are small.

5 Summary and Further Work

Procrustes analysis has been widely used in shape analysis. We have explored the potential application of this technique in DTI skeletal muscle research. In this valuable attempt, we have found some meaningful conclusions which primarily answer what are the mean fascicle shapes in MG region at different ankle positions and how fascicle shape varies within this muscle region. Although the samples used in the research are limited, the results from all participants are obvious and identical.

Procrustes analysis is a useful method to study the shape of objects. A further research topic is to use this technique to observe and analysis more muscle groups, including the shapes of muscle fascicles and muscle boundaries. For example, we are interested in studying the shape properties of same muscle group on different participants, and, different muscle groups on same participant. We are also interested in the relationship between the changing of muscle shape and force generation. Furthermore, applying machine learning in shape analysis is a very attractive research direction.

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