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SHORT COMMUNICATION

A QUANTITATIVE, THREE-DIMENSIONAL METHOD FOR ANALYZING ROTATIONAL MOVEMENT FROM SINGLE-VIEW MOVIES

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The study of animal movement is an important aspect of functional morphological research. The three-dimensional movements of (parts of) animals are usually recorded on two-dimensional film frames. For a quantitative analysis, the real movements should be reconstructed from their projections. If movements occur in one plane, their projection is distorted only if this plane is not parallel to the film plane. Provided that the parallel orientation of the movement with respect to the film plane is checked accurately, a two-dimensional method of analysis (ignoring projection errors) can be justified for quantitative analysis of *planar* movements.

Films of movements of skeletal elements of the fish head have generally been analyzed with the two-dimensional method (e.g. Sibbing, 1982; Hoogenboezem *et al.* 1990; Westneat, 1990; Claes and de Vree, 1991), which is justifiable for planar movements. Unfortunately, the movements of the head bones of fish are often strongly non-planar, e.g. the movement of the pharyngeal jaws and the gill arches. The two-dimensional method is inappropriate for studying such complex movements (Sibbing, 1982; Hoogenboezem *et al.* 1990). For a *qualitative* description of movement patterns, the conditions for the use of the two-dimensional method may be somewhat relaxed.

When two (or more) views of a movement are recorded simultaneously, the three-dimensional movements can readily be reconstructed using two two-dimensional images (e.g. Zarnack, 1972; Nachtigall, 1983; van Leeuwen, 1984; Drost and van den Boogaart, 1986). However, because of technical (and budget) limitations, simultaneous views of a movement cannot always be shot. In this paper, a method is presented for reconstructing the three-dimensional orientation and rotational movement of structures using single-view films and for calculating rotation in an object-bound frame. Ellington (1984) presented a similar method for determining three-dimensional wing movements from single-view films of flying insects. Ellington's method is based upon the bilateral symmetry of the wing movements. The present method does not depend on symmetry and can be applied to a variety of kinematic investigations. It eliminates a systematic error: the projection error. The measuring error is not discussed; it is the same in the two-dimensional and three-dimensional method of analysis.

Key words: cinematography, projection errors, vector method.

The three-dimensional method of analysis can only be applied when the following general requirements are fulfilled. (1) The magnification of the projection of the object should be known (e.g. with the aid of a scale bar parallel to the film plane). (2) At least two markers should always be visible in each structure to be analyzed. These markers should be as far apart as possible in the direction of the movement under study. Markers may be conspicuous and well-defined anatomical points or artificial points (e.g. surgically implanted pieces of platinum, which are commonly used in X-ray cinematography). (3) The distance between the markers in each structure should be known accurately in each frame (a constant distance is most convenient). This can be determined in the anaesthetized animal. (4) One should know whether the structures are pointing 'up' or 'down' with respect to the film plane. This cannot always be determined from the film frames. The easiest way to solve this problem is to make sure that the angle between each structure and the film plane stays well within the range 0–180 ˚; in other words, to make sure that the structure is either pointing 'up' or 'down' during the entire film sequence. For rotations with very large amplitude, this may be impossible. In general, direct or video-recorded observation of the animal during filming is enough to judge whether a particular film sequence is suitable for analysis (e.g. when the animal turns on its back the sequence may not be suitable).

If a structure has only two marker points, axial rotation (rotation around the line that connects the markers) cannot be measured. If this movement component is the object of study, a third marker point (obviously not in line with the other two markers) is necessary. I will only discuss the calculations for structures with two markers. The calculations with three markers are essentially the same. (With a third marker, two vectors can be defined for each structure, $\mathbf{G}_1$ and $\mathbf{G}_2$. Axial rotation is measured by calculating, in each frame, the component of $\mathbf{G}_2$ perpendicular to $\mathbf{G}_1$. The angle between these perpendicular components in subsequent frames is the axial rotation.)

To avoid an entirely abstract treatment of the method, it is illustrated by the movement of a gill arch of a white bream (*Blicca bjoerkna*). Two platinum markers were inserted in each gill arch, the copula communis (the fused basibranchials that connect the gill arches mid-ventrally) and the skull. The skull was the reference structure. All the above general requirements were fulfilled (for details and error analysis, see van den Berg *et al.* 1994).

The film-plane is the x,y-plane. The z-axis is perpendicular to this plane. All calculations in this paper are performed in this x,y,z-frame. The two markers in each structure define a vector, $\mathbf{G}$. For example, $\mathbf{G}$ may represent a gill arch. One marker is translated to the origin (0,0,0). The coordinates of the other marker are (x,y,z). $\mathbf{G}$ can now be expressed in terms of x, y and z. Coordinates x and y are determined directly from each film frame (Fig. 1A,B). The value of z is calculated using Pythagoras' rule (Fig. 1C):

$$z = \pm\sqrt{\mathbf{G}^2 - x^2 - y^2}\,, \qquad (1)$$

where $\mathbf{G}^2$ (=the length of $\mathbf{G}$ squared) and the sign of z are known (general requirements 3 and 4) (see Ellington, 1984).

When two structures are connected with a single joint (e.g. a gill arch and the copula communis), the three-dimensional angle α between these structures can be calculated as the angle between the vectors $\mathbf{G}_1$ and $\mathbf{G}_2$ representing these structures. If there is no

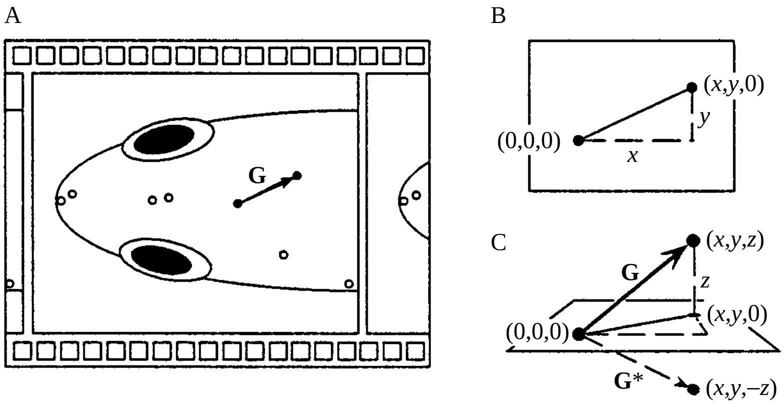

Fig. 1. (A) The markers in the fish head are indicated as small circles in this schematic film frame. The two black markers (at opposite ends of a gill arch) define a vector **G** (not in the film plane). (B) The *x* and *y* coordinates of **G** are calculated from its projection onto the film plane (=*x*,*y*-plane). (C) In this view, the film plane from B is shown from the side. The *z* coordinate of **G** is calculated with Pythagoras' rule, given the length of **G** and the orientation of **G** ('up' or 'down') with respect to the film plane. The orientation of **G** (i.e. the sign of *z*) must be known. The wrong direction of **G** is indicated as **G***.

marker exactly in the joint, the coordinates of the joint should be calculated using the coordinates of other marker points. The cosine of the angle α between $\mathbf{G}_1$ and $\mathbf{G}_2$ is given by:

$$\cos\alpha = \frac{\mathbf{G}_1 \cdot \mathbf{G}_2}{G_1 G_2} = \frac{x_1x_2 + y_1y_2 + z_1z_2}{G_1 G_2}\,, \qquad (2)$$

where x_1,y_1,z_1 and x_2,y_2,z_2 are the coordinates of vectors $\mathbf{G}_1$ and $\mathbf{G}_2$, respectively, and $G_1$and G_2 are the lengths of vectors $\mathbf{G}_1$and $\mathbf{G}_2$ (scalar).

The movement of a gill arch in a series of film frames (a film sequence) is the sum of its movement with respect to the skull and the movement of the skull with respect to the film frame. The separate components are interesting; their sum is not. Therefore, we want to separate these two components.

The movement of the skull can be split into a translation and a rotation component. The distance between the skull and the gill arches is not constant and is unknown. Therefore, the position (translation component) of the gill arches cannot be calculated relative to the skull in single-view films. However, rotation (e.g. depression) of the gill arches *can* be corrected for rotation of the skull.

The vector representing the skull in frame number *n* is $\mathbf{S}_\mathrm{n}$. The vector representing a gill arch is **G**. The angle between **G** and $\mathbf{S}_\mathrm{n}$ can easily be calculated from equation 2. However, we want to know the depression angle of **G**, which is the angle between **G** and a horizontal plane (plane *H*) in the fish (Fig. 2A,B). The calculation of such a depression angle is more complicated. First, a frame in which the fish is horizontal is chosen as the reference frame (Fig. 2A). In this frame, plane *H* is parallel to the *x*,*y*-plane (or film plane) (by definition). All vectors in the other film frames (Fig. 2B) must be transformed to the orientation of the reference frame (the method is described below). The depression angle of **G** equals the angle between the corrected vector **G** and the *x*,*y*-plane, since the *x*,*y*-

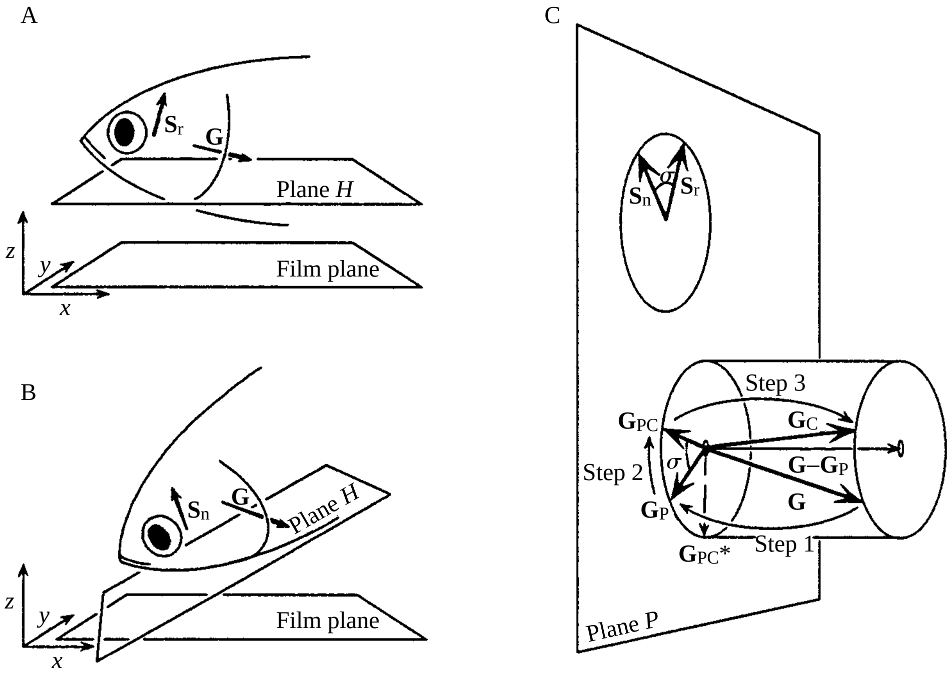

Fig. 2. The depression angle of the gill arch vector **G** is calculated in a fish-bound frame by correcting **G** for rotation of the skull vector **S**. (A) Vector **S** in the reference frame is $\mathbf{S}_r$. In the reference frame, plane *H* (the horizontal plane in the fish-bound frame) is parallel to the *x*,*y*-plane (by definition). (B) In film frame number *n*, the skull (vector $\mathbf{S}_n$) has rotated with respect to $\mathbf{S}_r$ over an angle σ. Plane *H* has also rotated over angle σ. The orientation of plane *H* with respect to vector **S** is unaltered. Vector **G** has to be transformed to the reference orientation given in A. (C) Vectors $\mathbf{S}_r$ and $\mathbf{S}_n$ define a plane *P*. This plane can have any orientation, depending on the way the skull has rotated (a combination of pitch, roll and yaw). **G** is projected on plane *P* ($\mathbf{G}_P$; step 1), rotated over angle σ ($\mathbf{G}_{PC}$; step 2) and restored to its original length ($\mathbf{G}_C$; step 3), by adding $\mathbf{G}-\mathbf{G}_P$. $\mathbf{G}_{PC}$* is the wrong solution of $\mathbf{G}_{PC}$ (rotated over angle $-\sigma$ instead of σ).

plane is always parallel to plane *H* after correction. The correction method is based on the movement of the skull vector $\mathbf{S}_n$ with respect to its reference orientation $\mathbf{S}_r$. The direction of $\mathbf{S}_r$ should preferably be perpendicular to the film frame (see Appendix).

By positioning $\mathbf{S}_r$ and $\mathbf{S}_n$ tail-to-tail, a plane *P* can be defined (Fig. 2C). Plane *P* is the plane of movement of the skull; it is unrelated to the film plane. The amount of movement is expressed as the angle σ between $\mathbf{S}_r$ and $\mathbf{S}_n$. Angle σ is a combination of the pitch, roll and yaw of the skull. Since $S_r=S_n$ (requirement 3), $\cos\sigma$ is given by:

$$\cos\sigma = \frac{\mathbf{S}_r \cdot \mathbf{S}_n}{S_r^2}\ . \tag{3}$$

In each film frame, **G** is transformed from the $\mathbf{S}_n$ orientation to the $\mathbf{S}_r$ orientation in three steps (Fig. 2C): step 1, **G** is projected on plane *P* ($\mathbf{G}_P$); step 2, $\mathbf{G}_P$ is rotated over angle σ ($\mathbf{G}_{PC}$); step 3, using $\mathbf{G}_{PC}$, the corrected direction of **G** ($\mathbf{G}_C$) is calculated; note that G_C=G.

When this is done, the depression angle of the gill arch is the angle between $\mathbf{G}_C$ and the *x*,*y*-plane. Note that, in the calculations below, the coordinates are not transformed to a frame defined by plane *P*, but always remain defined in the original *x*,*y*,*z*-frame of the film plane.

*Step 1: projection of vector **G** on plane* P. Just like any vector in plane *P*, vector $\mathbf{G}_P$ must be a linear combination of $\mathbf{S}_r$ and $\mathbf{S}_n$:

$$\mathbf{G}_P = \alpha_1 \mathbf{S}_r + \alpha_2 \mathbf{S}_n \,, \tag{4a}$$

where α_1 and α_2 are scalar factors.
$\mathbf{G}_P$ is a perpendicular projection of $\mathbf{G}$, therefore:

$$\begin{aligned} (\mathbf{G} - \mathbf{G}_P)\cdot\mathbf{S}_r &= 0\,, \\ (\mathbf{G} - \mathbf{G}_P)\cdot\mathbf{S}_n &= 0\,. \end{aligned} \tag{4b}$$

Substituting equation 4a into equation 4b gives two equations with two unknowns (α_1, α_2):

$$\begin{aligned} \alpha_1(\mathbf{S}_r\cdot\mathbf{S}_r) + \alpha_2(\mathbf{S}_r\cdot\mathbf{S}_n) &= \mathbf{G}\cdot\mathbf{S}_r\,, \\ \alpha_1(\mathbf{S}_r\cdot\mathbf{S}_n) + \alpha_2(\mathbf{S}_n\cdot\mathbf{S}_n) &= \mathbf{G}\cdot\mathbf{S}_n\,. \end{aligned} \tag{4c}$$

Using these equations, α_1 and α_2 and hence $\mathbf{G}_P$ can be determined.

Step 2: rotation of vector $\mathbf{G}_P$ over angle σ. The coordinates of $\mathbf{G}_{PC}$ (three unknowns: x_{PC}, y_{PC}, z_{PC}) are calculated using three equations, which are based on three conditions for the rotation (see Fig. 2C): condition 1, $\mathbf{G}_{PC}$ has the same length as $\mathbf{G}_P$; condition 2, $\mathbf{G}_{PC}$ is rotated over angle σ; condition 3, $\mathbf{G}_{PC}$ lies in plane *P*.

Condition 1: $G_{PC}=G_P$ or:

$$x_{PC}^2 + y_{PC}^2 + z_{PC}^2 = x_P^2 + y_P^2 + z_P^2\,. \tag{5}$$

Condition 2: $\mathbf{G}_{PC}$ is rotated over angle σ; combined with $G_{PC}=G_P$ (condition 1):

$$\cos\sigma = \frac{\mathbf{G}_P\cdot\mathbf{G}_{PC}}{G_P^2}\,,$$

combined with equation 3:

$$\mathbf{G}_P\cdot\mathbf{G}_{PC} = \frac{G_P^2}{S_r^2}\,\mathbf{S}_r\cdot\mathbf{S}_n\,,$$

or:

$$x_P x_{PC} + y_P y_{PC} + z_P z_{PC} = \frac{G_P^2}{S_r^2}\,(x_{Sr}x_{Sn} + y_{Sr}y_{Sn} + z_{Sr}z_{Sn})\,. \tag{6}$$

Condition 3: $\mathbf{G}_{PC}$ lies in plane *P*; all vectors in plane *P* are perpendicular to $\mathbf{S}_r\times\mathbf{S}_n$, therefore:

$$\mathbf{G}_{PC}\cdot(\mathbf{S}_r\times\mathbf{S}_n) = 0\,,$$

or:

$$x_{PC}x_{Sr\times Sn} + y_{PC}y_{Sr\times Sn} + z_{PC}z_{Sr\times Sn} = 0\,. \tag{7}$$

Combination of equations 5, 6 and 7 yields a quadratic equation with two solutions for $\mathbf{G}_{PC}$. These solutions represent rotation over angle σ in both directions in plane *P*

(Fig. 2C). The right solution is found by considering that the angle between $\mathbf{S}_r$ and $\mathbf{G}_{PC}$ should equal the angle between $\mathbf{S}_n$ and $\mathbf{G}_P$ (see Fig. 2C). Combined with S_r=S_n and G_{PC}=G_P we find:

$$\mathbf{S}_r \cdot \mathbf{G}_{PC} = \mathbf{S}_n \cdot \mathbf{G}_P\,. \tag{8}$$

Step 3: restoring $\boldsymbol{G}_{PC}$ *to its original length.* To put $\mathbf{G}_{PC}$ 'back in space', we simply add the part of **G** that is perpendicular to plane *P*. This part, $\mathbf{G}-\mathbf{G}_P$, is not affected by the rotation in plane *P* (Fig. 2C):

$$\mathbf{G}_C = \mathbf{G}_{PC} + (\mathbf{G} - \mathbf{G}_P)\,. \tag{9}$$

Note that the effect on **G** of the above correction for skull rotation is dependent on the angle between **G** and plane *P*. When this angle is large, the effect of the correction is small. When the angle is 90˚, its effect is even nil, since $\mathbf{G}_C$ equals **G**.

MPW FORTRAN subroutines (for Macintosh computers) with the present calculations are available on request.

The example of the gill arch movements of white bream (van den Berg *et al.* 1994) illustrates the importance of the three-dimensional method of analysis. The abduction angle α between the left first gill arch and the copula communis was calculated using both the two-dimensional and the three-dimensional method. The projected angle α_p (two-dimensional method) was 5–20˚ larger and had an amplitude two times larger (!) than the real angle α (three-dimensional method). The amplitude of depression angle β_{uncor} (the angle between the gill arch and the film plane) was about 1.5 times smaller than that of angle β (the angle between the gill arch and a horizontal plane in the fish) because of the pitch of the fish during food intake. Furthermore, the data for β_{uncor} suggested that the gill arch was placed in a special depressed position prior to gulping, while β showed that this was an artefact.

The gill arch movement in our example consisted of a combination of abduction (angle α) and depression (angle β). The large differences between the two-dimensional and three-dimensional methods in the example clearly show that the latter method is essential for a quantitative analysis of such non-planar movements from single-view films. The three-dimensional method is also essential when substantial changes in the orientation of the animal occur. In the example, pitch was an integral part of the feeding behaviour of the white bream, which led to both quantitative and qualitative (e.g. so-called 'special position of the gill arch') errors when the two-dimensional method was used. The three-dimensional method allows us to calculate rotations in an object-bound frame. The three-dimensional method of analysis must be strongly advised for both quantitative and qualitative studies of animal movement.

List of symbols

G	vector representing a gill arch
$\mathbf{G}_P$	the projection of **G** on plane *P*
$\mathbf{G}_{PC}$	$\mathbf{G}_P$ corrected for rotation of vector **S**
$\mathbf{G}_C$	**G** corrected for rotation of vector **S**

Plane *H*	horizontal plane in the fish, which is parallel to the film plane when the fish is in the reference orientation
Plane *P*	plane of movement of the skull, defined by vectors $\mathbf{S}_r$ and $\mathbf{S}_n$
Film plane	*x*,*y*-plane
S	vector representing the skull
$\mathbf{S}_r$	reference orientation of **S**
$\mathbf{S}_n$	vector **S** in frame number *n*
x, y, z	*x*, *y* and *z* coordinates of vector **G**
x_P, y_P, z_P	*x*, *y* and *z* coordinates of vector $\mathbf{G}_P$
x_{PC}, y_{PC}, z_{PC}	*x*, *y* and *z* coordinates of vector $\mathbf{G}_{PC}$
x_C, y_C, z_C	*x*, *y* and *z* coordinates of vector $\mathbf{G}_C$
x_{Sr}, y_{Sr}, z_{Sr}	*x*, *y* and *z* coordinates of vector $\mathbf{S}_r$
x_{Sn}, y_{Sn}, z_{Sn}	*x*, *y* and *z* coordinates of vector $\mathbf{S}_n$
$x_{Sr\times Sn}, y_{Sr\times Sn}, z_{Sr\times Sn}$	*x*, *y* and *z* coordinates of vector $\mathbf{S}_r \times \mathbf{S}_n$
α_1, α_2	scalar factors to express $\mathbf{G}_P$ in terms of $\mathbf{S}_r$ and $\mathbf{S}_n$
α	angle between gill arch and copula communis
α_p	the projection of angle α on the film plane
β	angle between gill arch and plane *H*
β_{uncor}	angle between gill arch and the film plane
σ	angle between vectors $\mathbf{S}_r$ and $\mathbf{S}_n$
$\mathbf{G} \cdot \mathbf{G}$	notation for the dot product
$\mathbf{G} \times \mathbf{G}$	notation for the cross product
G	notation for the length of a vector (=|G|)

Appendix

Vector $\mathbf{S}_r$ should be perpendicular to the film plane. There are two reasons for this. (1) The length of the projection of vector **S** on the film plane is the length S multiplied by the cosine of the angle between vector **S** and the film plane. The cosine is most sensitive to rotation when the angle is approximately 90°. This holds true for vector **G** as well: one should preferably film in the direction parallel to **G**, rather than perpendicular to it. In the latter case, the projection on the film plane is very insensitive to rotation of **G** (see Ellington, 1984, pp. 46–47). In other words, movements perpendicular to the film plane may easily go unnoticed in that case. (2) Axial rotation around vector $\mathbf{S}_r$ is not measured. When $\mathbf{S}_r$ is perpendicular to the film plane, this unmeasured rotation component is rotation in the film plane (yaw, in the example). During analysis of the film frames, both the marker projections and the outline of the fish head are copied on paper. The yaw component of head rotation can easily be compensated for by always positioning the outline of the fish head in the same way on the data tablet. Furthermore, if there is still some rotation in the film plane, this has no influence on *depression* angles.

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