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Original Research Paper

VOLUME TENSOR ESTIMATION USING A VIRTUAL LINE GRID: STUDY OF A DEVELOPING PHEASANT BRAIN

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ABSTRACT

The volume tensor provides a robust estimate of the shape and orientation of an object in space. In this paper, we introduce Fakir method for estimating the tensor of an object in 3D data set based on the intersections of objects boundary with virtual lines. We calculate the precision of shape estimates by predicting the variance of estimators of integrals based on systematic sampling. To demonstrate the ability of the Fakir method, we measure changes in shape and orientation of compartments in the pheasant brain during development.

Keywords: bird brain, development, Fakir probe, magnetic resonance imaging, volume Minkowski tensor.

INTRODUCTION

The shape and orientation of an object can be expressed by an equivalent ellipsoid (Ziegel *et al.*, 2015) and calculated using a centered second moment volume tensor. Current volume tensor estimators use either a triangulated surface (Schröder-Turk *et al.*, 2011) or random sections (Rafati *et al.*, 2016). However, neither is suitable when handling volumetric data for an object that cannot be easily segmented. Our aim therefore was to design and evaluate a method that uses intersections with a spatial grid sparser then a voxel lattice, enabling intersections to be assessed interactively.

The virtual grid approach was originally designed to measure surface areas (Barbier, 1860; Cruz-Orive, 1997; Kubínová and Janáček, 1998). It was later implemented as the Fakir method for estimating the surface area and volume of 3D objects in images via 3D modalities such as confocal microscopy (Kubínová *et al.*, 1999), magnetic resonance imaging or computed tomography (Jirák *et al.*, 2015). The estimators are based on two propositions of integral geometry (Santaló, 1976): 1) the mean number of intersections of the object boundary with the grid is in direct proportion to the surface area of the object. 2) the mean length of the grid lines inside the objects is in direct proportion to the volume of the object; where the mean is assessed with respect to the random position of the grid. The coefficients of proportionality are the products of the grid length density (m^{-2}) and constants equal to one half and one, respectively.

Using randomly oriented grids, the variance of an estimator can be determined from the grid density and properties of the objects measured (Kendall, 1948; Hlawka, 1950; Matheron, 1965). For example, the variance of volume estimator using a spatial grid with an isotropic uniform random (IUR) position can be efficiently estimated from the asymptotic term proportional to the surface area of the object divided by the square of the length density (Janáček, 2006; 2008) with a constant characteristic for the arrangement of the grid lines.

Arranging a line grid is the best way of reducing further workload. One particularly efficient line arrangement is found in a garnet crystal, where the atoms are aligned in three orthogonal and four diagonal directions (O´Keefe, 1992).

The aim of this paper is to demonstrate the ability of the Fakir method to evaluate a volume tensor and effectively estimate the equivalent ellipsoid and orientation of an object. We propose an formula for precision of the semi-axes approximation. The variance of semi-axes is calculated from spatial grid density and object properties. The approximation is valid for bounded objects with finite surface area (Janáček and Jirák, 2019).

We apply the Fakir method to assess changes in shape in the compartments of a phaesant brain during development. The results of the study (Jirák *et al.*, 2015) reveal changes to the volume in divisions of the pheasant brain with ontogeny.

MATERIAL AND METHODS

ESTIMATION OF AN EQUIVALENT ELLIPSOID FROM LINE SEGMENTS

Let K be a measurable subset of 3-dimensional Euclidean space $\mathbb{R}^3$. The elements of the second-moment Minkowski volume tensor $\Phi_{3,2,0}(K) = \{\phi_{i,j}\}$ are defined by integrals (Hug *et al.*, 2008):

$$\phi_{i,j} = \frac{1}{2}\iiint\limits_K x_i x_j dx_1 dx_2 dx_3,\ i,j = 1\ldots 3. \quad (1)$$

We combine $\Phi_{3,2,0}$ with center of mass and volume of the object to obtain the translation-invariant centered second-moment volume tensor $\Theta(K) = \{\tau_{i,j}\}$ (Ziegel *et al.*, 2015):

$$\tau_{i,j} = \frac{2\phi_{i,j}}{V} - c_i c_j,\ i,j = 1\ldots 3, \quad (2)$$

where

$$c_i = \frac{m_i}{V},\ m_i = \iiint\limits_K x_i dx_1 dx_2 dx_3,\ i = 1\ldots 3 \quad (3)$$

are coordinates of the centre of mass and

$$V = \iiint\limits_K dx_1 dx_2 dx_3 \quad (4)$$

is the volume of K.

Eigenvalues and eigenvectors of the centered tensor provide information on shape and orientation of the object, respectively. Let λ_i and v_i, $i = 1\ldots 3$ be the eigenvalues and eigenvectors of the centered tensor $\Theta(K)$. The anisotropy of the set is characterized by the Procrustes anisotropy (*PA*) (Dryden *et al.*, 2009):

$$PA(K) = \sqrt{\frac{3}{2}\frac{\sum_{i=1}^{3}\left(\sqrt{\lambda_i} - \frac{1}{3}\sum_{j=1}^{3}\sqrt{\lambda_j}\right)^2}{\sum_{i=1}^{3}\lambda_i}}. \quad (5)$$

PA achieves values between 0 and 1.

For convenience, the centred tensor $T(K)$ can be visualised by the equivalent ellipsoid with semi-axes in direction v_i with lengths s_i, $i = 1\ldots 3$, (Ziegel *et al.*, 2015), where:

$$s_i = \sqrt{5\lambda_i}. \quad (6)$$

The Fakir estimate of the tensor entries $\widetilde{\phi}_{i,j}$, $i,j = 1\ldots 3$ uses line grid G with intensity L_V (m^{-2}), which is randomly shifted by uniform random vector U:

$$\widetilde{\phi}_{i,j} = \frac{1}{2L_V}\int\limits_{G+U\cap K} x_i x_j dH(x), \quad (7)$$

where H is the 1-dimensional Hausdorff measure. The values of s_i in Eq. 6 are estimated from the intersections of the grid $G+U$ with object K. Let the intersections of the set K and the grid consist of N line segments with endpoints $\mathbf{a}_k = (a_{k,1}, a_{k,2}, a_{k,3})$ and $\mathbf{b}_k = (b_{k,1}, b_{k,2}, b_{k,3})$ and let l_k be the length of the k-th segment. Calculating the estimate $\widetilde{\phi}_{i,j}$ in Eq. 7 as the sum of integrals over individual line segments gives:

$$\widetilde{\phi}_{i,j} = \tfrac{1}{12L_V}\sum_{k=1}^{N} l_k \left(2a_{k,i}a_{k,j} + a_{k,i}b_{k,j} + \right. \\ \left. + b_{k,i}a_{k,j} + 2b_{k,i}b_{k,j}\right),\ i,j = 1\ldots 3. \quad (8)$$

Estimates of the volume V, of the center of mass c and of the first volume moment $m = Vc$ are:

$$\widetilde{c}_i = \frac{\widetilde{m}_i}{\widetilde{V}},\ \widetilde{m}_i = \frac{1}{2L_V}\sum_{k=1}^{N} l_k\left(a_{k,i} + b_{k,i}\right), \quad (9)$$

$$\widetilde{V} = \frac{1}{L_V}\sum_{k=1}^{N} l_k. \quad (10)$$

Formulas in Eq. 8, 9 and 10 are discrete analogues of Eq. 1, 3 and 4, respectively. The natural estimate of centered tensor element is then:

$$\widetilde{\tau}_{i,j} = \frac{2\widetilde{\phi}_{i,j}}{\widetilde{V}} - \widetilde{c}_i\widetilde{c}_j. \quad (11)$$

We estimate the equivalent ellipsoid and its Procrustes anisotropy by plugging the eigenvalues $\widetilde{\lambda}_i$ of the estimated tensor $\widetilde{\tau}_{i,j}$ into formulas in Eq. 5 and 6.

PRECISION OF SEMI-AXES ESTIMATES

The precision of estimates of semi-axis is calculated from the variances and covariances of the tensor components, *i.e.* from variances of estimates of integrals of polynomials, and from covariances of simultaneous estimates of two such integrals. Special case (when the polynomial is constant) is known, because variance of the volume estimate by isotropic Fakir probe

$$\widetilde{V}(K,U,R) = \frac{1}{L_V}\int\limits_{RG+U\cap K} dH(x),$$

where U is random shift and R is random rotation, is approximately

$$\mathrm{var}\left(\widetilde{V}(K,U,R)\right) \cong C_G S(K) L_V^{-2} \quad (12)$$

where C_G is the grid constant (Kendall, 1948; Hlawka, 1950; Matheron, 1965; Janáček, 1999).

The grid constant C_G can be calculated from the Fourier transform of the grid (Janáček and Kubínová, 2010). For the optimized Fakir grids we have

$$C_G = \frac{1}{8\pi^3}\left(3\zeta\left(Z_2,4\right) - \frac{21}{2}\zeta\left(4\right)\right)$$

with value 0.02707533 for the threefold grid,

$$C_G = \frac{1}{8\pi^3}\left(4\zeta\left(A_2,4\right) - \frac{63}{4}\zeta\left(4\right)\right)$$

with value 0.02453877 for the fourfold grid and

$$C_G = \frac{1}{8\pi^3}\left(3\zeta\left(Z_2,4\right) + 4\zeta\left(A_2,4\right) - \right.$$
$$\left. - \frac{21}{8}\left(10+\sqrt{3}\right)\zeta\left(4\right)\right)$$

with value 0.0317757 for the sevenfold grid, where

$$\zeta\left(Z_2,s\right) = \sum\nolimits'^{\infty}_{i,j=-\infty}\left(i^2+j^2\right)^{-\frac{s}{2}}$$

is the Epstein zeta function of square point grid, $\zeta\left(Z_2,4\right) \cong 6.02681$,

$$\zeta\left(A_2,s\right) = \sum\nolimits'^{\infty}_{i,j=-\infty}\left(2\frac{i^2+ij+j^2}{\sqrt{3}}\right)^{-\frac{s}{2}}$$

is the Epstein zeta function of unit triangular point grid, $\zeta\left(A_2,4\right) \cong 5.78336$ and

$$\zeta\left(s\right) = \sum_{i=1}^{\infty} i^{-s}$$

is the Riemann zeta function, $\zeta\left(4\right) \cong 1.082323$.

A generalization of Eq. 12 yields the approximate formula for covariance of the estimates of integrals of complex functions

$$\widetilde{I}\left(f_i,U,R\right) = \frac{1}{L_V}\int\limits_{RG+U\cap K} f_i\left(x\right)dH\left(x\right),\ i,j = 1\ldots 2.$$

which is:

$$\begin{aligned}&\operatorname{cov}\left(\widetilde{I}\left(f_1,U,R\right),\widetilde{I}\left(f_2,U,R\right)\right) \cong \\ &\cong C_G \iint_{\partial K} f_1\left(x\right)\overline{f_2\left(x\right)}dS\left(x\right)L_V^{-2}.\end{aligned} \qquad (13)$$

The approximate expansions in Eq. 12 and 13 are obtained by replacing the function $\Phi\left(\sqrt{L_V}\right)$ with properties $\Phi \geq 0$ and $\lim_{x\to+\infty}\frac{1}{x}\int_0^x \Phi\left(y\right)\,dy = 1$, provided that the functions are smooth and have bounded supports and the set has finite perimeter, by constant equal to 1 in the variance formula (Janáček and Jirák, 2019, Theorem 4).

The surface integral in Eq. 13 can be estimated from intersections x_k of the Fakir probe with the surface of the set K as

$$\iint\limits_{\partial K} h\left(x\right)dS\left(x\right) \cong \frac{2}{L_V}\sum_{x_k\in RG+U\cap\partial K} h\left(x_k\right).$$

We obtain an approximation of $\operatorname{cov}\left(\widetilde{\tau}_{i,j},\widetilde{\tau}_{k,l}\right)$ for $i,j,k,l = 1\ldots 3$ by linearization of the formula

$$\widetilde{\tau}_{i,j} = \frac{2\widetilde{\phi}_{i,j}}{\widetilde{V}} - \frac{\widetilde{m}_i\widetilde{m}_j}{\widetilde{V}^2},\ i,j = 1\ldots 3$$

(Eq. 11), and by the use of bilinearity of covariance:

$$\begin{aligned}\operatorname{cov}\left(\widetilde{\tau}_{i,j},\widetilde{\tau}_{k,l}\right) \cong\ & \frac{4}{\widetilde{V}^2}\operatorname{cov}\left(\widetilde{\phi}_{i,j},\widetilde{\phi}_{k,l}\right) - \frac{4\widetilde{\phi}_{i,j}}{\widetilde{V}^3}\operatorname{cov}\left(\widetilde{\phi}_{k,l},\widetilde{V}\right)\\
&-\frac{4\widetilde{\phi}_{k,l}}{\widetilde{V}^3}\operatorname{cov}\left(\widetilde{\phi}_{i,j},\widetilde{V},\right) + \frac{4\widetilde{\phi}_{i,j}\widetilde{\phi}_{k,l}}{\widetilde{V}^4}\operatorname{var}\left(\widetilde{V}\right)\\
&-\frac{2\widetilde{m}_l}{\widetilde{V}^3}\operatorname{cov}\left(\widetilde{\phi}_{i,j},\widetilde{m}_k\right) - \frac{2\widetilde{m}_k}{\widetilde{V}^3}\operatorname{cov}\left(\widetilde{\phi}_{i,j},\widetilde{m}_l\right)\\
&-\frac{2\widetilde{m}_j}{\widetilde{V}^3}\operatorname{cov}\left(\widetilde{\phi}_{k,l},\widetilde{m}_i\right) - \frac{2\widetilde{m}_i}{\widetilde{V}^3}\operatorname{cov}\left(\widetilde{\phi}_{k,l},\widetilde{m}_j\right)\\
&+\frac{2\widetilde{m}_k\widetilde{m}_l}{\widetilde{V}^4}\operatorname{cov}\left(\widetilde{\phi}_{i,j},\widetilde{V}\right) + \frac{2\widetilde{m}_i\widetilde{m}_j}{\widetilde{V}^4}\operatorname{cov}\left(\widetilde{\phi}_{k,l},\widetilde{V}\right)\\
&+\frac{2\widetilde{\phi}_{i,j}\widetilde{m}_l}{\widetilde{V}^4}\operatorname{cov}\left(\widetilde{m}_k,\widetilde{V}\right) + \frac{2\widetilde{\phi}_{i,j}\widetilde{m}_k}{\widetilde{V}^4}\operatorname{cov}\left(\widetilde{m}_l,\widetilde{V}\right)\\
&+\frac{2\widetilde{\phi}_{k,l}\widetilde{m}_j}{\widetilde{V}^4}\operatorname{cov}\left(\widetilde{m}_i,\widetilde{V}\right) + \frac{2\widetilde{\phi}_{k,l}\widetilde{m}_i}{\widetilde{V}^4}\operatorname{cov}\left(\widetilde{m}_j,\widetilde{V}\right)\\
&-\frac{4\widetilde{\phi}_{i,j}\widetilde{m}_k\widetilde{m}_l}{\widetilde{V}^5}\operatorname{var}\left(\widetilde{V}\right) - \frac{4\widetilde{\phi}_{k,l}\widetilde{m}_i\widetilde{m}_j}{\widetilde{V}^5}\operatorname{var}\left(\widetilde{V}\right)\\
&+\frac{\widetilde{m}_j\widetilde{m}_l}{\widetilde{V}^4}\operatorname{cov}\left(\widetilde{m}_i,\widetilde{m}_k\right) + \frac{\widetilde{m}_j\widetilde{m}_k}{\widetilde{V}^4}\operatorname{cov}\left(\widetilde{m}_i,\widetilde{m}_l\right)\\
&+\frac{\widetilde{m}_i\widetilde{m}_l}{\widetilde{V}^4}\operatorname{cov}\left(\widetilde{m}_j,\widetilde{m}_k\right) + \frac{\widetilde{m}_i\widetilde{m}_k}{\widetilde{V}^4}\operatorname{cov}\left(\widetilde{m}_j,\widetilde{m}_l\right)\\
&-\frac{2\widetilde{m}_j\widetilde{m}_k\widetilde{m}_l}{\widetilde{V}^5}\operatorname{cov}\left(\widetilde{m}_i,\widetilde{V}\right) - \frac{2\widetilde{m}_i\widetilde{m}_k\widetilde{m}_l}{\widetilde{V}^5}\operatorname{cov}\left(\widetilde{m}_l,\widetilde{V}\right)\\
&-\frac{2\widetilde{m}_i\widetilde{m}_j\widetilde{m}_l}{\widetilde{V}^5}\operatorname{cov}\left(\widetilde{m}_k,\widetilde{V}\right) - \frac{2\widetilde{m}_i\widetilde{m}_j\widetilde{m}_k}{\widetilde{V}^5}\operatorname{cov}\left(\widetilde{m}_j,\widetilde{V}\right)\\
&+\frac{4\widetilde{m}_i\widetilde{m}_j\widetilde{m}_k\widetilde{m}_l}{\widetilde{V}^6}\operatorname{var}\left(\widetilde{V}\right).\end{aligned}$$

We apply Eq. 13 to the summands on the right side of the equation above and by proper grouping of factors we obtain

$$\operatorname{cov}\left(\widetilde{\tau}_{i,j},\widetilde{\tau}_{k,l}\right) \cong$$

$$\frac{C_G}{\widetilde{V}^2 L_V^2} \iint_{\partial K} \left(\left(x_i - \widetilde{c}_i \right) \left(x_j - \widetilde{c}_j \right) - \widetilde{\tau}_{i,j} \right) \left(\left(x_k - \widetilde{c}_k \right) \left(x_l - \widetilde{c}_l \right) - \widetilde{\tau}_{k,l} \right) dS(x) .$$

Now we can calculate the variance of semi-axes by the use of linearization of formulas for eigenvalues and linearization of Eq. 6.

Characteristic polynomial $P(\lambda)$ and invariants T, Q, D of the tensor Θ are related with τ_{ij} by formula:

$$P(\lambda) = \begin{vmatrix} \tau_{11}-\lambda & \tau_{12} & \tau_{13} \\ \tau_{12} & \tau_{22}-\lambda & \tau_{23} \\ \tau_{13} & \tau_{23} & \tau_{33}-\lambda \end{vmatrix} = = -\lambda^3 + T\lambda^2 - Q\lambda + D.$$

Partial derivatives of the invariants are then

$$\frac{\partial T}{\partial \tau_{ii}} = 1,\ \frac{\partial T}{\partial \tau_{ij}} = 0,$$

$$\frac{\partial Q}{\partial \tau_{ii}} = \tau_{jj} + \tau_{kk},\ \frac{\partial Q}{\partial \tau_{ij}} = -2\tau_{ij},$$

$$\frac{\partial D}{\partial \tau_{ii}} = \tau_{jj}\tau_{kk} + \tau_{jk}^2,\ \frac{\partial D}{\partial \tau_{ij}} = 2\tau_{ik}\tau_{jk} - 2\tau_{ij}\tau_{kk},$$

where $\{i,j,k\} = \{1,2,3\}$.

Partial derivatives of centered tensor eigenvalues are calculated solving the equations with partial derivatives of invariants:

$$\frac{\partial \lambda_1}{\partial \tau_{ij}} + \frac{\partial \lambda_2}{\partial \tau_{ij}} + \frac{\partial \lambda_3}{\partial \tau_{ij}} = \frac{\partial T}{\partial \tau_{ij}},$$

$$(\lambda_2+\lambda_3)\frac{\partial \lambda_1}{\partial \tau_{ij}} + (\lambda_1+\lambda_3)\frac{\partial \lambda_2}{\partial \tau_{ij}} + (\lambda_1+\lambda_2)\frac{\partial \lambda_3}{\partial \tau_{ij}} = \frac{\partial Q}{\partial \tau_{ij}},$$

$$\lambda_2\lambda_3\frac{\partial \lambda_1}{\partial \tau_{ij}} + \lambda_1\lambda_3\frac{\partial \lambda_2}{\partial \tau_{ij}} + \lambda_1\lambda_2\frac{\partial \lambda_3}{\partial \tau_{ij}} = \frac{\partial D}{\partial \tau_{ij}}.$$

The derivatives of eigenvalues are then:

$$\frac{\partial \lambda_k}{\partial \tau_{ij}} = (\lambda_l - \lambda_m)\left(\lambda_k^2 \frac{\partial T}{\partial \tau_{ij}} + \lambda_k \frac{\partial Q}{\partial \tau_{ij}} + \frac{\partial D}{\partial \tau_{ij}}\right) Det^{-1},$$

where (k,l,m) is $(1,2,3)$, $(2,3,1)$ or $(3,1,2)$ and

$$Det = \lambda_1^2(\lambda_2-\lambda_3) + \lambda_2^2(\lambda_3-\lambda_1) + \lambda_3^2(\lambda_1-\lambda_2).$$

Variance of eigenvalues $\widetilde{\lambda}_m$, $m = 1\ldots3$ is approximately:

$$\mathrm{var}\left(\widetilde{\lambda}_m\right) \cong \sum_{\substack{i,j,k,l=1 \\ i\le j, k\le l}}^{3} \frac{\partial \lambda_m}{\partial \tau_{ij}} \frac{\partial \lambda_m}{\partial \tau_{kl}} \mathrm{cov}\left(\widetilde{\tau}_{i,j}, \widetilde{\tau}_{k,l}\right).$$

Finally, the variance of semi-axes lengths $\widetilde{s}_m$, $m = 1\ldots3$ is approximately:

$$\mathrm{var}\left(\widetilde{s}_m\right) \cong \frac{5}{4\widetilde{\lambda}_m} \mathrm{var}\widetilde{\lambda}_m. \qquad (14)$$

VERIFICATION OF THE VARIANCE FORMULA

An ellipsoid with unequal semi-axes was measured using a sevenfold grid of random orientation and position. The variance in length of the semi-axes was predicted by Eq. 14 and calculated based on this measurement. The results presented in Tab. 1 show excellent performance of the error prediction.

Ellipsoid (n=100)	s_1	s_2	s_3
Prediction	0.33	0.28	0.23
Simulation	0.34	0.30	0.24

Table 1. *Estimate of ellipsoid semi-axes (s_1=50, s_2=40, s_3=30, in arbitrary units) using a Fakir sevenfold grid (L_V=0.01183) repeated 100x with random grid orientation and position. Mean standard deviation values (prediction calculated using Eq. 14) of the ellipsoid semi-axes and standard deviation of the simulated estimate are also shown. Standard deviation of the predicted standard deviation was 0.01 in all semiaxes.*

SELECTING THE GRID PARAMETERS

The precision of the semi-axis length estimates using selected parameters (grid type and L_V) and given objects (adult male phaesant forebrain and hatchling forebrain) is better than 0.5%. Semi-axis length estimate with predicted errors in two selected samples are presented in Tab. 2

Forebrain	s_1	s_2	s_3
Adult	12.16 (0.03)	7.95 (0.02)	6.48 (0.02)
Hatch.	7.18 (0.03)	5.21 (0.02)	4.76 (0.02)

Table 2. *Semi-axis estimates ($s_1 \geq s_2 \geq s_3$, in mm) of selected samples (forebrain of adult male phaesant and a hatchling) by Fakir sevenfold grid (L_V = 0.76 mm^{-2}) presented with predicted standard deviation (calculated using Eq. 14) in parentheses.*

RESULTS

ANALYSIS OF THE DEVELOPING PHEASANT BRAIN

The heads of 2 hatchlings, 4 juvenile and 6 adult ring-necked pheasants (Phasianus colchicus) were fixed in formalin before scanning. Brain images were acquired at high resolution (voxel volume = 0.002775 mm^3) using a 4.7 T magnetic resonance (MR) spectrometer (Bruker BioSpec) equipped with a commercially available resonator coil, and 3D Rapid Acquisition incorporating a Relaxation Enhancement (RARE) multi-spin echo sequence (Jirák *et al.*, 2015). MR images were analyzed using home-made Fakir software. The volume and surface area of each brain

	Forebrain	Midbrain	Hindbrain
Hatch. (2)	0.30 (0.01)	0.83 (0.01)	0.39 (0.02)
Juv. (4)	0.36 (0.01)	0.85 (0.01)	0.44 (0.01)
Adults (6)	0.43 (0.01)	0.83 (0.01)	0.36 (0.02)

Table 3. *Procrustes anisotropy PA of pheasant (hatchlings, juveniles and adults; number of samples in parentheses) brain compartments calculated according to Eq. 5. Anisotropy mean values are presented with the standard error of the mean. The differences in forebrain and hindbrain anisotropy between age groups were statistically significant (ANOVA $p < 0.01$).*

a) Forebrain	s_1	s_2	s_3
Hatch. (2)	7.0 (0.0)	5.2 (0.1)	4.5 (0.1)
Juv. (4)	10.3 (0.2)	7.2 (0.1)	6.3 (0.1)
Adults (6)	11.7 (0.1)	7.8 (0.1)	6.3 (0.1)

b) Midbrain	s_1	s_2	s_3
Hatch. (2)	8.4 (0.1)	3.3 (0.1)	2.2 (0.0)
Juv. (4)	10.7 (0.1)	4.1 (0.1)	3.0 (0.1)
Adults (6)	11.1 (0.2)	4.1 (0.1)	3.6 (0.1)

c) Hindbrain	s_1	s_2	s_3
Hatch. (2)	5.1 (0.0)	3.4 (0.1)	3.0 (0.2)
Juv. (4)	7.4 (0.0)	4.6 (0.1)	4.2 (0.0)
Adults (6)	7.5 (0.1)	5.2 (0.2)	4.5 (0.1)

Table 4. *Semi-axis lengths ($s_1 \geq s_2 \geq s_3$, in mm) of ellipsoids calculated according to Eq. 6 for the phaesant (a) forebrain (b) midbrain and (c) hindbrain (hatchlings, juveniles and adults; number of samples is in parentheses). Mean values are presented with the standard error of the mean in parentheses.*

division were measured interactively using sevenfold Fakir probe with a grid density of 0.76 mm^{-2} (for results see Jirák *et al.* (2015)). Structures of the avian brain were identified using a histological atlas (Karten *et al.*, 2013).

The major axis is oriented laterally in the forebrain and midbrain and rostrally in the hindbrain. The significant increase of PA in the forebrain (Tab. 3) is caused by the increase in the length of the semi-major axis, which was more pronounced that the lengths of the other semi-axes (Tab. 4a). The major change in forebrain shape during development was the increase in relative width. In the same manner, we conclude that the hindbrain significantly elongated in rostral direction during brain development. The predicted errors of the semi-axis estimates for selected brain compartments and grid density 0.76 mm^{-2} using Eq. 14 were less than 0.4%.

DISCUSSION

Estimating linear dimensions of a 3D object using the volume tensor method may be more robust than direct measurement, depending on the subjective selection of extreme points on the object.

The Fakir probe efficiently estimates the volume tensor of a single object from 3D data using sparse systematic sampling without explicit segmentation. Predicting the precision of the method follows classical works (Hlawka, 1950; Matheron, 1965) but it had to be proved *de novo* using Wiener-Tauberian and geometric measure theory (Janáček and Jirák, 2019) in order to achive sufficient generality. In this study, we applied a special arrangement of line grids in a sevenfold grid (O´Keefe, 1992) to increase the precision of surface integral estimates (see Eq. 13) by negative covariance between estimates using sets of parallel lines in different directions.

The Fakir method is preferable to the surface-based method (Schröder-Turk *et al.*, 2011) in cases where automatic segmentation of an object is not possible. It can be used to efficiently measure the shapes of pheasant brain compartments and detect changes in shapes of the pheasant forebrain and hindbrain during development (Tables 3 & 4). The results may be of particular benefit for interpreting the fossilised braincases of extinct birds and dinosaurs (Beyrand, 2019).

The Fakir probe implemented with MS Visual C++, which includes semi-axis and precision calculations, can be downloaded from the author's website and used in morphometric studies of macro- or microscopic objects.

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REFERENCES

Barbier JE (1860). Note sur problème de l'aiguille et le jeu du joint couvert, Journal de Mathématiques Pures et Appliquès 5:273–87.

Beyrand V, Voeten DFAE, Bureš S, Fernandez V, Janáček J, Jirák D, Rauhut O, Tafforeau P (2019). Multiphase progenetic development shaped the brain of flying archosaurs. Sci Rep 9.

Cruz-Orive LM (1997). Stereology of single objects, Journal of Microscopy 186:93–107.

Dryden IL, Koloydenko A, Zhou D (2009). Non-Euclidean statistics for covariance matrices, with application to diffusion tensor imaging. Annal Appl Statist 3:1102–23.

Hlawka E (1950). Über integrale auf konvexen körpern I. Monatsh für Math 54 1-36.

Hug D, Schneider R, Schuster R (2008). Integral geometry of tensor valuations. Adv Appl Math 41:482–509.

Janáček J (1999). Errors of spatial grids estimators of volume and surface area. Acta Stereol 18:389–96.

Janáček J (2006). Variance of periodic measure of bounded set with random position. Comment Math Univ Carolinae 47:473–82.

Janáček J (2008). An asymptotics of variance of the lattice points count. Czechoslovak Mathematical Journal, 58 (133),751–75.

Janáček J, Kubínová L (2010). Variances of length and surface area estimates by spatial grids: preliminary study. Image Anal Stereol, 29:45–52.

Janáček J, Jirák D (2019). Variance of the isotropic uniform systematic sampling. submitted to IAS.

Jirák D, Janáček J, Kear BP (2015). A combined MR and CT study for precise quantitative analysis of the avian brain. Sci Rep 5.

Karten HJ, Brzozowska-Prechtl A, Lovell PV, Tang DD, Mello CV, Wang H, Mitra PP (2013). Digital atlas of the Zebra Finch (Taeniopygia guttata) brain: a high-resolution photo atlas. J Comp Neurology 521:3702–15.

O´Keefe M (1992). Cubic cylinder packings. Crystalography A48:879–84.

Kendall DG (1948). On the number of lattice points inside the random oval. Q J Math Oxford Ser 19 (3):1–26.

Kubínová L, Janáček J (1998). Estimating surface area by the isotropic fakir method from thick slices cut in an arbitrary direction. J Microsc 191 (2):201–11.

Kubínová L, Janáček J, Guilak F, Opatrný Z (1999). Comparison of several digital and stereological methods for estimating surface area and volume of cells studied by confocal microscopy. Cytometry 36:85–95.

Matheron G (1965). Les variables régionalisées et leur estimations. Masson et Cie, Éditeurs.

Rafati AH, Ziegel JF, Nyengaard JR, Jensen EBV (2016). Stereological estimation of particle shape and orientation from volume tensors. J Microsc 263:229–37.

Santaló LA (1976). Integral geometry and geometric probability. Addison–Wesley Publishing Company.

Schröder-Turk GE, Mickel W, Kapfer SC, Klatt MA, Schaller FM, Hoffmann MJF, Kleppmann N, Armstrong P, Inayat A, Hug D, Reichelsdorfer M, Peukert W, Schwieger W, Mecke K (2011). Minkowski tensor shape analysis of cellular, granular and porous structures. Adv Materials 23:2535–53.

Ziegel JF, Nyengaard JR, Jensen EBV (2015). Estimating particle shape and orientation using volume tensors. Scand J Statist 42:813–31.