Acta Applicandae Mathematicae **11** (1988), 199–211.

Integral Geometry and Three-Dimensional Reconstruction of Randomly Oriented Identical Particles from their Electron Microphotos

A. B. GONCHAROV
Cybernetics, Scientific Council of the Praesidium of the U.S.S.R. Academy of Sciences, 117333 Moscow, Vavilova 40, U.S.S.R.

(Received: 1 December 1985)

Abstract. A new method for the three-dimensional reconstruction of a structure from projections of randomly oriented particles on a plane is proposed. Reconstruction is performed in two steps. First, we find mutual orientation of particles, i.e., Euler angles, describing the angle of one particle with respect to another. Almost all the paper is devoted to solving this problem. Then we perform the three-dimensional reconstruction of an object from its projections in already-known directions.

The stability of the method with respect to experimental errors is shown. Three-dimensional reconstruction of asymmetric biological objects might be one of its applications.

AMS subject classification (1980). 44A15.

Key words. Integral geometry, Radon transform, electron microscopy, tomography.

1. Introduction

In 1917, Radon [1] considered the transformation assigning to a function $\rho(x)$ on the n-dimensional space, the function $\check{\rho}$ on the set of all hyperplanes in this n-dimensional space determined by integrals of ρ over hyperplanes

$$\check{\rho}(\omega, p) = \int \rho(x) \cdot \delta(\langle x \cdot \omega \rangle - p)\, dx, \quad |\omega| = 1.$$

Radon obtained an explicit inversion formula expressing ρ in terms of $\check{\rho}$. During the past 20 years, Radon's work has found wide application in Röntgen (X-ray), nuclear magnetic resonance, and ultrasonic tomography of the human body and other objects, electron microscopy of biomolecules, radioastronomy, and many other fields (see reviews [2] and [3]).

Radon's paper, together with the preceding paper by Minkowsky of 1915, started a new trend of mathematics: integral geometry. The main problem of integral geometry is to study an integral transformation assigning to a function ρ on a manifold M the function $\check{\rho}$ on a family of submanifolds $\check{M}$ of M defined by integrals of ρ over these submanifolds.

The new epoch in the development of integral geometry started after the works by Gelfand, Graev, and Shapiro [5, 6]. Until then, in problems of three-dimensional reconstruction, only the Radon transformation itself (on the plane and in the space) and Radon inversion formula had been used.

In this and subsequent papers, I propose a new approach to the three-dimensional recovering of the structure from the projections of randomly arranged particles on the plane. An essential role in this approach is played by Palay–Wiener's theorem for the Radon transformation proved by Gelfand and Graev in 1961, see [5, 7].

From the integral geometry viewpoint, one of the main results is the following: to recover (up to a motion of the whole space) a function $\rho(x)$ *with compact support* from its Radon transform $\check{\rho}(\omega, p)$ we do not need to know how $\check{\rho}(\omega, p)$ depends on ω. It only suffices to have a set of one-dimensional projections of $\check{\rho}(\omega, p)$ as functions of p without knowing in precisely what direction a projection is performed, i.e., without knowing ω. This result is considered in detail in [11] in the most difficult case $n = 2$.

Algorithms of three-dimensional reconstruction allow us to recover the particle's density distribution $\rho(x_1, x_2, x_3)$ from its planar projections in *known* directions usually determined by vectors on a sphere of radius 1 in the three-dimensional space. To determine the projection in direction τ is to determine integrals of $\rho(x_1, x_2, x_3)$ over the lines parallel to τ. For instance, to determine the projection in direction $(0, 0, 1)$ is to give the function $\rho(x_1, x_2) = \int \rho(x_1, x_2, x_3)\, dx_3$ (for details, see [3]).

There are problems, however, in which the orientation of τ_i is not known beforehand. A typical problem is that of the study of ribosomal particle structures in electron microscopy. These identical asymmetric particles are precipitated onto a layer so that the orientation on it is generally random. A photo of an electron microscope is the set of different projections p_{τ_i} of these particles, i.e., actually the set of projections of the same body whose orientation is unknown.

At a first glance, it seems that a projection of n arbitrarily oriented identical particles provides much less information on a distribution-function of a particle than n-projections of one particle in known directions. The main aim of this paper is to show that no information is actually lost, since we will show that the mutual arrangement of identical particles is recovered from their projection for $n \geqslant 3$.

Let us describe the setting in detail. First note that the centre of mass of a particle coincides with the projection of its centre. Therefore, we will assume that the projection of the centre of mass is known.

The arising problem of three-dimensional reconstruction is equivalent to the following (mathematical) problem: Given projections of particles randomly turned around their centre of mass on the plane x_1, x_2, recover the distribution function of particles.

More exactly, let the centre of mass be at the origin $(0, 0, 0)$. Let ω be an element of the rotation group of the three-dimensional space and $\rho_\omega(x_1, x_2, x_3)$ the distribution function of an electron density of a shifted (via ω) particle. How does one recover $\rho(x_1, x_2, x_3)$ from the functions

$$p_i(x_1, x_2) = \int \rho_{\omega_i}(x_1, x_2, x_3)\, \mathrm{d}x_3,$$

where $\omega_1, \ldots, \omega_n$ are unknown?

Kam [4] proposed an approximate method for solving this problem based mainly on the assumption of uniformity of the distribution of rotation ω_i over the rotation group of the three-dimensional space. The method of [4] fails completely for nonuniform distributions of rotations ω_i.

The main idea of our approach is to recover the mutual disposition of identical particles from their projections onto the plane $x_3 = 0$, denoted in what follows by α. It turns out that it can be done in a unique way.

Note immediately that since $\rho(x_1, x_2, x_3)$ is to be found up to a rotation, we may assume ω_1 to be the identity transformation. Then the rotations $\omega_2, \ldots, \omega_n$ are, in general, uniquely recovered from the projections. (If we do not assume ω_1 to be the identity, we may find $\omega_1^{-1}\omega_k$.)

After this we get the usual problem of three-dimensional reconstruction: recovering $\rho(x_1, x_2, x_3)$ from its projections in given directions.

We propose two different approaches to the problem of finding $\omega_2, \ldots, \omega_n$. One of them, a geometric one, is described in Section 2 and the other one in Section 3. The geometric approach is announced in [9]. It is inapplicable when the intersection of all the planes $\alpha, \omega_2^{-1}\alpha, \ldots, \omega_n^{-1}\alpha$ is a line. In [11], it is shown that for $n \geq 7$ the transformations ω_i can be recovered, nevertheless, with the help of the properties of the projection moments found by Gelfand and Graev [5, 7] (the Paley–Wiener theorem for Radon transformation). The results of [11] are announced in [10].

In this problem the moments of projections are used in the second approach to the problem of finding ω_i. This approach is based on the following construction. To $\rho(x_1, x_2, x_3)$ assign the positive definite quadratic form

$$Q_\rho(x_1, x_2, x_3) = \int \rho(y_1, y_2, y_3)(x_1y_1 + x_2y_2 + x_3y_3)^2\, \mathrm{d}y_1\, \mathrm{d}y_2\, \mathrm{d}y_3 \qquad (*)$$

Consider the ellipsoid $Q_\rho(x) = 1$. The ellipsoid obtained by rotating $Q_\rho(x)$ under ω corresponds to the function $\rho_\omega(x_1, x_2, x_3)$. The key idea of the second approach is to study these ellipsoids, since ω is uniquely determined by the image of a given three-axle ellipsoid. First, the form of the ellipsoid connected with $\rho(x_1, x_2, x_3)$ is determined, i.e., the lengths of its principal axes (for an asymmetric particle these lengths are different). Further, from the projection we find the sections of the ellipsoid $Q_{\rho_\omega}(x) = 1$ by the plane $x_3 = 0$ (see formula (*)). It remains for us to make use of the fact that there exists exactly four ways to arrange the three-axle ellipsoid of the known form with the centre at $(0, 0, 0)$ so that the section by the plane $x_3 = 0$ is of the given form. After this, simple additional arguments allow us to find the actual arrangement of the ellipsoid.

The present work is oriented toward the electron-microscopic study of biologic particles. Nevertheless, the possibility of applying this approach to other tomographic problems should be investigated more attentively, especially those problems where the object may randomly change its position in the space.

In the Crystallography Institute of the U.S.S.R. Academy of Sciences, a computational experiment with a model of ribosom has been performed which confirmed the possibility of a practical application of the geometric approach suggested here (1985). The detailed description of the experiment and its results are to be found in [12].

2. Geometric Method. Recovering the Mutual Orientation of Particles from the Projections

2.1. HOW TO RECOVER THE SYSTEM OF PLANES $\omega_i^{-1}\pi$

Let $n_{i_1 i_2}$ be the straight line, the intersection of $\omega_i^{-1}\pi$ and $\omega_{i_2}^{-1}\pi$. Represent these lines as $n_{i_1 i_2} = \pi \cap \omega_{i_1}\omega_{i_2}^{-1}\pi$.

Recall that by the projection of a function defined on the plane onto the line $n_{i_1 i_2}$, we mean its integrals over the system of lines perpendicular to this line. Then the projection of $p_{i_1}(x_1, x_2)$ onto $n_{i_1 i_2}$ coincides with the projection of $p_{i_2}(x_1, x_2)$ onto $n_{i_2 i_1}$. In fact, the projections coincide with the projections of $\rho_{\omega_{i_1}}(x_1, x_2, x_3)$ or $\rho_{\omega_{i_2}}(x_1, x_2, x_3)$ onto this line. It remains for us to note that the projection of $\rho_{\omega_{i_1}}(x_1, x_2, x_3)$ onto $n_{i_1 i_2}$, as well as the projection of $\rho_{\omega_{i_2}}(x_1, x_2, x_3)$ onto $n_{i_2 i_1}$, coincides with the projection of $\rho(x_1, x_2, x_3)$ onto $n_{i_1 i_2}$.

For an asymmetric particle on the plane π there exists, in general, no other pair of lines $\tilde{n}_{i_1 i_2}$, $\tilde{n}_{i_2 i_1}$ such that the projection of $p_{i_1}(x_1, x_2)$ onto $n_{i_1 i_2}$ coincides with the projection of $p_{i_2}(x_1, x_2)$ onto $n_{i_2 i_1}$.

Therefore, on π, we should take an ample set of lines through $(0, 0)$ and consider the projections $f_1^i(t), \ldots, f_N^i(t)$ of a function $p_i(x_1, x_2)$ known from an experiment onto these lines. For each pair of functions $p_{i_1}(x_1, x_2)$ and $p_{i_2}(x_1, x_2)$, compare the two sets $\{f_j^{i_1}(t)\}$ and $\{f_j^{i_2}(t)\}$ considering $f_j^{i_1}(t)$ and $f_j^{i_2}(t)$ *almost coincidental* if $\int |f_j^{i_1}(t) - f_j^{i_2}(t)|^2 \, dt$ is sufficiently small (ideally, equal to 0).

2.2. THE STABILITY OF THE METHOD WITH RESPECT TO EXPERIMENTAL ERRORS

The main point of our algorithm is to find the genuine pair of lines $n_{i_1 i_2}(i_1)$ and $n_{i_1 i_2}(i_2)$ on α. We may encounter several suspicious pairs.

In what follows, we explain why this does not lead to ambiguity in recovering the mutual configuration of the planes $\omega_i^{-1}\alpha$ and how to remove the false pairs of lines.

The configuration of the planes $\omega_i^{-1}\alpha$, $i = 2, \ldots, n$, is determined by $2(n-1)$ parameters, since the position of one plane is determined by a unique normal to it, i.e., by two parameters. We know all the angles $\varphi_{j_1 j_2}^i$, between n_{ij_1} and n_{ij_2}. For

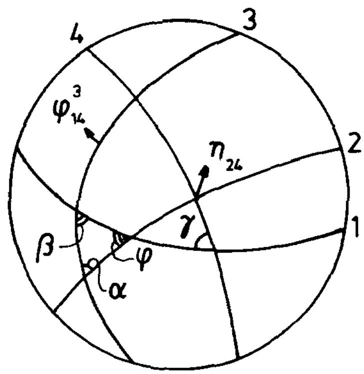

Fig. 1.

a fixed i, there are $n-2$ independent angles among them, since the planes split the plane $\omega_j^{-1}\alpha$ into $n-1$ sectors, but the sum of all the angles equals 2π. This implies that among the angles $\varphi^i_{j_1 j_2}$ there are $n(n-2)-2(n-1)=n^2-4n+2$ independent identities. To get one of them, consider an arbitrary quadruple of planes $\omega_{j_1}^{-1}\alpha$, $\omega_{j_2}^{-1}\alpha$, $\omega_{j_3}^{-1}\alpha$, $\omega_{j_4}^{-1}\alpha$ (Figure 1) and their intersections with the unit sphere. The lengths of the arch-segments obtained on the sphere equal the angles $\varphi^i_{j_1 j_2}$. Then (see Figure 1)

$$\frac{\sin \check{AK} \cdot \sin \check{PC} \cdot \sin \check{BL}}{\sin \check{KB} \cdot \sin \check{AP} \cdot \sin \check{CL}} = 1.$$

For an asymmetric particle, it is natural to assume that the function defined on the line $n_{i_1 i_2}$ (namely the projection of $p_{i_1}(x_1, x_2)$ onto this line) is not even. It will be convenient to imagine that each projection of $p_i(x_1, x_2)$ is defined on its own plane P_i, though these planes are naturally identified with the plane $Z=0$.

The above makes it clear that there is exactly one way to identify the line $n_{i_1 i_2}$ on the plane p_{i_1} with the line $n_{i_2 i_1}$ on the pane p_{i_2} so as to make the functions defined on them coincidental.

LEMMA. *The arrangement of three (or more) planes (not passing through one line) is uniquely defined up to a motion or a reflection of this space if*

(1) *on each plane there are given the lines of its intersection with the other planes;*

(2) *there is given a way to identify the corresponding lines on different planes (such as $n_{i_1 i_2} \subset p_{i_1}$ and $n_{i_2 i_1} \subset p_{i_2}$).*

Thanks to the lemma, we may find the planes $\pi, \omega_2^{-1}\pi, \ldots, \omega_i^{-1}\pi$ up to the reflection with respect to π. A rotation of the three-dimensional space is not yet defined by the image of a fixed plane π, since we may rotate it afterwards around the axis perpendicular to the image of π.

However, we have additional information: the rotation ω_i^{-1} transforms π and the line n_{1i} on it into $\omega_i^{-1}\pi$ with the given line on it, so that the functions on these lines are identified. This determines the transformation ω_i^{-1} uniquely.

In practice, to construct the planes $\pi, \omega_i^{-1}\pi, \omega_j^{-1}\pi$ we are to recover the trihedral angle from the planar angles between the pairs of lines n_{1i}, n_{1j}; n_{i1}, n_{ij}; n_{j1}, n_{ji}. The angle between a pair of these lines is well-defined if, on each line, a direction is determined so that, these directions coincide with respect to a given way of identifying the lines. (The angle between nonordered lines is ambiguously determined: it is either φ or $\pi - \varphi$.) It is easy to verify that the planes of the faces of the obtained trihedral angle do not depend on the arbitrariness in the choice of consistent directions of the lines. For example, if we reverse the direction of the lines n_{1i} and n_{i1}, then two of the three angles will be replaced by the complementary ones and we get another trihedral angle, namely the adjoint one. The planes of the faces of this trihedral angle are the same.

3. Finding Mutual Orientation of Particles by the Moment Method

Let e_1, e_2, e_3 be an orthonormal basis in the space with coordinates x_1, x_2, x_3. Let ω_{ij} be the matrix of an orthonormal transformation ω in this basis, i.e.,

$$\omega \cdot e_i = \sum_{1 \leq j \leq 3} \omega_{ij} e_j .$$

Then

$$\rho_\omega(x_1, x_2, x_3) = \rho(\omega_{ij} x_j).$$

We have chosen the centre of mass of the particle as an origin. Therefore

$$\int \rho(x_1, x_2, x_3) x_k \,\mathrm{d}x_1 \,\mathrm{d}x_2 \,\mathrm{d}x_3 = 0, \quad k = 1, 2, 3.$$

Consider the set of the second moments of a function f:

$$\lambda_{k_1, k_2}(f) = \int f(x_1, x_2, x_3) x_{k_1} x_{k_2} \,\mathrm{d}x_1 \,\mathrm{d}x_2 \,\mathrm{d}x_3, \quad 1 \leq k_1, k_2 \leq 3.$$

Then

$$\lambda_{k_1 k_2}(\rho_\omega) = \sum_{1 \leq l_1, l_2 \leq 3} \omega_{l_1 k_1} \cdot \omega_{l_2 k_2} \lambda_{l_1 l_2}(\rho). \tag{1}$$

In fact, performing the change of variables $y_i = \sum_{1 \leq j \leq 3} \omega_{ij} x_j$ (therefore, $x_k = \sum_{1 \leq l \leq 3} \omega_{lk} y_l$ since for an orthogonal matrix ω, $(\omega^{-1})_{ij} = \omega_{ji}$) we get

$$\lambda_{k_1k_2}(\rho_\omega) = \int \rho(\omega_{ij}x_j)x_{k_1}x_{k_2}\,dx_1\,dx_2\,dx_3$$
$$= \int \rho(y_1, y_2, y_3)\Big(\sum_{1\le l_1\le 3} \omega_{l_1k_1}y_{l_1}\Big)\cdot\Big(\sum_{1\le l_2\le 3} \omega_{l_2k_2}y_{l_2}\Big)\,dy_1\,dy_2\,dy_3$$
$$= \sum_{1\le l_1,l_2\le 3} \omega_{l_1k_1}\cdot\omega_{l_2k_2}\lambda_{l_1l_2}(\rho).$$

Set

$$\Lambda(\rho_\omega) = \begin{pmatrix} \lambda_{11}(\rho) & \lambda_{12}(\rho) & \lambda_{13}(\rho) \\ \lambda_{12}(\rho) & \lambda_{22}(\rho) & \lambda_{23}(\rho) \\ \lambda_{13}(\rho) & \lambda_{23}(\rho) & \lambda_{33}(\rho) \end{pmatrix}.$$

Let $W = (\omega_{ij})$. Formula (1) can be rewritten in the form

$$\Lambda(\rho_\omega) = W^t\cdot\Lambda(\rho)\cdot W = W^{-1}\cdot\Lambda(\rho)\cdot W.$$

Therefore,

$$I_1(\rho_\omega) = \operatorname{tr}\Lambda(\rho_\omega), \qquad I_2(\rho_\omega) = \operatorname{tr}\Lambda^2(\rho_\omega), \qquad I_3(\rho_\omega) = \det\Lambda(\rho_\omega) \tag{2}$$

do not depend on ω, though it occurs in their definition.

To each vector $\boldsymbol{\lambda}_2(\rho)$ of Λ assign the quadratic form

$$Q_{\boldsymbol{\lambda}_2(\rho)}(x)7 = \sum_{1\le i,j\le 3} \lambda_{ij}x_ix_j$$
$$= \sum_{1\le i,j\le 3} x_ix_j\cdot\int \rho(y_1, y_2, y_3)y_iy_j\,dy_1\,dy_2\,dy_3$$
$$= \int \rho(y_1, y_2, y_3)\cdot(y_1x_1 + y_2x_2 + y_3x_3)^2\,dy_1\,dy_2\,dy_3. \tag{3}$$

Note that the distribution function of the electron density of the particle is nonnegative. Hence, $Q_{\boldsymbol{\lambda}_2(\rho)}(x) > 0$ for $x \neq 0$.

Consider the ellipsoid $Q_{\boldsymbol{\lambda}_2(\rho)}(x) = 1$. The lengths of its principal axes are $1/\sqrt{s_i(\rho)}$, where $s_i(\rho)$ are the eigenvalues of $\Lambda(\rho)$. Formula (3) implies that under ω, this ellipsoid turns into an ellipsoid corresponding to $\boldsymbol{\lambda}_2(\rho_\omega)$. The main idea of the method proposed below is to study the ellipsoid corresponding to $\rho_\omega(x_1, x_2, x_3)$. If the lengths of the principal axes of the ellipsoid corresponding to $\rho(x_1, x_2, x_3)$ are different, then a nonidentity transformation ω transforms it into another, different ellipsoid.

It is natural to assume that the lengths of the principal axes are different for an asymmetric particle. Therefore, in this case $\boldsymbol{\lambda}_2(\rho_\omega) = \boldsymbol{\lambda}_2(\rho)$ implies that ω is the identity transformation. Moreover, if we know $\boldsymbol{\lambda}_2(\rho)$, then ω is uniquely determined by $\boldsymbol{\lambda}_2(\rho_\omega)$. Therefore, we should find all the vectors.

Note that the components $\lambda_{11}(\rho_\omega)$, $\lambda_{12}(\rho_\omega)$, $\lambda_{22}(\rho_\omega)$ are calculated from the projection of $p_\omega(x_1, x_2)$ since

$$\lambda_{i_1 i_2} = \int \rho_\omega(x_1, x_2, x_3) x_{i_1} x_{i_2} \, dx_1 \, dx_2 \, dx_3$$
$$= \int \rho_\omega(x_1, x_2) x_{i_1} x_{i_2} \, dx_1 \, dx_2, \quad \text{for } 1 \leqslant i_1, i_2 \leqslant 2.$$

Geometrically, this means that we know the section of the ellipsoid corresponding to $\boldsymbol{\lambda}_2(\rho_\omega)$ by the plane $x_3 = 0$. Let us find the remaining components of $\boldsymbol{\lambda}_2(\rho_\omega)$.

First let us calculate $I_1(\rho)$, $I_2(\rho)$, $I_3(\rho)$. For this, let us make use of the geometric method expressed above to find two transformations, say, ω_2 and ω_3 different from the rotation around Ox_3. Namely, take two projections for which this algorithm is best applicable.

After the transformations ω_2 and ω_3 are found, we solve the system of linear equations with three unknowns $\lambda_{13}(\rho)$, $\lambda_{23}(\rho)$, $\lambda_{33}(\rho)$:

$$\sum_{1 \leqslant l_1, l_2 \leqslant 3} \omega_{l_1 k_1}^{(s)} \cdot \omega_{l_2 k_2}^{(s)} \cdot \lambda_{l_1 l_2}(\rho) = \lambda_{k_1 k_2}(\rho_{\omega_s}), \quad 1 \leqslant k_1 \leqslant k_2 \leqslant 2, \quad s = 2, 3,$$

where $\omega_{ij}^{(a)}$ is the matrix of the transformation ω_a. Since all the parameters are only known approximately, we may consider a similar system for ω_3 and find the minimum of the function of three variables $\lambda_{i3}(\rho)$ $(1 \leqslant i \leqslant 3)$:

$$\sum_{2 \leqslant s \leqslant 3} \sum_{1 \leqslant k_1, k_2 \leqslant 2} \left(\sum_{1 \leqslant l_1, l_2 \leqslant 3} \omega_{l_1 k_1}^{(s)} \cdot \omega_{l_2 k_2}^{(s)} \cdot \lambda_{l_1 l_2}(\rho) - \lambda_{k_1 k_2}(\rho_{\omega_s}) \right)^2.$$

Substituting the obtained values $\lambda_{ij}(\rho)$ into the formulas

$$\begin{aligned} I_1(\rho) &= \lambda_{11}(\rho) + \lambda_{22}(\rho) + \lambda_{33}(\rho), \\ I_2(\rho) &= \lambda_{11}^2(\rho) + \lambda_{22}^2(\rho) + \lambda_{33}^2(\rho) + 2\lambda_{12}^2(\rho) + 2\lambda_{13}^2(\rho) + 2\lambda_{23}^2(\rho), \\ I_3(\rho) &= \lambda_{11}(\rho) \cdot \lambda_{22}(\rho) \cdot \lambda_{33}(\rho) + 2\lambda_{12}(\rho) \cdot \lambda_{13}(\rho) \cdot \lambda_{23}(\rho) - \\ &\qquad - \lambda_{13}^2(\rho) \cdot \lambda_{33}(\rho) - \lambda_{23}^2(\rho) \cdot \lambda_{33}(\rho) - \lambda_{13}^2(\rho) \cdot \lambda_{22}(\rho), \end{aligned} \tag{4}$$

we get $I_1(\rho)$, $I_2(\rho)$, $I_3(\rho)$. Therefore,

$$\lambda_{33}(\rho_\omega) = I_1(\rho) - \lambda_{11}(\rho_\omega) - \lambda_{22}(\rho_\omega).$$

To define $\lambda_{23}(\rho_\omega)$ and $\lambda_{13}(\rho_\omega)$ we have the system of quadratic equations

$$\begin{aligned} &\lambda_{13}^2(\rho_\omega) + \lambda_{23}^2(\rho_\omega) \\ &\quad = \tfrac{1}{2}(I_2(\rho) - \lambda_{11}^2(\rho_\omega) - \lambda_{22}^2(\rho_\omega) - \\ &\qquad - (I_1(\rho) - \lambda_{11}(\rho) - \lambda_{22}(\rho))^2 - 2\lambda_{12}^2(\rho_\omega)), \\ &\lambda_{13}^2(\rho_\omega)(I_1(\rho) - \lambda_{11}(\rho_\omega)) + \lambda_{23}(\rho_\omega) \cdot \lambda_{11}(\rho_\omega) - \\ &\qquad - 2\lambda_{13}(\rho_\omega) \cdot \lambda_{23}(\rho_\omega) \cdot \lambda_{12}(\rho_\omega) \\ &\quad = \lambda_{11}(\rho_\omega) \cdot \lambda_{22}(\rho_\omega)(I_1(\rho) - \lambda_{11}(\rho) - \lambda_{22}(\rho)) - I_3(\rho), \end{aligned} \tag{5}$$

which has, in general, four solutions. Note that the presence of exactly four solutions (a priori, we know that there exists at least one solution corresponding to the genuine position of the particle) is easy to see in the geometric interpretation via the ellipsoid.

In fact, we know

$$I_1(\rho) = s_1(\rho) + s_2(\rho) + s_3(\rho), \qquad I_2(\rho) = s_1^2(\rho) + s_2^2(\rho) + s_3^2(\rho),$$
$$I_3(\rho) = s_1(\rho) \cdot s_2(\rho) \cdot s_3(\rho)$$

and therefore

$$s_1(\rho) \cdot s_2(\rho) + s_1(\rho) \cdot s_3(\rho) + s_2(\rho) \cdot s_3(\rho) = \tfrac{1}{2}(I_1^2(\rho) - I_2(\rho))$$

hence, we may find $s_1(\rho)$, $s_2(\rho)$, $s_3(\rho)$, which are the roots of the cubic equation

$$z^3 - I_1(\rho)z^2 + \tfrac{1}{2}(I_1^2(\rho) - I_2(\rho)) \cdot z - I_3(\rho) = 0.$$

Therefore the ellipsoids corresponding to different solutions $\boldsymbol{\lambda}_2$ are of the same form. Hence, if the numbers $s_i(\rho)$ are different, i.e., $\boldsymbol{\lambda}_2$ corresponds to the three-axle ellipsoid (in particular, this is so for an asymmetric particle), all the ellipsoids corresponding to the solutions $\boldsymbol{\lambda}_2$ are obtained from someone by reflections with respect to the planes through the main axes of the ellipse cut on the plane $x_3 = 0$ by the ellipsoid and perpendicular to this plane.

Recall that (for a three-axle ellipsoid) for each obtained $\boldsymbol{\lambda}_2$ there exists a unique ω, such that $\boldsymbol{\lambda}_2 = \boldsymbol{\lambda}_2(\rho_\omega)$. To find this ω we should find the eigenvectors and eigenvalues of the symmetric matrix Λ recovered from $\boldsymbol{\lambda}_2$ and $\Lambda(\rho)$.

Let $f_1(\omega)$, $f_2(\omega)$, $f_3(\omega)$ and f_1, f_2, f_3 be the eigenvectors of Λ and $\Lambda(\rho)$, respectively, of length 1 such that to the eigenvectors $f_i(\omega)$ and f_i the same eigenvalues correspond. Let us decompose $f_i(\omega)$ with respect to f_j:

$$f_i(\omega) = \sum_{1 \leq j \leq 3} \tilde{\omega}_{ij} f_j .$$

The numbers $\tilde{\omega}_{ij}$ are the matrix elements of the desired transformation ω (with respect to f_1, f_2, f_3). The matrix of ω in the basis e_1, e_2, e_3 are obtained by the formula

$$\omega_{ij} = \sum_{1 \leq s,k \leq 3} a_{is} \cdot \omega_{sk} \cdot \omega_{jk} , \quad \text{where } e_i = \sum_{1 \leq j \leq 3} a_{ij} f_j . \tag{6}$$

To find the genuine value of ω among the four possible ones, make use of the algorithm based on the rotation of one projection around another. For each of the possible values ω^α, find the line on the plane $x_3 = 0$ spanned by the unit vector $\cos \xi_\alpha \cdot e_1 + \sin \xi_2 \cdot e_2$, such that the line $\omega^\alpha \cdot l_\alpha$ belongs to the same plane. This means that $\omega_{13}^\alpha \cos \xi_\alpha + \omega_{23}^\alpha \sin \xi_\alpha = 0$, i.e., $\tan \xi_\alpha = -(\omega_{13}^\alpha / \omega_{23}^\alpha)$. Here ω_{ij}^α are the matrix elements of ω^α. We have shown that if $\omega^\alpha = \omega$, then the projection of $p_\omega(x_1, x_2)$ onto $\omega^\alpha \cdot l_\alpha$ coincides with the projection of $p(x_1, x_2)$ onto l_α. Finding, thanks to formula (6), the lines l_α and $\omega^\alpha \cdot l_\alpha$ and calculating the corresponding projections, we find the genuine transformation.

If $\omega_{13}^3 = \omega_{23}^3$, then ω^α is the rotation by the angle ξ_α around Ox_3. Therefore, after the rotation of $p_\omega(x_1, x_2)$ around $(0, 0)$ we should get $p_{\omega_1}(x_1, x_2)$.

The advantage of the above approach is that the algorithm which makes use of the rotation of one projection around another is needed in full only to find rotations ω_2 and ω_3. All the remaining operations are considerably simpler.

To get completely rid of the rotation of one projection with respect to another, we introduce the third moments

$$\lambda_{i_1 i_2 i_3}(\rho) = \int \rho(x_1, x_2, x_3) x_{i_1} x_{i_2} x_{i_3}\, dx_1\, dx_2\, dx_3, \quad 1 \leqslant i_1, i_2, i_3 \leqslant 3.$$

Clearly, $\lambda_{i_1 i_2 i_3}(\rho)$ do not depend on the permutation of indices. The components of $\lambda_{i_1 i_2 i_3}(\rho)$, for $1 \leqslant i_1, i_2, i_3 \leqslant 2$, are calculated from the projection. There are four independent components among them, whereas the total number of (independent) components is 10. Therefore, we have to find six components, namely

$$\lambda_{113}(\rho), \lambda_{123}(\rho), \lambda_{133}(\rho), \lambda_{223}(\rho), \lambda_{233}(\rho), \lambda_{333}(\rho).$$

A formula similar to (5) holds:

$$\lambda_{k_1 k_2 k_3}(\rho) = \sum_{1 \leqslant l_1, l_2, l_3 \leqslant 3} \omega_{l_1 k_1} \cdot \omega_{l_2 k_2} \cdot \omega_{l_3 k_3} \cdot \lambda_{l_1 l_2 l_3}(\rho).$$

Consider a system of 21 equations:

$$\sum_{1 \leqslant l_1, l_2 \leqslant 3} \omega_{l_1 k_1}^{(s)} \cdot \omega_{l_2 k_2}^{(s)} \lambda_{l_1 l_2}(\rho) = \lambda_{k_1 k_2}(\rho_{\omega_s}), \quad 1 \leqslant k_1 \leqslant k_2 \leqslant 2,$$

$$\sum_{1 \leqslant l, l_2, l_3 \leqslant 3} \omega_{l_1 k_1}^{(s)} \cdot \omega_{l_2 k_2}^{(s)} \lambda_{l_1 l_2 l_3}(\rho) = \lambda_{k_1 k_2 k_3}(\rho_{\omega_s}), \quad 1 \leqslant k_1 \leqslant k_2 \leqslant k_3 \leqslant 2,$$

$$s = 2, 3, 4,$$

for 18 unknowns $\varphi_1^{(s)}, \varphi_2^{(s)}, \theta^{(s)}$ (the Euler angles of transformations $\omega^{(s)}$); $\lambda_{13}(\rho)$, $\lambda_{23}(\rho)$, $\lambda_{33}(\rho)$; $\lambda_{113}(\rho)$, $\lambda_{123}(\rho)$, $\lambda_{133}(\rho)$, $\lambda_{223}(\rho)$, $\lambda_{233}(\rho)$, $\lambda_{333}(\rho)$.

Recall, (see [8]) that the matrix of ω in the basis e_1, e_2, e_3 is expressed via Euler angles $\varphi_1, \varphi_2, \theta$ by the formula

$$\left\| \begin{matrix} \cos\varphi_1 \cdot \cos\varphi_2 - \cos\theta \cdot \sin\varphi_1 \cdot \sin\varphi_2 & \sin\varphi_1 \cdot \cos\varphi_2 + \cos\theta \cdot \cos\varphi_1 \cdot \sin\varphi_2 & \sin\varphi_2 \cdot \sin\theta \\ -\cos\varphi_1 \cdot \sin\varphi_2 - \cos\theta \cdot \sin\varphi_1 \cdot \cos\varphi_2 & -\sin\varphi_1 \cdot \sin\varphi_2 + \cos\theta \cdot \cos\varphi_1 \cdot \cos\varphi_2 & \cos\varphi_2 \cdot \sin\theta \\ \sin\varphi_1 \cdot \sin\theta & -\cos\varphi_1 \cdot \sin\theta, & \cos\theta \end{matrix} \right\|$$

where $0 \leqslant \varphi_1, \varphi_2 \leqslant 2\pi$, $0 \leqslant \theta \leqslant \pi$.

This system is overdefined and has a unique solution which can be found with a computer by the least-squares method, i.e., computing the minimum of the function in 18 variables

$$\sum_{2\leqslant s\leqslant 4}\ \sum_{1\leqslant k_1,k_2\leqslant 2}\left(\sum_{1\leqslant l_1,l_2\leqslant 3}\omega_{l_1k_1}^{(s)}\cdot\omega_{l_2k_2}^{(s)}\cdot\lambda_{l_1l_2}(\rho)-\lambda_{k_1k_2}(\rho_{\omega_s})\right)^2+$$

$$+\sum_{1\leqslant k_1\leqslant k_2\leqslant k_3}\ \sum_{1\leqslant l_1,l_2,l_3\leqslant 3}(\omega_{l_1k_1}^{(s)}\cdot\omega_{l_2k_2}^{(s)}\cdot\omega_{l_3k_3}^{(s)}\cdot\lambda_{l_1l_2l_3}(\rho)-\lambda_{k_1k_2k_3}(\rho_{\omega_s}))^2).$$

Further, by the above method we find four possible values ω_j^{α} ($\alpha=1,2,3,4$) for each ω_j, $5\leqslant j\leqslant n$.

Note that the matrix elements $\omega_{i_1i_2}^{(j)}$ of the transformation ω_j satisfy the system

$$\sum_{1\leqslant l_1,l_2,l_3\leqslant 3}\omega_{l_1k_1}^{(j)}\cdot\omega_{l_2k_2}^{(j)}\cdot\omega_{l_3k_3}^{(j)}\lambda_{l_1l_2l_3}(\rho)=\lambda_{k_1k_2k_3}(\rho_{\omega_j}),$$

where $1\leqslant k_1\leqslant k_2\leqslant k_3\leqslant 2$. Therefore, substituting the matrix elements of transformations into this system, we find the genuine values of ω_j.

In the real situation, the values of $p_{\omega_i}(x_1, x_2)$ are known only approximately and do not always equal $\int \rho_{\omega_i}(x_1, x_2, x_3)\,dx_3$. To choose the more appropriate of the above methods, one should understand their advantages and disadvantages.

If the projection $p_{\omega_i}(x_1, x_2)$ becomes too 'dizzy' the farther it goes from the centre of mass (i.e., the degree of reliability with which the function $p_{\omega_i}(x_1, x_2)$ is known diminishes as the distance of the centre of mass grows), then it is advisable to make less use of the moments $\lambda_{i_1i_2i_3}(\rho)$, since a third degree polynomial grows much faster than a second degree polynomial and, therefore, the error of calculating $\lambda_{i_1i_2i_3}(\rho)$ is considerably greater than that for $\lambda_{i_1i_2}(\rho)$.

On the other hand, if $p_{\omega_i}(x_1, x_2)$ are uniformly unprecise, then we had better deal with the integral characteristics of the function $p_{\omega_i}(x_1, x_2)$ (like the second and third moments) instead of the values of $p_{\omega_i}(x_1, x2)$ at certain lines.

4. Finding the Mutual Orientation of Particles when the Electron Microscope is Supplied with a Goniometer

If our electron microscope is supplied with a goniometer, then we may turn the film with particles on it by known angles φ_1 and φ_2 around the axis Ox_1 (clockwise) (such a possibility is important in defining a rhibosom structure). In this case, the transformations ω_i are very easy to find. Making use of the formula (5), we get the system of four linear equations

$$\lambda_{i2}(\rho_\omega)\cdot\cos\varphi_s+\lambda_{i3}(\rho\omega)\cdot\sin\varphi_s=\lambda_{i2}(\rho_\omega^{\varphi_s}),\quad \text{where } i=1,2;\quad s=1,2,$$

solving which we find $\lambda_{13}(\rho_\omega)$, $\lambda_{23}(\rho_\omega)$, $\lambda_{33}(\rho_\omega)$.

By ρ_ω^{φ} we have denoted the distribution function of the electron density of the particle obtained from the initial particle after the rotation ω with the subsequent rotation by the angle φ clockwise around the Ox_1 axis. We may now find the values $\lambda_{13}(\rho_\omega^{\varphi_s})$, $\lambda_{23}(\rho_\omega^{\varphi_s})$, $\lambda_{33}(\rho_\omega^{\varphi_s})$ by the formula ($s=1,2$):

$$\lambda_{13}(\rho_\omega^{\varphi_s}) = -\lambda_{12}(\rho_\omega)\cdot\sin\varphi_s + \lambda_{13}(\rho_\omega)\cos\varphi_s,$$

$$\lambda_{23}(\rho_\omega^{\varphi_s}) = (\lambda_{33}(\rho_\omega) - \lambda_{22}(\rho_\omega))\cos\varphi_s\cdot\sin\varphi_s + \\ + \lambda_{23}(\rho_\omega)(\cos^2\varphi_s - \sin^2\varphi_s),$$

$$\lambda_{33}(\rho_\omega^{\varphi_s}) = \lambda_{33}(\rho_\omega)\cos^2\varphi_s - \lambda_{23}(\rho_\omega)\cos\varphi_s\cdot\sin\varphi_s + \lambda_{22}(\rho_\omega)\sin^2\varphi_s.$$

5. Several Remarks on Finding the Mutual Orientation of Identical Particles with Nontrivial Symmetry Group G

All the subgroups in the group of rotations of the three-dimensional space are divided into three classes. We should operate differently in each of the classes:

(a) The symmetry group G is such that there exists a three-axle ellipsoid invariant with respect to G. Then, in principle, we might proceed as earlier.

(b) Any ellipsoid with the group G is an ellipsoid of rotation. In this case, a typical G-invariant ellipsoid is different from the sphere and, therefore, has a distinguished axis. It is natural to assume that, in particular, such is the ellipsoid corresponding to the distribution function of the particle under investigation. Therefore, we should look at the distinguished axis of the ellipsoid. As a result, the transformation ω is found up to rotation around some axis (namely, around the distinguished axis of the ellipsoid corresponding to $\rho_\omega(x_1, x_2, x_3)$. To find the precise value of ω, we may apply the algorithm based on rotating one projection around another. Note that this is easier to apply in this situation than in general case, since for each angle φ (rotation of one projection around the other one) it is easy to analytically find the two lines $l_1(\varphi)$ and $l_2(\varphi)$ in the plane $x_3 = 0$, so that the restriction of the function $p_\omega(x_1, x_2)$ onto $l_1(\varphi)$ is the same as that of $p_{\omega_1}(x_1, x_2)$ onto $l_2(\varphi)$. Therefore, in the general situation, we should compare the restriction of the corresponding functions onto all the possible pairs of lines (depending on two parameters), whereas in our situation we are to investigate the restrictions of the functions onto a one-parameter family of pairs of lines which is far more simpler, e.g., of the set of 50 lines we should compare 2500 pairs of functions in the general case and only 50 in our case.

An alternative way is to make use of the third moments $\lambda_{i_1 i_2 i_3}(\rho)$.

(c) G is such that any G-invariant ellipsoid is a sphere. An equivalent condition is that there are no nonzero G-invariant vectors in three-dimensional space. Then, in principle, we may similarly make use of higher moments. However, G is actually great enough to perform a three-dimensional reconstruction from several photos (see the Introduction).

Therefore, our method works better when the considered particles are less symmetric. Therefore, it is a nice compliment to the conventional methods of three-dimensional reconstruction of complicated biologic objects that work well when the object possesses a sufficiently large symmetry group.

References

1. Radon, J.: Über die Bestimmung von Functionen durch ihre Integralwarte längs gewisser Mannigfaltigkeiten. *Ber. Verh. Sächs. Acad.* **69** (1917), 262–277.
2. Pikalov, V. V. and Preobrazhensky, N. G.: Computational tomography and physical experiment, *Russian Phys. Surveys*, 1983, **141**, No. 3 (1983), 469–498 (in Russian).
3. Wainstein, B. K.: Three-dimensional electron microscopy of biology micromolecules, *Russian Phys. Surveys*, 1973, **109**, No. 3 (1973) 455–497 (in Russian).
4. Kam, Z.: Three-dimensional reconstruction of aperiodic objects, *J. Theoret. Biol.* **82** (1980), 15–32.
5. Gelfand, I. M., Graev, M. J., and Vilenkin, N. Ya.: *Integral Geometry and Related Problems of Representation Theory.*, Fizmatgiz, Moscow, 1962 (in Russian).
6. Gelfand, I. M., Graev, M. J., and Shapiro, Z. Ya.: Integral geometry on k-dimensional planes, *Funct. Anal. Appl.* No. 1 (1967), 15–31 (in Russian).
7. Gelfand, I. M., Gindikin, S. G., and Graev, M. J.: *Integral Geometry in Affine and Projective Spaces. Results of Science and Technics* (Modern Problems of Mathematics, v. 16), VINITI, Moscow, 1980 (in Russian).
8. Gelfand, I. M., Minlos, R. A., and Shapiro, Z. Ya.: *Representations of the Rotation Group and the Loretz Group*, Fizmatgiz, Moscow, 1958 (in Russian).
9. Wainstein, B. K. and Goncharov, A. B.: Recording of the spatial orientation of randomly arranged identical particles of unknown structure from their projections, *Doklady USSR Acad. Sci.* (1986) (in Russian).
10. Goncharov, A. B.: Methods of integral geometry and finding the mutual orientation of randomly arranged particles on the plane from their projections on the line, *Doklady USSR Acad. Sci.* (1986) (in Russian).
11. Goncharov, A. B.: Methods of integral geometry and recovering a function with compact support from its projections in unknown directions. *Acta Applic. Math.* **11** (1988), 213–222.
12. Goncharov, A. B., Wainstein, B. K., Ryskin, A. I., Vagin, A. A.: preprint.