VISUALIZATION AND EXTRACTION OF STRUCTURAL COMPONENTS FROM RECONSTRUCTED VOLUMES

Edgar Garduno

A DISSERTATION

in

BIOENGINEERING

Presented to the Faculties of the University of Pennsylvania in Partial Fulfillment of the Requirements for the Degree of Doctor of Philosophy

2002

Gabor T. Herman, Ph.D.

Supervisor of Dissertation

Gershon Buchsbaum, Ph.D.

Graduate Group Chairperson

DEDICATION

Para Oliva, Leopoldo y Gustavo quienes siempre han estado …

To Oliva, Leopoldo y Gustavo who have always been present…

Por todos los ladrones y ángeles que pasarón, y los que pasarán …

Con todo el corazón y cariño.

ACKNOWLEDGEMENTS

This has been a long road since that Fall of 1974 when “Kairos” was the first relay. Many years later the “Centro Activo Freire”. Those years in the U.N.A.M. have long passed. Now the University of Pennsylvania. This work would not have been possible without the assistance or support of many individuals and institutions.

First of all I would like to thank the guidance and friendship of Dr. Gabor T. Herman during these years. Without his guidance this project would not have been possible. Dr. Herman is only one of many who have examined this work, I would like to thank Dr. Kenneth R. Foster, Dr. Robert M. Lewitt, Dr. Dimitris N. Metaxas, Dr. Jayaram K. Udupa, and Dr. Felix W. Wehrli, the members of the committee in charge of reviewing this dissertation.

This dissertation could fall into the category of “Nomad Ph.D.s” as I have visited several laboratories in different parts of the U.S. and abroad. I first arrived to the Medical Image Processing Group at the University of Pennsylvania, then to the Biocomputing Unit at the C.N.B. (Spain), later to the Center for Computer Science and Applied Mathematics (Temple University) and finally to the Mathematical Sciences Research Institute (Berkeley), respectively. The final experiments and the final version of my thesis were finished in the Discrete Imaging and Graphics Group at the Graduate Center of the City University of New York. In all these laboratories and institutions I was always welcome and assisted by their members. In particular I want to thank the support from José María Carazo, Gabor T. Herman and Jayaram K. Udupa. I am also very thankful to Samuel Matej, Robert M.

Lewitt, Roberto Marabini, Rasmus Schröder, Carlos Oscar Sánchez, Bruno Carvalho, T. K. Narayan, José Jesú Fernández, and Pedro de Alarcon for their help, guidance and their time.

During these years of graduate school I have received the invaluable financial and economical support from several institutions whose support and trust I will always thank: the Consejo Nacional de Cienca y Tecnología (México) and the International Institute of Education (México and U.S.), the National Institutes of Health through grants HL28438 and HL70472, the National Science Foundation through the grant DMS96122077 and the Consejo Superior de Investigación Científica, (Spain).

Clearly, it would be impossible to “survive” graduate school without feeling like home. In these years I have met amazing people who did exactly that, my gratitude and love to all my dear friends and “relatives” all around the world: Margie, Vasilis, Katerina, Octavio, Daniel, Donna, Lisa, Kristina, Jonas, Teresa, Natalia, Pedro, Paola and Julia.

There has been a group of people who have been there no matter the difficulty, the distance, and always in my heart. Nada de esto sería posible sin Oliva, Leopoldo y Gustavo: ¡¡mil gracias!!.

ABSTRACT

VISUALIZATION AND EXTRACTION OF STRUCTURAL COMPONENTS FROM RECONSTRUCTED VOLUMES

Edgar Garduno

Gabor T. Herman, Ph.D.

Algorithms for image reconstruction from projections have expanded our ability to visualize the structure of objects. Image reconstruction covers a wide range of physical applications from electron microscopy of molecular structures to radio astronomy of cosmic structures. In this work we will focus on molecular structures, namely biological macromolecules imaged by a transmission electron microscope (*TEM*) and then reconstructed from such images.

Biological macromolecules are the main structural components from which living matter is built. This group of molecules consists of four broad subclasses: proteins, nucleic acids, polysaccharides and lipids. All of these macromolecules exhibit a complex structure which determines the interaction of the macromolecule with itself or with others. In particular, proteins have a functional role which is not present in the other three macromolecules. Different proteins carry out their diverse functions due to the extremely wide range of configurations they can exhibit. Indeed, it is believed that this specialization is largely due to the structure they possess.

TEM technology has been commonly used to obtain reconstructions of macromolecu-

lar structures because of the meaningful information this technology produces. Algebraic Reconstruction Techniques (*ART*) are methods for the reconstruction of macromolecular complexes in TEM. The process of reconstructing a macromolecular complex yields three-dimensional arrays of real numbers representing the macromolecular complex. After reconstruction, the approximation of the macromolecular complex is generally corrupted by noise, resulting from the noise in the data collection, and by errors inherent in the reconstruction algorithm. As a result, the reconstruction is typically an imprecise approximation of the macromolecular complex. Consequently, this approximation has to be further processed to obtain an accurate representation of the macromolecule.

Reconstructions with ART produce volumes expressed as linear combinations of some basis functions. Recently, spherically symmetric functions (*blobs*) have been introduced as efficacious basis functions for reconstruction. In this dissertation we propose a method of selecting blob parameters to obtain accurate reconstructions and visual representations of the three-dimensional structure of macromolecular complexes.

ART produces as output the set of weights for the basis functions, that we refer to as the set of coefficients. In order to produce fast and accurate visual representations of macromolecules we first propose a method that takes advantage of the properties of ART and blobs, together with simple thresholding of the set of coefficients that provides a "good" estimate of those blobs that contribute to the formation of the object of interest and, thus, simplifying the search for significant blobs. Furthermore, we also take advantage of the

various ways of placing the blobs in the three-dimensional space to produce more accurate visual representations and reduce the number of coefficients.

One characteristic of biological macromolecules, which distinguishes them from specimens used in other fields, is that they very frequently present some kind of symmetry. In fact, the functional form of many biological macromolecules is obtained by merging or joining several copies of one or several subunits. We propose to take advantage of the repetitive presence of subunits to improve the signal-to-noise ratio of the reconstruction of a macromolecular complex.

CONTENTS

Chapter 1

INTRODUCTION

Medical imaging has provided physicians with new tools to diagnose or plan treatment on patients with non-invasive or almost non-invasive techniques. Nowadays the imaging techniques in use in medicine are capable of producing two-dimensional or three-dimensional images from the human body. These devices measure the interaction of organs and tissues with some type of energy, e.g., Computerized Tomography uses X-rays, Positron Emission Tomography uses positrons. The measurements are treated as line integrals, of some spatially-varying physical parameter which is related to the local interactions of the tissue with the energy, through the body from the source to the detector. The distribution of the spatially-varying physical parameter is recovered from these measurements. Medicine and biology also rely on other imaging techniques to visualize microorganisms, pathogens and some organelles. Technology has even provided tools for the dissection of objects at the micro-scale.

At the micro-scale and the body-scale humankind has acquired sufficient knowledge

to recognize structures and shapes so that it is possible to distinguish between a “normal” structure and an “irregular” one. Thus, it is possible to distinguish between a healthy bone and osteoporotic bone, or between a *staphylococcus* and a *vibro cholerae* bacterium from acquired images.

There are several mechanisms by which cells interact with each other, microorganisms and pathogens attack cells, cells protect the body, or organelles produce substances. Microorganisms, pathogens, organelles and the substances they produce are made of molecules that cannot be observed with optical devices. Therefore, it has been necessary to invent other instruments to obtain information on objects at a such small scale, in particular on macromolecules. However, many of the instruments capable of yielding images of nano-scale objects produce only two-dimensional images. For reasons that are exposed later in this dissertation, it is important to obtain the three-dimensional structure of objects at the nano-scale and consequently it is necessary to apply image processing techniques to the two-dimensional images to produce three-dimensional representations. Whereas at the body-scale and micro-scale it is possible to compare the images with the objects imaged, at the nano-scale it is impossible with the current technology to make such a comparison, see Fig. 1.1.

Images produced by a device are usually post-processed. There are two main reasons for the further processing of the images, the first being that imaging devices are not perfect and they introduce noise into the images. Another reason for processing is to compen-

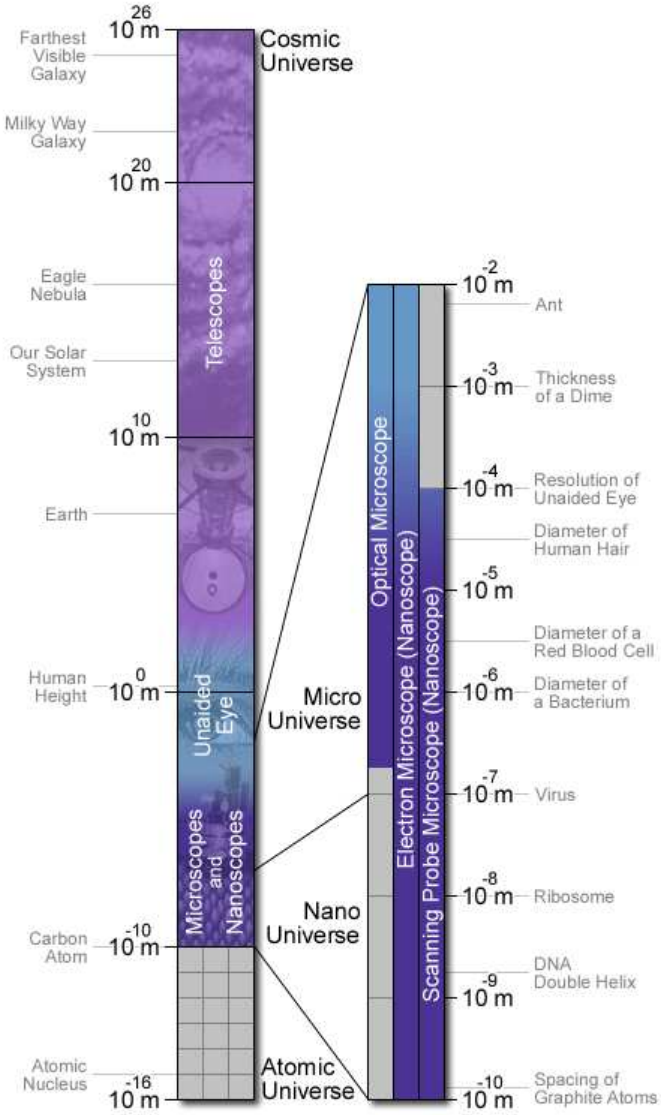

Fig.1.1: Relative size of several objects from the cosmic-scale to the nano-scale together with the instruments used to observe them (image courtesy of the INVSEE project at Arizona State University, Dr. B. L. Ramakrishna, Project Director, INVSEE).

sate for artifacts in computer-produced three-dimensional images. Finally, the user of the images may want to obtain higher level knowledge from the images, such as extracting meaningful portions of the objects or visualizing the objects.

In Chapter 2 we present a summary of the importance of studying macromolecules and proteins, and the methods and techniques used to obtain three-dimensional density functions, together with some of their limitations. In Chapter 3 we propose a method to improve the visualization of the computer representation of macromolecules and in Chapter 4 we propose methods to produce computer representations of the three-dimensional density functions by improving the raycasting method introduced in Chapter 3 and by an approximation, by polygons, to the surface of the density function. Finally, in Chapter 5 we propose to take advantage of the repetition of subunits that form some macromolecular complexes to improve the accuracy of their representation.

BIBLIOGRAPHY

[1] G. D. Abram and L. Treinish, "An extended data flow architecture for data analysis and visualization," in *Proceedings of IEEE Visualization 1995*, pp. 263–269, IEEE Computer Society, 1995.

[2] N. M. Alpert, J. F. Bradshaw, D. Kennedy, and J. A. Correia, "The principal axes transformation – a method for image registration," *Journal of Nuclear Medicine*, vol. 31, pp. 1717–1722, 1990.

[3] L. A. Amos, R. Henderson, and P. N. T. Unwin, "Three-dimensional structure determination by electron microscopy of two-dimensional crystals," *Progress in Biophysical and Molecular Biology*, vol. 39, pp. 183–231, 1982.

[4] L. K. Arata, A. P. Dhawan, J. P. Broderick, M. F. Gaskil-Shipley, A. V. Levy, and N. D. Volkow, "Three-dimensional anatomical model-based segmentation of MR brain images through principal axes registration," *IEEE Transactions on Biomedical Engineering*, vol. 42, pp. 1069–1078, 1995.

[5] E. Artzy, G. Frieder, and G. T. Herman, "The theory, design, implementation and evaluation of a three-dimensional surface detection algorithm," *Computer Graphics and Image Processing*, vol. 15, pp. 1–24, 1981.

[6] T. S. Baker and J. E. Johnson, "Low resolution meets high: Towards a resolution continuum from cells to atoms," *Current Opinion in Structural Biology*, vol. 6, pp. 585–594, 1996.

[7] H. Bandemer and S. Gottwald, *Fuzzy Sets, Fuzzy Logic, Fuzzy Methods with Applications*. Chichester: Wiley, 1995.

[8] M. Bárcena, T. Ruiz, L. E. Donate, S. E. Brown, N. E. Dixon, M. Radermacher, and J. M. Carazo, "The DnaB·DnaC complex: A structure based on dimers assembled around an occluded channel," *European Molecular Biology Organization Journal*, vol. 20, pp. 1462–1468, 2001.

[9] A. Ben-Israel and T. N. E. Greville, *Generalized Inverses: Theory and Applications*. New York: Wiley, 2nd ed., 2001.

[10] F. C. Bernstein, T. F. Koetzle, G. J. Williams, E. Meyer, M. D. Brice, J. R. Rodgers, O. Kennard, T. Shimanouchi, and M. Tasumi, "The Protein Data Bank. A computer-based archival file for macromolecular structures," *Journal of Molecular Biology*, vol. 112, pp. 535–542, 1977.

[11] P. J. Besl and N. D. McKay, "A method for registration of 3-D shapes," *IEEE Transactions on Pattern Analysis and Machine Intelligence*, vol. 14, pp. 239–255, 1992.

[12] J. C. Bezdek and S. K. Pal, *Fuzzy Models for Pattern Recognition: Background, Significance, and Key Points*, ch. 1, pp. 1–34. New York: IEEE, Inc., 1992.

[13] G. Blais and M. D. Levine, "Registering multiview range data to create 3D computer objects," *IEEE Transactions of Pattern Analysis and Machine Intelligence*, vol. 17, pp. 820–824, 1995.

[14] J. F. Blinn, "A generalization of algebraic surface drawing," *ACM Transactions on Graphics*, vol. 1, pp. 235–256, 1982.

[15] I. Bloch, "Fuzzy techniques in medical image processing: On fuzzy distances and their use in image processing under imprecision," in *Third IEEE EMBS International Summer School on Biomedical Imaging, Ile de Berder, France, June 13–21, 1998*, vol. 1, IEEE, 1998.

[16] R. N. Bracewell, *The Fourier Transform and its Applications*. New York: McGraw-Hill Book Company, 3rd ed., 1999.

[17] E. O. Brigham, *The Fast Fourier Transform and Its Applications*. Englewood Cliffs, New Jersey: Prentice-Hall, Inc., 1988.

[18] L. G. Brown, "A survey of image registration techniques," *ACM Computing Surveys*, vol. 24, pp. 325–376, 1992.

[19] M. D. Brunch, *NMR Spectroscopy Techniques*. New York: M. Dekker, 1996.

[20] B. M. Carvalho, E. Garduño, and G. T. Herman, "Multiseeded fuzzy segmentation on the face centered cubic grid," in *Proceedings Second International Conference on Advances in Pattern Recognition ICAPR 2001* (S.-V. Ltd., ed.), ICAPR, Pattern Analysis and Applications Journal, 2002.

[21] B. M. Carvalho, C. J. Gau, G. T. Herman, and T. Y. Kong, "Algorithms for fuzzy segmentation," *Pattern Analysis and Applications*, vol. 2, pp. 73–81, 1999.

[22] I. Catalaa, R. I. Grossman, D. L. Kolson, J. K. Udupa, L. G. Nyul, L. G. Wei, X. Zhang, M. Polanski, L. J. Mannon, and J. C. McGowan, "Multiple sclerosis: Magnetization transfer histogram analysis of segmented normal-appearing white matter," *Radiology*, vol. 216, pp. 351–355, 2000.

[23] D. C. Champeney, *Fourier Transforms and Their Physical Applications*. New York: Academic Press, 1973.

[24] W. Chiu, "What does electron cryomicroscopy provide that X-ray crystallography and NMR spectroscopy cannot?," *Annual Review of Biophysics and Biomolecular Structure*, vol. 22, pp. 233–255, 1993.

[25] A. P. Dhawan, L. K. Arata, A. V. Levy, and J. Mantil, "Iterative principal axes registration method for analysis of MR-PET brain images," *IEEE Transactions on Biomedical Engineering*, vol. 11, pp. 1079–1087, 1995.

[26] J. Dubochet, M. Adrian, J.-J. Chang, J.-C. Homo, J. Lepault, A. W. McDowall, and P. Schultz, "Cryo-electron microscopy of vitrified specimens," *Quaternary Review of Biophysics*, vol. 21, pp. 129–228, 1988.

[27] J. Dubochet, J. Lepault, R. Freeman, J. A. Berriman, and J.-C. Homo, "Electron microscopy of frozen water and aqueous solutions," *Journal of Microscopy (Oxford)*, vol. 128, pp. 219–237, 1982.

[28] T. L. Faber and E. M. Stokely, "Orientation of 3-D structures in medical images," *IEEE Transactions on Pattern Recognition*, vol. 10, pp. 626–636, 1988.

[29] J. D. Foley, A. V. Dam, S. K. Feiner, and J. F. Hughes, *Computer Graphics: Principles and Practice*. New York: Addison-Wesley Co., 2nd ed., 1996.

[30] J. Frank, "SPIDER – a modular software system for electron image processing," *Ultramicrosy*, vol. 6, pp. 343–358, 1981.

[31] J. Frank, *Three-Dimensional Electron Microscopy of Macromolecular Assemblies*. New York: Academic Press, 1995.

[32] J. Frank, M. Radermacher, P. Penczek, J. Zhu, M. Ladjadj, and A. Leith, "SPIDER and WEB: Processing and visualization of images in 3D electron microscopy and related fields," *Journal of Structural Biology*, vol. 116, pp. 190–199, 1996.

[33] G. Frieder, G. T. Herman, C. Meyer, and J. K. Udupa, "Large software problems for small computers: An example from medical imaging," *IEEE Software*, vol. 2, pp. 37–47, 1985.

[34] J. C. Fulton, R. I. Grossman, J. K. Udupa, L. J. Mannon, M. Grossman, L. G. Wei, M. Polansky, and D. L. Kolson, "MR lesion load and cognitive function in patients with relapsing-remitting multiple sclerosis," *American Journal of Neuroradiology*, vol. 20, pp. 1951–1955, 1999.

[35] E. Galin and S. Akkouche, "Incremental polygonization of implicit surfaces," *Graphical Models*, vol. 69, pp. 19–39, 2000.

[36] E. Garduño, G. T. Herman, and H. Katz, "Boundary tracking in 3D binary images to produce rhombic faces for a dodecahedral model," *IEEE Transcations on Medical Imaging*, vol. 17, pp. 1097–1100, 1998.

[37] H. Goldstein, C. P. Poole Jr., and J. L. Safko, *Classical Mechanics*. New York: Addison-Wesley Co., 3rd ed., 2001.

[38] D. S. Goodsell and A. J. Olson, "Structural symmetry and protein function," *Annual Review of Biophysics and Biomolecular Structure*, vol. 29, pp. 105–153, 2000.

[39] D. Gordon and J. K. Udupa, "Fast surface tracking in three-dimensional binary images," *Computer Vision, Graphics, and Image Processing*, vol. 45, pp. 196–214, 1989.

[40] R. Gordon, R. Bender, and G. T. Herman, "Algebraic reconstruction techniques (ART) for three-dimensional electron microscopy and X-ray photography," *Journal of Theoretical Biology*, vol. 29, pp. 471–481, 1970.

[41] G. J. Grevera, J. K. Udupa, and D. Odhner, "An order of magnitude faster isosurface rendering in software on a PC than using dedicated, general purpose rendering hardware," *IEEE Transactions on Visualization and Computer Graphics*, vol. 6, pp. 335–345, 2000.

[42] N. Grigorieff, T. A. Ceska, K. H. Downing, J. M. Baldwin, and R. Henderson, "Electron-crystallographic refinement of the structure of bacteriorhodopsin," *Journal of Molecular Biology*, vol. 259, pp. 393–421, 1996.

[43] A. Gross and L. J. Latecki, "Digital geometric methods in image analysis and compression," in *Lecture Notes in Computer Science, Computer Vision ACCV'98*, vol. 1351, pp. 184–191, ACCV, Hong Kong, China, 1998.

[44] A. Gross and R. Lusinyants, "Fast nonparametric detection of regular structure using digital geometric primitives," in *Vision Geometry VI* (R. A. Melter, A. Y. Wu, and L. J. Latecki, eds.), vol. 3168 of *Proceedings of SPIE*, pp. 338–347, 1997.

[45] R. M. Haralick and L. G. Shapiro, "Image segmentation techniques," *Computer Vision, Graphics, and Image Processing*, vol. 29, pp. 100–132, 1985.

[46] R. Henderson, J. M. Baldwin, T. A. Ceska, F. Zemlin, E. Beckmann, and H. K. Downing, "Model for the structure of bacteriorhodopsin based on high-resolution electron cryo-microscopy," *Journal of Molecular Biology*, vol. 213, pp. 899–929, 1990.

[47] R. Henderson, J. M. Baldwin, J. Downing, J. Lepault, and F. Zemlin, "Structure of purple membrane from *Halobacterium halobium*: Recording, measurement and evaluation of electron micrographs at 3.5 Å resolution," *Ultramicroscopy*, vol. 19, pp. 146–178, 1986.

[48] R. Henderson and P. Unwin, "Three-dimensional model of purple membrane obtained by electron microscopy," *Nature*, vol. 257, pp. 28–32, 1975.

[49] G. T. Herman, *Image Reconstruction from Projections: The Fundamentals of Computerized Tomography*. New York: Academic Press, 1980.

[50] G. T. Herman, "Algebraic reconstruction techniques in medical imaging," in *Medical Imaging, Systems Techniques and Applications - Computational Techniques* (C. T. Leondes, ed.), pp. 1–42, Amsterdam: Gordon and Breach Science Publishers, 1997.

[51] G. T. Herman, *Geometry of Digital Spaces*. Boston: Birkhäuser, 1998.

[52] G. T. Herman and B. M. Carvalho, "Multiseeded segmentation using fuzzy connectedness," *IEEE Transactions on Pattern Analysis and Machine Intelligence*, vol. 23, pp. 460–474, 2001.

[53] G. T. Herman, R. Marabini, J. M. Carazo, E. Garduño, R. M. Lewitt, and S. Matej, "Image processing approaches to biological three-dimensional electron microscopy," *International Journal of Imaging Systems & Technology*, vol. 11, pp. 12–29, 2000.

[54] G. T. Herman and L. B. Meyer, "Algebraic reconstruction techniques can be made computationally efficient," *IEEE Transactions on Medical Imaging*, vol. 12, pp. 600–609, 1993.

[55] G. T. Herman and H. K. Tuy, "Background from mathematical analysis," in *Basic Methods of Tomography and Inverse Problems* (P. C. Sabatier, ed.), pp. 21–68, Bristol: IOP Publishing, 1987.

[56] G. T. Herman and D. Webster, "A topological proof of a surface tracking algorithm," *Computer Vision, Graphics, and Image Processing*, vol. 5, pp. 162–177, 1983.

[57] W. Hoppe, J. Gaβmann, N. Hunsmann, H. J. Schramm, and M. Sturm, "Three-dimensional reconstruction of individual negatively stained yeast fatty acid synthetase molecules from tilt-series in the electron microscope," *Hoppe-Seylers Zeitschrift für Physiologische Chemie*, vol. 355, pp. 1483–1487, 1974.

[58] B. K. P. Horn, "Closed form solution of absolute orientation using unit quaternions," *Journal of the Optical Society of America A*, vol. 4, pp. 629–642, 1987.

[59] D. P. Huttenlocher, G. A. Klanderman, and W. J. Rucklide, "Comparing images using the Hausdorff distance," *IEEE Transactions on Pattern Analysis and Machine Intelligence*, vol. 15, pp. 850–863, 1993.

[60] T. N. Jones and D. N. Metaxas, "Image segmentation based on the integration of pixel affinity and deformable models," in *Proceedings of the IEEE Computer Vision and Pattern Recognition 1998*, pp. 330–337, IEEE Computer Society, 1998.

[61] S. Kaczmarz, "Angenäherte Auflösung von Systemen Linearer Gleichungen," *Bulletin de l'Académie Polonaise des Sciences, Lettres, Series A*, vol. 35, pp. 355–357, 1937.

[62] A. C. Kak and M. Slaney, *Principles of Computerized Tomographic Imaging*. New York: IEEE Press, 1988.

[63] G. Karp, *Cell and Molecular Biology: Concepts and Experiments*. New York: John Wiley and Sons, Inc., 1996.

[64] J. Kendrew, "Myoglobin and the structure of proteins," *Science*, vol. 139, pp. 1259–1266, 1966.

[65] P. E. Kinahan, S. Matej, J. S. Karp, G. T. Herman, and R. M. Lewitt, "A comparison of transform and iterative techniques for a volume-imaging PET scanner with a large acceptance angle," *IEEE Transactions on Nuclear Science*, vol. 42, pp. 2281–2287, 1995.

[66] W. Kühlbrandt, "Bacteriorhodopsin – the movie," *Nature*, vol. 406, pp. 569–570, 2000.

[67] W. Kühlbrandt, D. N. Wang, and Y. Fujiyoshi, "Atomic model of plant light-harvesting complex by electron crystallography," *Nature*, vol. 367, pp. 6144–6621, 1994.

[68] K. Kyte, *Structure in Protein Chemistry*. New York: Garland Publishers, 1995.

[69] R. M. Lewitt, "Multidimensional digital image representations using generalized Kaiser–Bessel window functions," *Journal of the Optical Society of America A: Optics, Image Science, and Vision*, vol. 7, pp. 1834–1846, 1990.

[70] R. M. Lewitt, "Alternatives to voxels for image representation in iterative reconstruction algorithms," *Physics in Medicine and Biology*, vol. 37, pp. 705–716, 1992.

[71] G. Lohmann, *Volumetric Image Analysis*. New York: Wiley-Teubner, 1998.

[72] J. B. A. Maintz and M. A. Viergever, "A survey of medical image registration," *Medical Image Analysis*, vol. 2, pp. 1–37, 1998.

[73] R. Marabini, G. T. Herman, and J. M. Carazo, "3D reconstruction in electron microscopy using ART with smooth spherically symmetric volume elements (blobs)," *Ultramicroscopy*, vol. 72, pp. 53–65, 1998.

[74] R. Marabini, G. T. Herman, C. O. Sánchez-Sorzano, and J. M. Carazo, "Volumetric constraints in 3D tomography applied to electron microscopy," in *Proceedings IEEE International Symposium on Biomedical Imaging: Macro to nano* (IEEE, ed.), (Washington D. C.), pp. 641–644, IEEE Engineering in Medicine and Biology Society and IEEE Signal Processing Society, 2002.

[75] R. Marabini, I. M. Masegosa, M. C. San Martin, S. Marco, J. J. Fernández, L. G. de la Fraga, C. Vaquerizo, and J. M. Carazo, "Xmipp: An image processing package for electron microscopy," *Journal of Structural Biology*, vol. 116, pp. 237–240, 1996.

[76] R. Marabini, E. Rietzel, R. Schröder, G. T. Herman, and J. M. Carazo, "Three-dimensional reconstruction from reduced sets of very noisy images acquired following a single-axis tilt schema: Application of a new three-dimensional reconstruction algorithm and objective comparison with weighted backprojection," *Journal of Structural Biology*, vol. 120, pp. 363–371, 1997.

[77] R. Marabini, M. C. San Martin, and J. M. Carazo, "Electron tomography of biological specimens," in *Contemporary Perspectives in Three-Dimensional Biomedical Imaging* (R. J. L. Coatrieux, ed.), vol. 30 of *Studies in Health Technology and Informatics*, pp. 53–77, Amsterdam, Netherlands: IOS Press, 1997.

[78] S. Matej, G. T. Herman, T. K. Narayan, S. S. Furuie, R. M. Lewitt, and P. E. Kinahan, "Evaluation of task-oriented performance of several fully 3D PET reconstruction algorithms," *Physics in Medicine and Biology*, vol. 39, pp. 355–367, 1994.

[79] S. Matej and R. M. Lewitt, "Efficient 3D grids for image-reconstruction using spherically-symmetrical volume elements," *IEEE Transactions on Nuclear Science*, vol. 42, pp. 1361–1370, 1995.

[80] S. Matej and R. M. Lewitt, "Practical considerations for 3-D image reconstruction using spherically symmetric volume elements," *IEEE Transactions on Medical Imaging*, vol. 15, pp. 68–78, 1996.

[81] H. Miyakawa, "Sampling theorem of stationary stochastic variables in multidimensional space," *Journal of the Institute of Electronic and Communication Engineers of Japan*, vol. 42, pp. 421–427, 1959.

[82] D. C. Montgomery and G. C. Runger, *Applied Statistics and Probability for Engineering*. New York: John Wiley and Sons, Inc., 1994.

[83] R. F. Mould, *Introductory Medical Statistics*. Bristol, England: Adam Hilger, 2nd ed., 1989.

[84] S. Muraki, "Volumetric shape description of range data using "Blobby Model"," *Computer Graphics*, vol. 25, pp. 227–235, 1991.

[85] S. Muraki, "Multiscale volume representation by a DoG wavelet," *IEEE Transactions on Visualization and Computer Graphics*, vol. 1, pp. 109–116, 1995.

[86] F. Natterer, *The Mathematics of Computerized Tomography*. Chichester, England: John Wiley & Sons, 1986.

[87] F. Natterer and F. Wübbeling, *Mathematical Methods in Image Reconstruction*. Philadelphia, PA: Society for Industrial and Applied Mathematics, 2001.

[88] E. Nogales, S. G. Wolf, and K. H. Downing, "Structure of the $\alpha\beta$ tubulin dimer by electron crystallography," *Nature*, vol. 391, pp. 199–203, 1998.

[89] L. G. Nyul and J. K. Udupa, "Fuzzy-connected 3D image segmentation at interactive speeds," in *Proceedings of SPIE: Medical Imaging*, vol. 3979, (San Diego, CA), pp. 212–223, International Society of Optical Engineering, 2000.

[90] L. G. Nyul and J. K. Udupa, "MR image analysis in multiple sclerosis," *Neuroimaging Clinics of North America*, vol. 10, pp. 799–, 2000.

[91] H. Oettl, R. Hegerl, and W. Hoppe, "Three-dimensional reconstruction and averaging of 50S ribosomal subunits of *Escherichia coli* from electron micrographs," *Journal of Molecular Biology*, vol. 163, pp. 431–450, 1983.

[92] N. R. Pal and S. K. Pal, "A review on image segmentation techniques," *Pattern Recognition*, vol. 26, pp. 1277–1294, 1993.

[93] S. K. Pal and D. K. Majumder, *Fuzzy Mathematical Approach to Pattern Recognition*. New Delhi: John Wiley and Sons, Inc., 1997.

[94] X. Pennec and J. P. Thirion, "Validation of 3D registration methods based on points and frames," in *Proceedings of the 5th International Conference on Computer Vision (ICCV'95)*, (Cambridge, MA), pp. 557–562, June 1995.

[95] L. Perroton, "A new 26-connected objects surface-tracking algorithm and its related pram version," *International Journal of Pattern Recognition and Artificial Intelligence*, vol. 9, pp. 719–734, 1995.

[96] M. Perutz, "X-ray analysis of hemoglobin," *Science*, vol. 140, pp. 863–869, 1963.

[97] D. P. Petersen and D. Middleton, "Sampling and reconstruction of wave-number-limited functions in N-dimensional Euclidean spaces," *Information and Control*, vol. 5, pp. 279–323, 1962.

[98] R. Radermacher, T. Wagenknecht, A. Verschoor, and J. Frank, "Three-dimensional reconstruction from a single-exposure, random conical tilt series applied to the 50S ribosomal subunit of *Escherichia coli*," *Journal of Microscopy*, vol. 146, pp. 113–136, 1987.

[99] R. W. Ramirez, *The FFT: Fundamentals and Concepts*. Englewood Cliffs, New Jersey: Prentice-Hall, Inc., 1985.

[100] L. Reimer, *Transmission Electron Microscopy: Physics of Image Formation and Microanalysis*, vol. 36 of *Springer series in optical sciences*. Berlin: Springer-Verlag, 1984.

[101] G. Rhodes, *Crystallography Made Crystal Clear: A Guide for Users of Macromolecular Models*. San Diego, CA: Academic Press, 2nd ed., 2000.

[102] A. Rosenfeld, "Fuzzy digital topology," *Information and Control*, vol. 40, pp. 76–87, 1979.

[103] P. K. Saha and J. K. Udupa, "Fuzzy connected object delineation: Axiomatic path strength definition and the case of multiple seeds," *Computer Vision and Image Understanding*, vol. 83, pp. 275–295, 2001.

[104] P. K. Saha and J. K. Udupa, "Relative fuzzy connectedness among multiple objects: Theory, algorithms, and applications in image segmentation," *Computer Vision and Image Understanding*, vol. 82, pp. 42–56, 2001.

[105] P. K. Saha, J. K. Udupa, E. F. Conant, D. P. Chakraborty, and D. Sullivan, "Breast tissue density quantification via digitized mammograms," *IEEE Transactions on Medical Imaging*, vol. 8, pp. 792–803, 2001.

[106] P. K. Saha, J. K. Udupa, and D. Odhner, "Scale-based fuzzy connected image segmentation: Theory, algorithms, and validation," *Computer Vision and Image Understanding*, vol. 77, pp. 145–174, 2000.

[107] M. C. San Martin, C. Gruss, and J. M. Carazo, "Six molecules of SV40 large T antigen assemble in a propeller-shaped particle around a channel," *Journal of Structural Biology*, vol. 268, pp. 15–20, 1997.

[108] T. Schormann and K. Zilles, "Limitations of the principal-axes theory," *IEEE Transactions on Medical Imaging*, vol. 16, pp. 942–947, 1997.

[109] R. R. Schröder, "Zero-loss energy-filtered imaging of frozen-hydrated proteins: Model calculations and implications for future developments," *Journal of Microscopy*, vol. 166, pp. 389–400, 1992.

[110] R. R. Schröder, W. Hofmann, and J.-F. Menetret, "Zero-loss energy filtering as improved imaging mode in cryoelectronmicroscopy of frozen hydrated specimens," *Journal of Structural Biology*, vol. 105, pp. 28–34, 1990.

[111] D. E. Schumm, *Essentials of Biochemistry*. Boston, Little: Brown and Company, Inc., 2nd ed., 1995.

[112] I. I. Serysheva, E. V. Orlova, W. Chiu, M. B. Sherman, S. L. Hamilton, and M. van Heel, "Electron cryomicroscopy and angular reconstitution used to visualize the skeletal muscle calcium release channel," *Nature: Structural Biology*, vol. 2, pp. 18–24, 1995.

[113] G. C. Sharp, S. W. Lee, and D. K. Wehe, "ICP registration using invariant features," *IEEE Transactions of Pattern Analysis and Machine Intelligence*, vol. 24, pp. 90–102, 2002.

[114] U. Tiede, K. H. Hoehne, M. Bomans, A. Pommert, M. Riemer, and G. Wiebecke, "Investigation of medical 3D-rendering algorithms," *IEEE Computer Graphics and Applications*, vol. 10, no. 2, pp. 41–53, 1990.

[115] J. K. Udupa and G. T. Herman, *3D Imaging in Medicine*. Boca Raton, Florida: CRC Press, Inc., 1991.

[116] J. K. Udupa, L. G. Nyul, Y. L. Ge, and R. I. Grossman, "Multiprotocol MR image segmentation in multiple sclerosis: Experience with over 1,000 studies," *Academic Radiology*, vol. 8, pp. 1116–1126, 2001.

[117] J. K. Udupa and D. Odhner, "Shell rendering," *IEEE Computer Graphics and Applications*, vol. 13, no. 6, pp. 58–67, 1993.

[118] J. K. Udupa, P. K. Saha, and R. Lotufo, "Fuzzy connected object definition in images with respect to co-objects," in *Medical Imaging 1999: Image Processing* (K. Hanson, ed.), vol. 3661 of *Proceedings of SPIE*, pp. 236–245, The International Society for Optical Engineering, 1999.

[119] J. K. Udupa and S. Samarasekera, "Fuzzy connectedness and object definition: Theory, algorithms, and applications in image segmentation," *Graphical Models and Image Processing*, vol. 58, pp. 246–261, 1996.

[120] J. K. Udupa, L. Wei, S. Samarasekera, Y. Miki, A. van Buchem, and R. I. Grossman, "Multiple sclerosis lesion quantification using fuzzy-connectedness principles," *IEEE Transactions on Medical Imaging*, vol. 16, pp. 598–609, 1997.

[121] P. A. van den Elsen, E.-J. D. Pol, and M. A. Viergever, "Medical image matching - a review with classification," *IEEE Engineering in Medicine and Biology*, vol. 12, pp. 26–39, 1993.

[122] M. van Heel, "Angular reconstitution: A posteriori assignment of projection direction for 3D reconstruction," *Ultramicroscopy*, vol. 21, pp. 111–124, 1987.

[123] M. van Heel, B. Gowen, R. Matadeen, E. V. Orlova, R. Finn, T. Pape, D. Cohen, H. Stark, R. Schmidt, M. Schatz, and A. Patwardhan, "Single-particle electron cryomicroscopy: Towards atomic resolution," *Quarterly Reviews of Biophysics*, vol. 33, pp. 307–369, 2000.

[124] L. G. Wade, *Organic Chemistry*. Englewood Cliffs, New Jersey: Prentice Hall, Inc., 3rd ed., 1995.

[125] R. H. Wade, "A brief look at imaging and contrast transfer," *Ultramicroscopy*, vol. 46, pp. 145–156, 1992.

[126] A. H. Watt, *3D Computer Graphics*. Reading, Mass.: Addison-Wesley Pub. Co., 2nd ed., 1993.

[127] Z. Y. Zhang, "Iterative point matching for registration of free-form curves and surfaces," *International Journal of Computer Vision*, vol. 13, pp. 119–152, 1994.

[128] H. J. Zimmermann, *Fuzzy Set Theory and Its Applications*. London: Kluwer Academic Publishers, 3rd ed., 1996.

INDEX