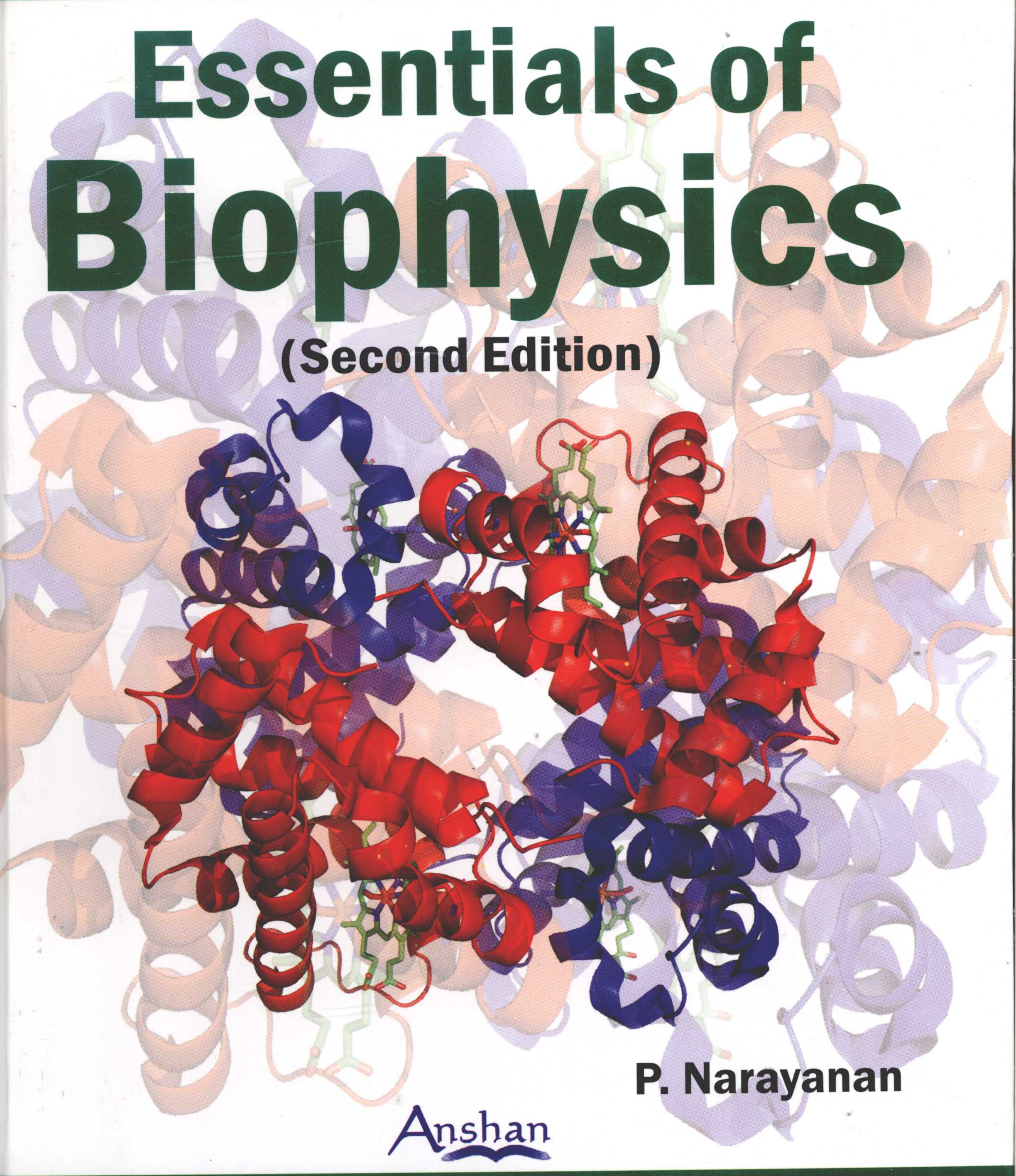
Essentials of Biophysics

(Second Edition)



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Anshan

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Preface to the Second Edition

Biophysics is a multidisciplinary subject—a "bridge" between physical sciences (Physics, Chemistry and Mathematics) with biological, medical and allied sciences. There is a direct correlation and equivalence between physical and biological/medical sciences. X-ray crystallography, NMR and other spectroscopies are increasingly used in structure-function elucidation of biomolecular complexes at atomic and molecular resolutions. While imaging techniques (*e.g.,* microscopies and radiography) have been in use in medicine and biology, advances in microscopic and topographic technologies have ushered in high-resolution and better quality imaging, which have become regular features in modern-day medical diagnostics and therapy protocols.

Recent advances in physics and allied areas– scanning-, and surface-probe imaging methods, Bose-Einstein condensate matter and the possibility of single-atom lasers, laser-traps ("optical- tweezers"), Bioinformatics and nanotechnology, use of biomolecules in computer chips, memory devices (neuronal networks)—, and advances in molecular biology (*e.g.*, genome projects) have brought Biophysics to the centre stage as via medium between the physical and biological/ medical sciences.

As biology and medical sciences are becoming more data-rich, it is becoming increasingly apparent that analytical and computational methods, characteristics of physical sciences, are required to streamline and interpret vast amount of complex data and, therefore, there has been a growing awareness to link physical and biological/medical sciences more closely in research and teaching.

The main aim of this book, therefore, is to present practically all the topics that can be classified as essential to Biophysics. Such topics include chemical bonding (essentials of Quantum physics), Cellular, Molecular and Membrane biophysics, Biophysical chemistry of biomolecules, physiochemical techniques (spectroscopies, microscopies, NMR, X-ray diffraction etc.,), Bioenergetics, Biomechanics, Photo-, Chemo-, and Radiation biophysics, and Bioinformatics.

The contents of the book have been arranged under sections and subsections. An overview is given for each section that comprises several chapters with a common theme. In addition, synopsis is included for each chapter to provide readers a gist of the chapter in compact form. Further mathematical formulas and formalisms, Biostatistics and useful information are provided in appendix to make the book selfsufficient. A list of terms, concepts and phrases in the glossary and index would facilitate the readers a quick access to the technical terms and subject matter in the text. With these efforts, it is hoped that this book would serve the purpose that it is intended for and would catalyze in some way in bringing the subject of Biophysics to play a central role in the advancement of biological, biomedical and related areas.

In this book, a conscious effort has been made to avoid over-emphasis on overtly Physics or overtly Biology topics. Therefore, there is less attention given to Electronics, Computers, and Instrumentation and Genetics, Molecular biology, Biochemistry, Immunology, and Microbiology etc. They are separate subjects in their own right and there are number of books available on these subjects. This book is intended for all students and researchers with Physics, Chemistry, Biology and Medicine background. It is to serve people with biological and medical background to make them knowledgeable about physical principles and techniques, which have become an integral part of biological and medical sciences. It is also intended to serve as an introductory source to make the physicists and chemists and other physical scientists aware of the essential aspects of biological sciences and the trends and progresses in *natural sciences.*

P. Narayanan

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Introduction

The subject of Biophysics has always been and is going to be even more, multidisciplinary, bridging the areas of physical (Physics, Chemistry and Mathematics) and natural sciences (Biology, Pharmacology and Medicine). It is a discipline concerned with the application of the principles of physics and the other physical sciences to the solution of biological/ medical problems. Its main emphasis is on the quantitative analysis of the physical and chemical aspects of the functions of biological molecules, organisms and entities, *vis a vis* their structures at the cellular, molecular and atomic levels. The techniques and methodologies that Biophysics relies on are closer to Physics and Chemistry, but the arena of applications are in the biological, medical and related sciences. There is a direct correlation and equivalence between physical and biological/medical sciences (Table I.1).

Table I.1 Correlation between physics and biology/medicine.

Physics	*Biology/Medicine*
Statics (kinematics)	Skeletal function; orthopedics; internal body mechanics.
Dynamics (Motion)	Spatial movement; flagella, heart and lung function.
Mass transport	Fluid flow; metabolic functions.
Charge transport	Ion transport; nerve impulse transduction; molecular signaling.
Surface tension	Capillary action; fluid transport in plants and cells.
Osmosis	Fluid transport in cells.
Molecular Physics	Structure-function elucidation.
Thermodynamics	Bioenergetics; metabolic and reaction kinetics.
Optics	Spectrosocpics; microscopies; lasers; optics of vision.
Acoustics	Mechano-transduction.
Radiation Physics	Radioactivity; radioassays; diagnostics; imaging and therapy.
Tomographies	Diagnostics and therapy.

Biophysics had a good beginning, due to the efforts of the early stalwarts who had a progressive and wide-ranging outlook on science and technology. Many of the early discoveries in physical sciences were made by such scientists who had diverse scientific backgrounds and interests, and were not constrained by the narrow over-compartmentalisation of knowledge. In fact, quite a number of discoveries in the 19th century in physical sciences were from scientists who were trained in biological and medical sciences. They applied physicochemical methods of analysis to Biology and, being from the biological sciences, they were in a better position to understand the problems and complexities of extending physicochemical methods to the natural sciences.

To cite a few cases: **Luigi Galvani** was a man of medicine who was instrumental in the development of electrochemistry and neurophysiology. **Thomas Young** was a pioneer in optics and vision, in addition to his keen interest in deciphering the Egyptian hieroglyphs. **Hermann von Helmoltz** was a trained physician, but his contributions to Physics and Biology are vast and wide-ranging— from electromagnetism, thermodynamics (conservation of energy), physiological optics (he invented the ophthalmoscope), colour theory and colour vision to physiological acoustics. To such group can be added the names of pioneers like **von Humboldt**. With such a stimulating and holistic approach, the flow of knowledge, from physical to natural sciences and *vice versa*, was on equal footing.

Developments in science in the later part of the 19th century and the early part of 20th century were lopsided, with a narrow compartmentalisation of subjects. The historical development of science, the practical necessity for divisions and the rigid adherence to their narrow areas by the practicing scientists were mostly responsible to such a state of affairs. This situation led to Biophysics, an interdisciplinary subject, relegated and forgotten.

By and large, Biology as practiced till the mid 20th century (till the late 1940s) was descriptive and speculative. Scientists from Biology and Medicine were trained with little or no knowledge of the fundamentals Physics and Chemistry and also the quantitative aspect that physical sciences inculcate. Being devoid of such training, it is not surprising that there was no major cross-fertilisation of scientific ideas from the biological to the physical sciences. While the physical sciences were making epoch-making discoveries from the early part of the 20th century, a large body of biological scientists concentrated their best efforts on the classification and morphological studies, and qualitative trial-and-error and hit-and-miss methods for solution. Such a development in the natural sciences had a tremendous negative impact on Biophysics and its neglected status.

There has been a discernible change, from the mid 20th century on wards in the approach and attitude to tackling the problems in biological, medical and allied sciences with more emphasis on the quantitative approach. This has been partly due to the realisation that—

1. Traditional departments (compartmentalisation) can be physical barriers to cross-fertilisation of ideas.
2. A deeper understanding of complex biological systems will need a more quantitative type of biology that is closely integrated with the physical sciences.
3. Active participation of physical scientists in biomedical-related sciences is essential.
4. Advances in physical and physicochemical techniques and instrumentation. Many of today's high-profile biological/biomedical advances have their roots in physics-related principles and technologies. Biomedical instrumentation is widely used in medical fields.

Some of the physical scientists who provided impetus for the progress of Biophysics in the 20th century include **George Gamow, Max Delbrück, Erwin Schrödinger, Lawrence Bragg, Linus**

Pauling, J.D. Bernal, Dorothy Hodgkin, Max Pertuz, Francis Crick, G.N. Ramachandran, Hodgkin and **Huxley, Claude Shanon, Alan Turing** and many others. The recent change in the mindset towards biology has also been due to the realisation that—

(i) Understanding of the complex arrangement of living matter cannot be achieved by classification, speculation and insights, but by quantitative physicochemical methods.

(ii) Life processes, in spite of varied structural organisation and complexity, do not violate the basic laws of Physics.

(iii) All biological problems can be addressed at the cellular, molecular and atomic levels, due to recent advances in physicochemical techniques and computer-based processing and modeling methods applied to biological systems.

Modern quantitative biology (Genetics, Molecular biology, Biochemistry, Microbiology etc.) relies on the physicochemical methods to understand life processes at the cellular, molecular and atomic levels. Some of these physical techniques are electron microscopies, spectroscopies, hydrodynamical methods, magnetic resonance, and X-ray diffraction. In the early half of the 20th century there was a greater emphasis on Radiation biophysics. This was because of the preoccupation of a large school of physical scientists in nuclear-related fields and due to generous availability of funds, facilities and governmental and industrial patronage. Radiation physics, applied to biological systems, was considered synonymous with Biophysics. The shift towards Molecular Biophysics has been primarily due to feasibility of obtaining complete three-dimensional architectures of biological macromolecules by X-ray diffraction at molecular/atomic resolutions. Starting from **Max Perutz** and **John Kendrew**, who were instrumental in solving the three-dimensional structures of proteins, hemoglobin and myoglobin, respectively, X-ray crystallographers played (and are playing) a very important role in extending the quantitative approach in biological and medical sciences. The **Watson-Crick** model of the DNA molecule brought together the structural and biochemical approaches to Genetics and Molecular biology, and has been instrumental to bringing the Molecular biology to the forefront. Recent advances in solution studies by multidimensional nuclear magnetic resonance (NMR) spectroscopic techniques have brought forth an additional quantitative physical modality for elucidating the three-dimensional structure and functional features of smaller molecular size proteins, nucleic acids and other biomolecuar entities of importance.

Progress made in the chemical foundation of life, that is Biochemistry, has been due to important developments in Molecular chemistry by hydrodynamical methods, sequence determination by conventional and advanced methods (e.g. mass spectrometry), and tracer and enzyme-labelled techniques. Contributions by various groups to Bioenergetics (**Katalsky, Prigogione** and others of the Brussels's group); and in computer technology— the development of mathematical logic and formalisms for developing artificial intelligence and robotics (Cybernetics)—, initiated by **von Neumann**, **Norbert Wiener**, **Alan Turing**, **Claude Shanon** and others brought about the influx of computer era in biology and medicine, and emergence of Bioinformatics to the centre stage in biological/biomedical sciences.

Spectacular advances in Molecular biology in recent years have also contributed greatly in bringing Biophysics to the centre stage. For example: application of innovative physical techniques in Molecular biology— fluorescence methods, tagged with auto-fluorescent proteins (such as green fluorescence protein), enables the imaging of molecular interactions and dynamics in the cell; gene selection and amplification techniques (by polymerase chain reaction and other methods) and selective methods/mutation methods (such as site-directed mutagenesis) allow greater sophistication and precision to biologists to modify or manipulate biological macromolecules in a more rational and planned way. The feasibility of obtaining three-dimensional structures of biological molecular species by X-ray diffraction and NMR spectroscopic methods in a reasonable time, and computer-modelling (*computational biology/molecular tinkering)* methods offer solutions to rational design protocols to obtain new types of molecular species (*virtual mutants*). Molecular engineering, which is a combination of molecular biological methods, physical techniques (primarily X-ray diffraction and NMR spectroscopy), and knowledge-based computer modeling procedures (*Bioinformatics*) will play a central role, in the progress of biological and medical sciences, in our understanding of the functions of biological systems *vis a vis* their structure, and in designing new molecular species, tailor-made to improve or suit specific requirements.

Imaging techniques— radiological and optical, electron and other microscopies have been in use in medicine and biology. But, recent advances in imaging techniques— radiological (CT), nuclear (SPECT and PET), magnetic resonance (MRI and fMRI) and ultrasonography— have greatly enhanced the clarity and resolution of images, very useful in diagnostic procedures and organ function as well as therapy. Further, developments in scanning-probe microscopy (SPM) technology— laser scanning confocal (LSM), scanning-tunneling (STM), atomic force (AFM), aperture-less and chemical microscopy— augur fundamental changes in developing new methods to examine physical and chemical properties of single macromolecules.

Application of recent developments in physical sciences— *monoatomic* condensed matter (Bose-Einstein condensate), quantum dots, single-atom lasers, laser-traps ("*optical tweezers*"), Bioinformatics and nanotechnology will be the basis for future developments, for exquisitely sensitive measurements on biomolecules at atomic resolution, use of biomolecules in computer chips, memory deices (neuronal networks) and solar-energy cells etc., thus bring Biophysics to the centre stage, as *via* medium between physical and natural (biological/ biomedical) sciences.

As biological and medical sciences are becoming data-rich (e.g. with the advent of genome projects), it is becoming increasingly apparent that analytical and computational methods, characteristics of physical sciences, are required to streamline and interpret vast quantity of complex of data. With this growing awareness, there have been conscientious efforts to link physical and natural sciences more closely in research and teaching, to impart biologists the ability to separate biological problems into segments that are amenable to exact physical interpretation and to formulate hypotheses that can be tested by experiment. It is hoped that by imparting biological streams with more Physics, Chemistry and Mathematics— that is, orienting them to Biophysics (*Integrative biology*)— they can be made to expand their horizons and adapt quantitative approaches in research and teaching, for example, the ability to use complex physical theories (e.g. X-ray diffraction techniques) to analyse natural entities (cells; molecules etc.).

But, imparting physical sciences courses to biological sciences should not be done as a separate exercise or as an additional material. The approach should be integral and holistic, with concepts and principles of physics and chemistry and necessary mathematical formalisms and quantitative tools of analytical analysis forming an integral part of such an approach in biological and medical sciences. In the absence of such a holistic approach, even subjects like Biochemistry and Molecular Biology, which have closer affinity to physical sciences, would become "soft" subjects. Therein lies the importance and centrality of Biophysics, in the emerging areas in biological and medical sciences.

Abbreviations

AAS : Atomic Absorption Spectroscopy
AES : Atomic Emission Spectroscopy
AFM : Atomic Force Microscopy
$\alpha t \alpha$: Helix-Turn-Helix
CARS : Coherent AntiStokes Raman Spectroscopy
CD : Circular Dichorism
CIA : Chemiluminescence ImmunoAssay
DIC : Differential Interference Contrast
ENDOR : Electron Nuclear Double Resonance
ESR : Electron Spin Resonance
FID : Free Induction Decay
FRET : Fluorescence Resonance Energy Transfer
FT : Fourier Transform
GC : Gas Chromatography
HPLC : High-Performance Liquid Chromatography
IEP : IsoElectric Point
IMRA : ImmunoMetricRadioAssay
IPG : Immobilised pH Gradient
LCAO : Linear Combination of Atomic Orbitals
MRI : Magnetic Resonance Imaging
MS : Mass Spectrometry
NMR : Nuclear Magnetic Resonance
PAGE : PolyAcrylamide Gel Electrophoresis

PET	:	Positron Emission Tomography
PMT	:	PhotoMultiplier Tube
RIA	:	RadioImmunoAssay
SDS	:	SodiumDodecyl Gel Electrophoresis
SEM	:	Scanning Electron Microscopy
STM	:	Scanning-Tunnelling Microscopy
SPECT	:	Single-Photon Emission Computed Tomography
STEM	:	Scanning Transmission Electron Microscopy
TEM	:	Transmission Electron Microscopy

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