Bone MarroW: Structure and Function

Mehdi Tavassoli and
Joseph Mendel Yoffey

Bone Marrow
Structure and Function

Mehdi Tavassoli, M.D., F.A.C.P.
Professor of Medicine
University of Mississippi School of Medicine
Chief, Hematology-Oncology
Director, Cell Biology Laboratory
Veterans Administration Medical Center
Jackson, Mississippi

Joseph Mendel Yoffey, D.Sc., M.D., F.R.C.S. (ENG.)
HON. LL. D. (Manchester)
Emeritus Professor of Anatomy
University of Bristol
Visiting Professor of Anatomy
The Hebrew University-Hadassah
Medical School, Jerusalem

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Foreword

Science in our time is incredibly fashion-oriented. Certain subjects and areas may now ride the crest of popularity only to be buried in the well of oblivion before long. They may be reincarnated again. The reasons for this periodicity in the popularity of scientific subjects are not clear, but the tendency has been increasingly amplified as science has become a mass profession and therefore heavily dependent on public funds. The nefarious influence of mass media may be another factor.

A deleterious effect of this fashion orientation is the neglect that non-fashionable areas may suffer. Certain disciplines may entirely be submerged and never recover. Others may move with some regularity from the top to the bottom and vice versa. And yet others may not be totally neglected but the angle of study may become fixed, thus providing a channelized rather than comprehensive view.

Studies of the bone marrow have suffered this last distortion. An observer sitting through one of the frequent meetings and symposia on this subject may come out with the impression that the bone marrow grows in culture dishes and not inside the bone. Undoubtedly, the application of tissue culture techniques, and particularly the clonal analysis of hemopoietic progenitor cells, has given a great impetus in furthering our understanding of hemopoiesis. It has opened new vistas in this field and brought to the surface many new questions for resolution. By simplifying the system, the application of tissue culture permits the analysis of different steps in hemopoiesis. Yet, the marrow is clearly a complex tissue which can also synthesize and integrate all these different steps in such a way that it can respond, in an orderly fashion, to physiological regulatory stimuli. The present volume is an attempt to present a different view of the bone marrow: a view that has been considerably neglected in recent years; a view that considers the marrow as the organ which it actually is. In this attempt the book intends to rectify, to a certain extent, the channelization of views on this subject.

Because we intend to treat only certain neglected aspects of bone marrow studies, the book has been designed as a collection of essays on particular aspects of this subject and not as a complete treatise on bone marrow as a whole. Some of the material has previously appeared in a different form and has been revised and updated in this volume. Because much of the data, collected in this volume, are scattered throughout the literature, this book can be of some reference use as well. Chapters I, II, III, IV, VI, and XI were written by M. Tavassoli and chapters V, VIII, IX, X, XII, XIII, XIV are the work of J.M. Yoffey. Chapter VII is a collaborative effort. J.M. Yoffey would particularly like to place on record his thanks to the following for their kind permission to draw freely on previously published material: (1) Messrs. Edward Arnold, publishers of "Bone Marrow Reactions" in 1966, (2) The Charles C. Thomas Co., publishers of "Bone Marrow in Hypoxia and Rebound" in 1974, and (3) The Academic Press. For permission to use previously published materials in chapters I and XI, M. Tavassoli is grateful to McGraw-Hill and Grune & Stratton, and adds his thanks to Academic. We are also grateful to Mrs. Jackie Davis whose highly organized administrative assistance was indispensable to the completion of this volume.

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About the Authors

Mehdi Tavassoli is Professor of Medicine at the University of Mississippi Medical Center. He was trained as a hematologist at Tufts University in Boston where he subsequently began a research career in hemopoiesis. For a decade he was affiliated with Scripps Clinic and Research Foundation in LaJolla, California. His research interest centers around the bone marrow and its hemopoietic function, a subject about which he has published extensive original work. He was among the first to demonstrate the microenvironmental requirements for hemopoiesis and to study their nature, and interactions with hemopoietic cells. He has received numerous awards for his original research and has trained a number of investigators who have subsequently done, in their own right, meritorious research in hemopoiesis.

A pioneer investigator and acknowledged world authority on the lymphoid tissues and bone marrow, **Joseph Mendel Yoffey** has pursued an illustrious and influential career devoted to the service of anatomy. In the 1940s he became especially interested in the lymphocytes and related cells in mammalian bone marrow. He developed the concept, which has since stimulated extensive research, that the bone marrow and lymphoid tissues constitute a single system, "The Lymphomyeloid Complex," unified by a continuous interchange of cells. The bone marrow is the central organ of the complex, and provides its main cellular driving force.

After reaching the conventional retirement age he went as a Visiting Professor to the Australian National University to produce a final edition of his major book. Since 1967 he has resided in Jerusalem and held an active Professorship in the Department of Anatomy, Hebrew University-Hadassah Medical School.

I.

Historical Perspective

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INTRODUCTION

The marrow of our bones is the seedbed of our blood. Like blood, it is essential to life. It is, after the blood itself, the largest and most widely dispersed organ in our body. We harbor more than 1 trillion cells in our marrow at any one time. Every day more than 200 billion red cells, 10 billion white cells, and 400 billion platelets are produced in the marrow. Here is where all lymphocytes and scavenger monocytes originate. A variety of other functions are attributed to the marrow. Birds carry air in their marrow not only to aid in levitation but apparently to serve a respiratory function as well [Meyer and Meltzer, 1916]. There is an interesting cyclic change in pigeons: Before ovulation, the marrow cavity is almost entirely obliterated by bone, which is then resorbed during ovulation and the bone minerals are used to form the egg shell [Bloom et al, 1941].

As one might surmise, a production center of this magnitude is highly vulnerable to malfunction or to the deleterious effects of various factors such as anticancer drugs. In fact, the marrow is currently the single most important limiting factor in cancer treatment. The reason that the treatment of cancer is often not definitive is because the marrow cannot tolerate it. However, the marrow is endowed with considerable potential for self-renewal, which mitigates the impact of its exquisite sensitivity. In this regard its wide dispersion is a distinct advantage.

We have not known all this for very long. For centuries, poets, healers, and philosophers saw and described the close link between blood and life. Not so the marrow. Its role as the seedbed of blood lay hidden, like a seed in the soil. It began to sprout hardly more than a hundred years ago when Ernst Neumann (Fig. I.1) and Giulio Bizzozero (Fig. I.2) established the link between blood and marrow. Ever since, marrow research has been a fertile field, fruitful not only to medicine but to the fundamental understanding of life itself. Scientists have used the marrow as a model for the elucidation of basic questions in biology. In some instances, new fields of biomedical

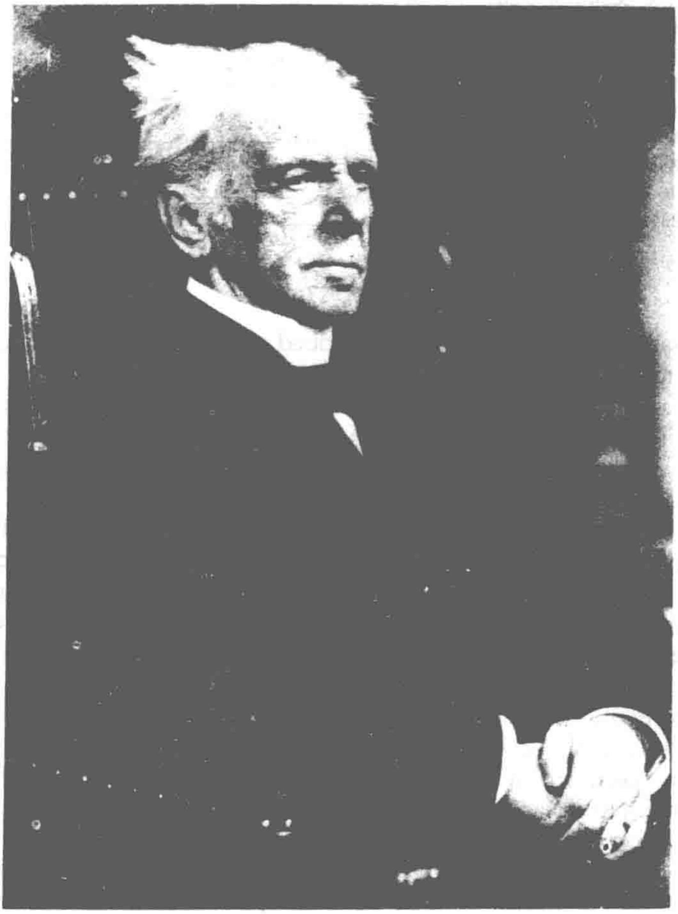

Fig. I-1. Ernst Neumann (1834–1918).

Fig. I-2. Giulio Bizzozero (1846–1901).

research have emerged from studies of the marrow—eg, radiobiology, cell kinetics, and transplantation.

A TRACE IN THE REALM OF IDEAS

In most languages, marrow denotes the inmost of the central part. Metaphorically, it connotes the essence, the substance, the vital part, or the "goodness." Thus, in the prologue of Gargantua, Rabelais invites us to "break the bone and to suck the substantive marrow." And in Hamlet, we are told:

> It takes
> From our achievement through perform'd a height
> The pith and marrow of our attribute.

From the ancient days, the marrow of animals was used for food and was considered to be rich and nutritious. During the 12th century, marrow was considered a "dainty," and cookbooks gave recipes for preparing it. In 1539, Sir Thomas Elyot thought, "Marrowe is more dilectable than the brayne."

In modern times, as everything came to have a scientific aroma, the nutritious effect of marrow was tested by physicians. In the 1890s, first Brown-Sequard [Brown-Sequard and d'Arsonval, 1891, 1892] and then others fed marrow to patients with blood dyscrasias, but to no avail. The matter was then laid to rest only to be revived in the 1920s. Whipple's study of the effects of different foods on hemoglobin production stimulated further interest. Isolated, anecdotal case reports claimed that patients recovered from blood dyscrasias after eating marrow. By 1929, however, it was clear that the only nutritious effect of marrow was due to its iron content. These experiments were the forerunners of marrow transplantation, as some physicians naively hoped that they could transfer living cells by feeding the marrow [Pegg, 1966].

During the 16th and 17th centuries, the marrow was considered a source of warmth, energy, and inner heat: "Thy bone is marrowless, thy blood is cold," said Shakespeare. "Love" was said to burn, or to "melt the marrow." Perhaps in this connotation, the marrow was also considered the seat of vitality and strength: "Marrowy and vigorous manhood," said Oliver Wendell Holmes. "Spending his manlie marrow in her armes," said Shakespeare. Prior to the discovery of its blood-forming function, the marrow was believed to be the source of bone nutrition. Identification of large bones with physical strength and manhood might have led to the designation of the marrow as a source of strength. In 1926, Mechanik, who was measuring the volume of the marrow, found that "under comparable conditions, man has more marrow than woman, a highly noteworthy characteristic of the normal sex differences in man until now unknown." The major product of the marrow, red cell mass, also is known to be lower in the female sex than the male.

THE PATH OF A DISCOVERY

Historic events do not take place in a vacuum. The course of history is a continuum wherein every event relates to a preceding one and leads to the next. Neumann's revelation that the marrow is the seedbed of blood was the culmination of a search for the origin of red cells that had begun much earlier.

Red cells were first described in the 17th century, but it was not until the 19th century that a search for their origin could begin. The intervening period, the entire 18th century, was spent in a seemingly endless squabble, which achieved little more than establishing the identity of the red cell. In fact, for biology as a whole, this was a century of indolence, torpor, and inaction. Nothing positive could be achieved without the synthesis of a conceptual frame that could serve as a *point de depart* for future work; this came in 1838 with the formulation of the cell theory.

The formulation of a cell theory, the conceptualization of the cell (the "little room") as the fundamental unit of life, was the dawn of a new era in biology. It was conceived in 1838 by Mathias Schleiden [Schleiden, 1838] and Theodor Schwann [Schwann, 1847]. From then on, biology moved rapidly. The rest of the 19th century was the *aurea aetas*, when the foundations of many disciplines were laid—bacteriology and immunology, pathology and histochemistry, modern biochemistry and genetics, and antisepsis and modern surgery. This was the century that provided great workers in biology. The essence of this period is well reflected in two quotations from Claude Bernard. In 1855, when he was appointed professor of experimental medicine, the opening sentence of his inauguration lecture was, "Experimental medicine which I am supposed to teach you, does not exist." Some 15 years later, as the president of the Paris Academy of Sciences, he amended this statement: "The dawn of experimental medicine is now visible on the scientific horizon."

It was within this scientific ambience that the search for the origin of red cells began and for several decades was focused on embryonic life. This was only natural: Scholars of this period did not know that blood formation is a continuous process, and takes place throughout life. The finite life-span of red cells, and therefore the necessity for their continuous replenishment, was not recognized. As late as 1923, Peyton Rous wrote, "So subtly is normal blood destruction conducted and the remains of the cells disposed of, that, were it not for indirect evidence, one might suppose the life of most red corpuscles to endure with that of the body" [Rous, 1923]. As late as 1905, Jolly found remnants of the nucleus in some red cells and none in others. He postulated two cell lineages and wrote, "In search of their origin, I have naturally searched the blood of mammalian embryos" [Jolly, 1905]. Evidently, the assumption was that blood cells, once formed in the embryo, remain in the body throughout life.

Neumann is rightly credited with the recognition of the marrow as the seat of blood formation. However, it is generally unrecognized that, conceptually, his most fundamental contribution was his recognition that blood formation is a continuous process, occurring during postnatal life. It was this concept that formed the frame of reference for much of the work that followed. His first brief communication of 1868 does not reflect this, suggesting that he attained this conceptual view gradually. But, the opening paragraph of his 1869 note [Neumann, 1869a] reads:

> The present work intends to demonstrate the physiologic importance of the bone marrow and that it is an important organ for blood formation which has not been recognized. It operates continually in a *de novo* formation of red blood cells.

To reach this conclusion, Neumann used deductive logic based on a premise that later proved incorrect. For a different reason, however, the conclusion remains valid: Neumann thought that proliferation of marrow cells occurred inside the blood vessels of the bone marrow, and reasoned that these continuously proliferating cells must also continuously move out into the general circulation; otherwise the blood circulation in the marrow would stop. We now know that red cell proliferation does not take place inside the blood vessels, but Neumann's conclusion remains valid because all blood formation takes place within a fixed volume inside a rigid frame of bone, where for every cell that is born, within or outside the blood vessels, one must leave to maintain the fixed volume.

Here, a corrective note is necessary. Most historical introductions on the marrow suggest that a substantive contribution was made by Claude Bernard [Michels, 1931; Ness and Stengle, 1974]. These all refer to Volume 68 of *Comptes Rendues* of the Paris Academy of Sciences. Examination of the original document [Neumann, 1869a] indicates that in this particular year, Claude Bernard, in his capacity as a member of the Academy, introduced a paper by Neumann, who was not a member. The title reads, "The Function of Bone Marrow in the Formation of Blood. Note by Mr. Neumann, presented by Mr. Claude Bernard."

Opposition to Neumann's discovery was most intense in Paris, where almost every eminent histologist had a theory on red cell production (vide infra). Bernard recognized Neumann's depth of vision and strongly supported his views. But there is nowhere, in this or other volumes of *Comptes Rendues*, an indication that Claude Bernard himself made a substantive contribution to this subject.

A VISIONARY DUO

Neumann's discovery was announced in the form of a preliminary report, which appeared as the lead article in the issue of 10 October 1868 of the

Centralblatt fur die medizinischen Wissenshaften [Neumann, 1868]. Here is a translation of "About the Significance of Bone Marrow for Blood Formation, Preliminary Communication by Prof. E. Neumann":

> In the so-called red bone marrow of man as well as the rabbit, one can regularly find, in addition to the well-known marrow cell, certain other elements which have not been mentioned until now; namely nucleated red blood cells, in every respect corresponding to embryonic stages of the red blood cells.
>
> Also in the marrow rich in fat, the same cells are present but in lower quantity and their number decreases parallel to the decrease in the number of marrow cells and the increase in the number of fat cells.
>
> It is possible to trace the origin of these elements to the marrow cells. The high content of colorless elements in the blood of the marrow makes it likely that there is a migration of contractile marrow cells into the vessels.
>
> A thorough description of my observations will be published.

The promised thorough description appeared the next year in an extensive article in *Archiv der Heilkunde* [Neumann, 1869b]. In the interim, however, two communications appeared in Italian and were soon translated in the *Centralblatt* [Bizzozero, 1868, 1869]. They were both by Bizzozero, confirming the observation that nonnucleated red blood cells are formed from nucleated red cells in the marrow. Bizzozero extended the blood-forming function of the marrow to include the formation of white cells.

A careful reading of these interesting communications leaves one with the impression that perhaps Bizzozero might have come to this conclusion even before Neumann, but that he was unsure of the reception he might receive if his findings were announced. The rapidity with which Bizzozero's announcement appeared following publication of Neumann's announcement supports this speculation. It is worth mentioning that Neumann was a well-established professor in the European tradition [Askanazy, 1918], whereas Bizzozero was but a 22-year-old recent graduate facing considerable opposition in his hometown of Pavia. His appointment to the faculty of medicine was pushed through, thanks to the recommendation of his mentor, Mantegazza, in the face of opposition by other faculty members, who cited his youth [Ghisalberti, 1960]. It should also be noted that in some areas, the views of Neumann and Bizzozero were not exactly identical. Retrospectively, in all these instances, Bizzozero proved to be correct.

Of the two, however, Neumann was a more persistent student of the subject. He continued his work on the marrow, and toward the end of the century produced other classic contributions. Among his "firsts" was the identification of leukemia [Neumann, 1870] as a disease of the marrow. He coined the term "myelogenous leukemia" [Neumann, 1878].

Like Immanuel Kant, Neumann preferred to remain a lifelong citizen of Konigsberg, where he taught and worked almost all his life on blood production and blood pigments. His superb literary taste, reflected in his masterful German writings, provides the profile of a German scholar in the classical sense. Bizzozero, by contrast, led a very unsettled life. Born in Varese, he studied in Milan and completed his medical studies in Pavia. He subsequently trained with Virchow in Berlin and, for a brief period, settled in Torino. He then moved to Rome where he became a senator. The scope of his scientific interest was also varied. His early interest in the vascular system was soon replaced by interest in the marrow, but after a decade, he focused on the coagulation mechanism and recognized and coined the term "platelet." Toward the end of his life, he developed choroiditis, which interfered with the microscopic work. His interest then turned to issues affecting public health. He died at the turn of the century, rather prematurely, at the age of 55.

Even before Neumann and Bizzozero, the transition of the nucleated to nonnucleated red cell had been seen in the liver by Kolliker [1846], a German scholar. The French anatomist Charles Robin [1849] had also come close to this discovery, but he did not recognize the kinship of red cells to marrow cells. He coined the term "marrow cells" (*medullocelles*), which apparently is what Neumann referred to as *bekannten Markzellen* [Neumann, 1868]—the well-known marrow cells.

This frontier of knowledge was thus being explored intensively. Had not Neumann made his discovery known, it would surely have been made by others. It is the curious nature of science that, in Bergsonian terms, it has its own *elan vital*, its own momentum. With some exceptions, humanity is but an instrument of this momentum to expand the boundary of knowledge: "It is not the men that make science; it is science that makes the men [Chargaff, 1968]".*

*Variations on this theme also appear in Paul Valery's *Mauvaises pensées ou autres* [1941, Paris, Corti], wherein he concludes, "Ce qui fait un ouvrage n'est past celui qui y met son nom. Ce qui fair un ouvrage n'a pas de nom." (The one who does a piece of work has no name.) Bertolt Brecht's *Galileo* is even more emphatic on this note: "There is no scientific work that one man alone can write" (Collected Plays, 1972, New York, Vintage.).