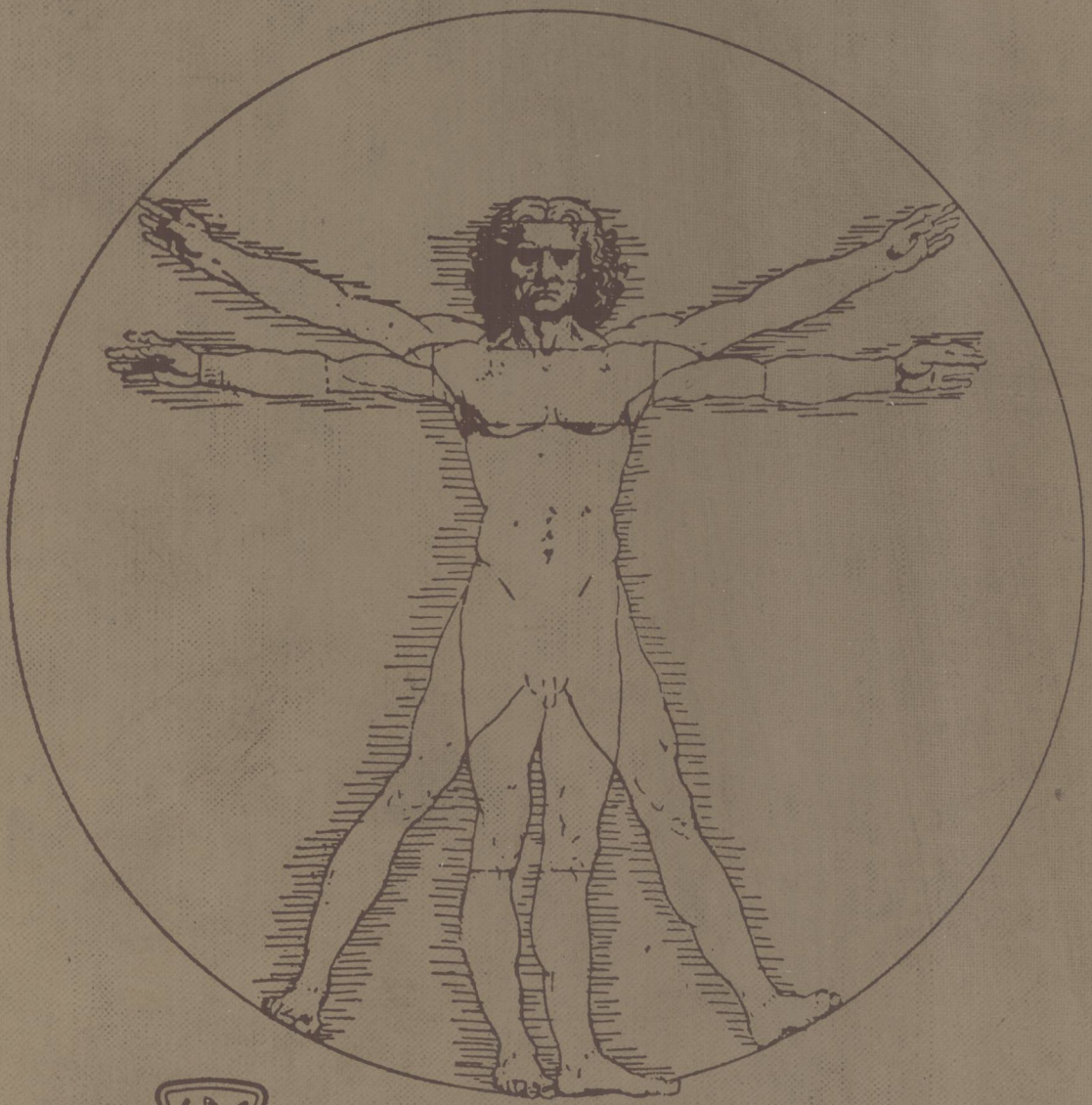
Biocompatible Polymers, Metals, and Composites

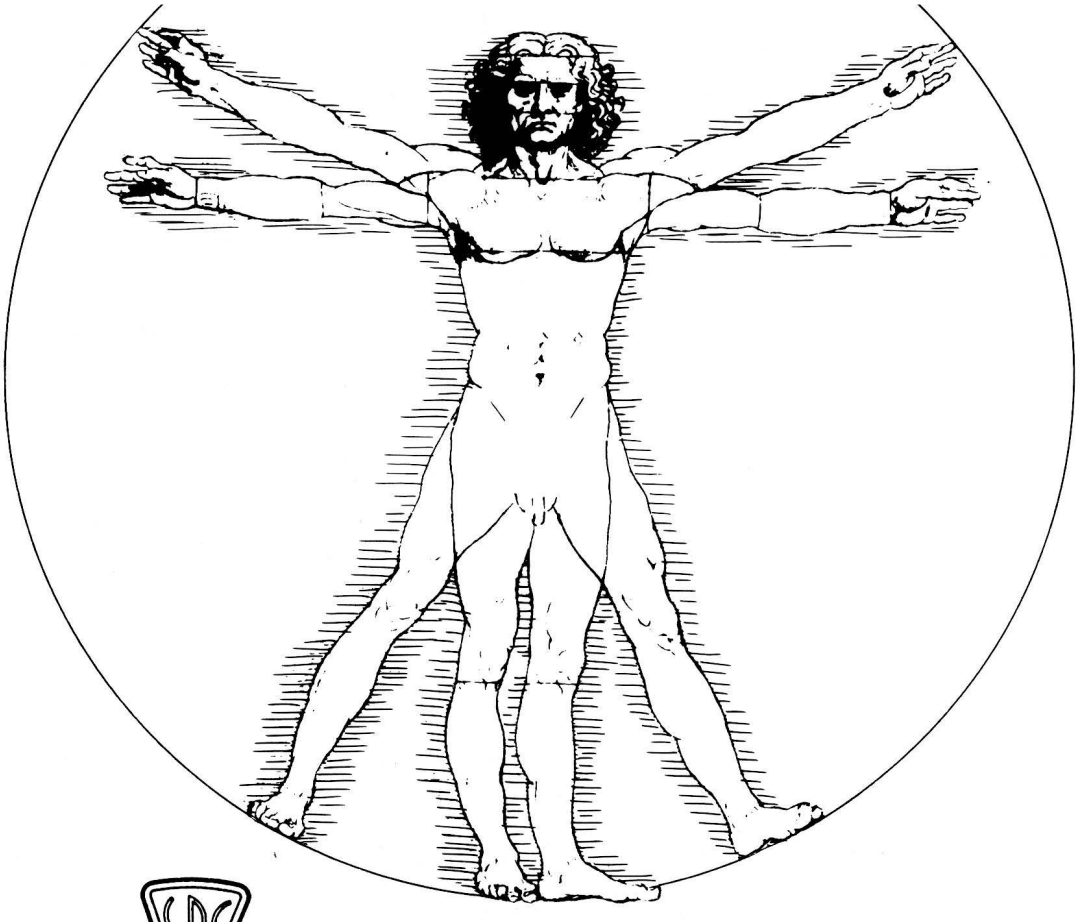
Edited by: M. Szycher, Ph.D.



Sponsored by the SOCIETY OF PLASTICS ENGINEERS, INC.

Biocompatible Polymers, Metals, and Composites

Edited by: M. Szycher, Ph.D.



Sponsored by the SOCIETY OF PLASTICS ENGINEERS, INC.

851 New Holland Avenue
Box 3535
Lancaster, Pennsylvania 17604

Printed in U.S.A.
Library of Congress Card No. 82-74316
ISBN 87762-323-6

DEDICATION

This book is dedicated to my wife, Laurie, and to my sons, Mark and Scott, for their continued support and encouragement.

Foreword

Plastics in biomedical applications continue to be one of the most rapidly growing areas in plastics today. Certainly the technical challenge is one of the most demanding since polymer performance criteria are frequently confounded by polymer compatability with aggressive biological systems. Dr. Michael Szycher, the editor of this landmark volume on biocompatible polymers, metals and composites, is a recognized authority in this highly technical area. He has gathered together many of the leading technical people to contribute their ideas to this book. The book is thoughtfully formatted to allow for easy reader access and this care has been translated into the individual chapter construction. Dr. Szycher is one of the founders of the Medical Plastics Division of the Society of Plastics Engineers and is coeditor of the successful SPE-sponsored volume, "Synthetic Biomedical Polymers", also published by Technomic.

The Technical Volumes Committee of SPE has been charged with surveying and evaluating need for specific technical books since 1956. In that time, it has been the catalyst for publication of more than 50 books in important areas of the plastics industry that have not previously had definitive treatments. The committee works with authors and publishers, reviews and contents of manuscripts to determine their utility to SPE members, and most importantly, thoroughly reviews final manuscripts to ensure accuracy of the technical material.

This concern with technical accuracy and detail is typical of the way in which the Society deals with its other activities—education programs, conferences, periodicals, and meetings. Its greatest resource is its 26,000 practicing plastics engineers and technologists, a resource that has made SPE the largest and most respected organization of its type in plastics worldwide.

The Society of Plastics Engineers is pleased to sponsor and endorse this foundation volume edited by an outstanding SPE member and authored by many of the foremost technical people in the biomedical area today.

SOCIETY OF PLASTICS ENGINEERS, INC.
14 FAIRFIELD DRIVE, BROOKFIELD CENTER, CONNECTICUT 06805
TELEPHONE 203-775-0471

Preface

Technologies have a life cycle of their own, similar in many ways to living organisms. An integral part of both the technological and organic life cycle is the maturation process. For any technical discipline, the maturation process involves greater rationalization; that is, the eventual replacement of the arts-practitioner, who relies heavily on experience and trial-and-error methods, by the professional who operates with prior understanding of fundamental concepts, and is thus capable of predicting, in advance, the expected behavior of a system based on scientifically defined principles.

The field of biomaterials is relatively new, and as such is in the process of undergoing a maturation process. It is apparent that biomaterials technology relies heavily on the classical sciences (chemistry, physics), but is also reinforced by basic engineering, anatomy, histology, hematology, and other disciplines too numerous to mention. A quick review of the biomaterials literature points out that there has indeed been a transition from the pragmatism or serendipity of early investigators, into a more controlled, scientifically based approach. Earlier attempts concentrated on selecting commercially available polymers, cleaning or purifying them as much as possible, and using them as a starting point in the fabrication of prototype prostheses. Lately, the tendency has been to specifically target the most suitable biomaterials to the intended application, biomaterials so specialized and expensive that they may never become commercially viable, except for their use in biomedical applications.

With these thoughts in mind, the intent of this book has been to emphasize, from a scientific viewpoint, the development, manufacture, and testing of those biomaterials most compatible with the human body. The result is that we have cut across disciplines, centers of medical research, and topics, in a deliberate effort to present the broadest view of the field. The contributors to this book represent many of the disciplines cited previously, and the topics originate from investigations conducted in the private, government, and academic sectors.

This book would not have been possible without the efforts of the contributors, who represent the vanguard of this young, emerging field. The sponsorship of the Society of Plastics Engineers, and the support of Thermo Electron Corporation have been invaluable. Specifically, I would like to acknowledge the interest and cooperation of those people who have been there on a daily basis: Victor L. Poirier, Manager of the Biomedical Group, Thermo Electron, for his unwavering support, Dorothy Lee Carchia for her deroic efforts in editing and proofreading, and Thomas L. Coyne for the design of the cover that graces this book.

Michael Szycher, Ph.D.
Director
Biomaterials Research
Thermo Electron Corporation

Waltham, Massachusetts
January 1982

Contents

III
Current Topics in Biomaterials

IV
Emerging Technologies

V

Processing of Biocompatible Materials

Chapter 1

Thrombosis, Hemostasis, and Thrombolysis at Prosthetic Interfaces

Michael Szycher, Ph.D.
Thermo Electron Corporation
Biomedical Systems Division
101 First Avenue
P.O. Box 459
Waltham, MA 02254

I. ABSTRACT

Exposure of blood to artificial surfaces leads to adsorption of plasma proteins within a matter of seconds. Following this adsorption, there may be subsequent deposition of platelets and leukocytes, leading to blood coagulation on the surface of the biomaterial, a phenomenon known as mural thrombosis.

Blood coagulation results from the conversion of a soluble circulating plasma protein, fibrinogen, into insoluble fibrin. Though fibrinogen is soluble in plasma, it is suspended on the brink of insolubility and indeed can be readily polymerized into fibrin on the surface of artificial materials under a variety of conditions. The fibrin that is deposited on the walls quickly forms a meshwork of fibrils, the basis of a coagulum.

The formation of a coagulum may be preceded by platelet adhesion; these two phenomena occur more or less independently. It appears that in the latter stages of blood coagulation on a foreign surface, there is a mutual and cooperative interaction between the coagulation process and platelet aggregation. As it forms, the coagulum entraps other blood elements. Red blood cells appear to play primarily a passive role, adding only bulk. White blood cells are attracted by the thrombus, invade it, and concentrate there. Early in the process, through enzymatic release, leukocytes may contribute to fibrin formation and platelet recruitment; later, these leukocytes may take part in the digestion and resolution of the thrombus (thrombolysis).

II. INTRODUCTION

The increasing utilization of foreign materials in contact with blood has gained great importance in medical practice in recent years. In the past two decades there has been a virtual revolution in the development and clinical applications of blood-handling equipment.

To mention just a few examples, we can cite the now widespread utilization of hemodialysis equipment – better known to the lay public as artificial kidney machines. Hemodialysis machines accept blood from a patient suffering from a derangement in kidney function, purify the blood from certain toxic metabolites, and return the cleansed blood to the patient.

Another example is the worldwide use of cardiopulmonary bypass. This device has permitted the development of open-heart surgery. The instrument pumps blood through the patient, while the heart is stopped to allow for a surgical procedure to take place. At the same time, while the blood is circulating extracorporeally, the device also oxygenates the blood, thus performing the dual functions of both heart and lungs.

Other notable examples include angiographic catheters, chronic insulin monitors, and intraaortic balloon pumps. More recently, an artificial heart device has been developed. This temporary Left Ventricular Assist Device is now undergoing its third year of clinical trials at participating hospitals in the Boston area. This artificial heart is designed to help patients who cannot be weaned from cardiopulmonary bypass, or in patients whose myocardium is profoundly hypokinetic following cardiogenic shock.

The successful development of the Left Ventricular Assist Device, as well as cardiopulmonary bypass, are made possible only through the use of synthetic polymers that are chronically nonthrombogenic. That is, when these devices begin to pump blood, they do not cause blood to coagulate uncontrollably or to embolize, since this would immediately defeat the intended purpose.

This is a remarkable achievement, since normally blood coagulates rapidly upon mere contact with foreign substances. To be able to produce cardiovascular prostheses that are nonthrombogenic, we must first gain a thorough understanding of the basic mechanisms underlying blood coagulation. Only then can we hope to select the biomaterials and configurations most suitable for our purposes.

III. BACKGROUND

In normal arteries and veins, a continuous monolayer of cells, called the endothelium, forms a unique thromboresistant layer between circulating blood and potentially thrombogenic subendothelial tissues. The integrity of the endothelium is a fundamental requirement for maintenance of normal structure and function within the entire cardiovascular system.

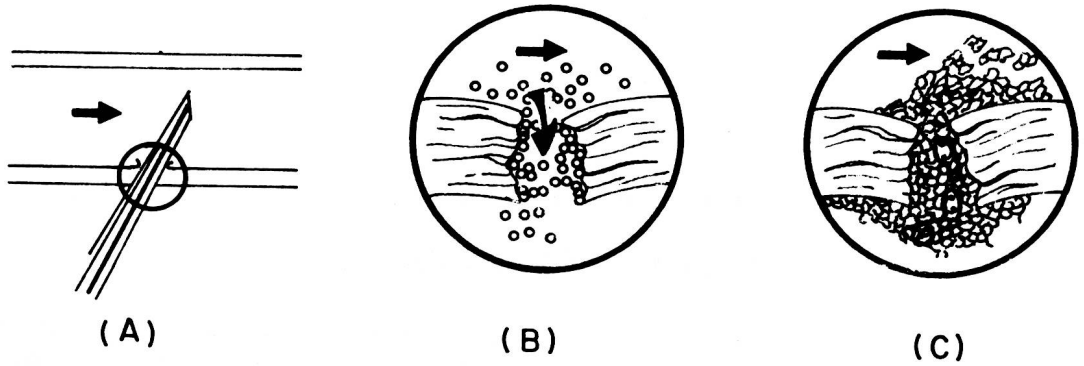
Natural living endothelium is nature's premier nonthrombogenic blood interface. This remarkable hemocompatibility of living blood vessels is undoubtedly derived from the ability of the vessels to take an active role in their thromboresistance; vascular endothelium is a biochemically versatile tissue capable of many synthetic and metabolic functions. For example, endothelial cells produce prostaglandin PGI (prostacyclin), the most potent antiplatelet inhibitor known in biology; endothelial cells also synthesize a plasminogen activator which solubilizes mural fibrin; endothelial cells possess a surface enzyme capable of neutralizing adenosine diphosphate, thereby reducing platelet agglomeration; finally, normal endothelium

has been shown to synthesize heparin – like anticoagulant substances. Thus, this thin cell, hardly visible in routine histologic sections, guarantees freedom from thrombosis and thromboembolism under normal circumstances.

However, what happens where we implant in the cardiovascular system a synthetic polymer that does not possess a living endothelium? Let us assume that we have synthesized an experimental biomaterial, and we wish to use it in chronic contact with blood. If we fashion this biomaterial in the form of an aortic graft, and implant it in the descending aorta, what physiological reactions can we expect? What would happen to the blood? What would happen to the blood-contacting surface of the biomaterial? To answer these questions, we must first examine the normal reactions of the blood, because it is precisely the normal reactions of the blood which will ultimately determine the success or failure of the implant.

IV. MURAL THROMBOSIS AND HEMOSTASIS

Under normal conditions, when a needle punctures a vessel wall, as depicted in Figure 1A, a series of responses is promptly elicited. As the needle is withdrawn from the vessel wall, as shown in Figure 1B, platelets adhere to the raw edges of the wound and then aggregate in an effort to plug the defect in the vessel wall, and thus prevent continuing hemorrhage; while this plug develops across the defect, a similar reaction takes place on the traumatized intimal surface to form an in situ mural thrombus. Platelets also adhere to the exposed connective tissue on the outside of the vessel, as shown in Figure 1C. Fibrin coagulation quickly follows platelet aggregation to further stabilize the mass. If not stabilized, surface fragments are apt to be dislodged by the blood, and carried away as emboli in the bloodstream.



(A) Needle punctures a vessel wall; (B) Platelets adhere to wound; (C) Fibrin coagulation quickly follows, with occasional incorporation of leukocytes. If not stabilized, microemboli may be carried by bloodstream. Arrow = direction of blood flow.

Figure 1. Normal Hemostatic Mechanism.

This simple model exemplifies two basic concepts, thrombosis and hemostasis. Thrombosis is the manifestation of those functions of blood normally responsible for the formation of a clot. The principal mechanisms that have evolved in blood are platelet adhesion, platelet aggregation, and finally, fibrin coagulation (polymerization and crosslinking). Hemostasis refers to the combination of processes that account for the cessation of bleeding after surgery, or trauma to the vascular system; one process contributing to hemostasis involves the

mechanism of thrombosis. Thus, hemostasis implies a return to normalcy, a restoration of equilibrium; therefore when we design and implant a cardiovascular prosthesis, we always strive to regain hemostasis as soon as possible.

Traditionally, blood coagulation was thought to be accomplished by the interaction of platelets and the formation of fibrin; in modern times the incredible complexity of blood coagulation is slowly being unraveled, dramatically illustrated by the many chapters in this book that deal specifically with the thrombogenicity of natural and synthetic blood interfaces.

Blood itself is composed of a liquid medium, plasma, and cellular elements, which constitute approximately 46 percent of the volume, shown diagrammatically in Figure 2. Cellular elements are subdivided into erythrocytes, leukocytes, and platelets, illustrated in Appendix A. Generally, erythrocytes do not form part of the coagulation process per se, although they are generally trapped within the sticky coagulum, giving rise to the familiar dark coloration we associate with a clot. By far the most important role is played by the platelets. Platelet adhesion and aggregation is one of the first steps in the formation of mural thrombi. An implanted biomaterial which is attractive to platelets can be expected to thrombose shortly thereafter.

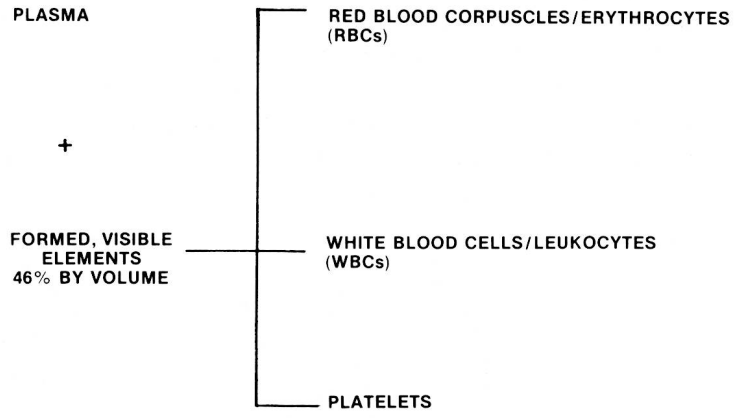


Figure 2. Human Blood Constituents.

V. PLATELETS (THROMBOCYTES)

As we have seen, hemostasis is one of the essential survival mechanisms; in man, hemostasis depends on the clotting of blood pumps, and on the aggregation of a particular type of circulating cell, the platelet. Platelets have only two well-established physiological functions: (1) to form hemostatic plugs in injured vessels, and (2) to provide a phospholipid material (platelet Factor 3) that greatly accelerates plasma coagulation.

Platelets are the smallest of the formed elements of the blood, averaging 1.5 μm in diameter and from 0.5 to 1 μm in thickness as determined by electron microscopy [1]. Normally, about $250 \times 10^3/mm^3$ (range 140 to $440 \times 10^3/mm^3$) are present in the circulation (range 140 to $440 \times 10^3/mm^3$ [2]. Microscopically, they appear as flat discs [3] and remain viable for 9 to 12 days in man, as estimated by in vitro whole population labeling with ^{51}Cr-chromate [4,5].

Ultrastructurally, the interior of the platelet is seen to contain a variety of granules, the least numerous being the mitochondria, comprising roughly 15 percent of the granule population. Two other intracellular storage granules are present: they are called, respectively, dense

granules and α-granules. Interactions between platelets and certain substances (i.e., collagen, synthetic foreign surfaces) cause the release of granular material into the circulating blood; the specific release of granular material from the platelets in response to extracellular stimulation has been termed the platelet release reaction [6,7]. The term "platelet release reaction" is thus used to describe the specific extrusion to the platelet's environment of the content of platelet storage granules.

The platelet release reaction is of paramount importance in our understanding of mural thrombi on biomaterials, since the platelet granules represent individually functioning biological units capable of attracting more platelets, and also capable of initiating thrombosis. Platelets circulate in blood without adhering to each other or to normal endothelium. Their unique properties lie in changes that occur when the endothelium is broken, or when the platelets are exposed to a variety of foreign artificial surfaces.

Platelets adhere to biomaterials by means of pseudopodia; following this adhesion (the mechanisms of which are poorly understood), platelets undergo a variety of internal changes, known as the platelet release reaction, which results in their sticking to each other, a process known as platelet aggregation. This aggregation appears to be triggered by the release of adenosine diphosphate (ADP) which is present in high concentration within the platelets themselves. The ADP responsible for platelet aggregation is stored in electron-dense dark granules. ADP also causes the activation of a membrane phospholipase, which hydrolyzes membrane phospholipids to yield arachidonic acid. Arachidonic acid, in turn, is converted by a platelet enzyme called cyclooxygenase into a prostaglandin endoperoxide. This endoperoxide is then converted into a substance called thromboxane A_2, which is the most potent platelet aggregant known in medicine. Therefore platelets are self-sufficient laboratories, containing all the necessary ingredients needed for adhesion, aggregation, and the initiation of a mural thrombus.

The production of an irreversible aggregate of platelets is dependent on the platelet release reaction. ADP released from adherent platelets is sufficient to induce aggregation of a much larger number, so the release reaction may also be viewed as a biologic amplifier that rapidly converts a minimal stimulus into a massive response [8]. ADP extruded during the release reaction is only one of a number of pharmacologically active substances located within the platelet-dense bodies. Table 1 shows the contents of substances known to reside within the platelets.

To recapitulate, platelets respond to a variety of substrates, both natural and synthetic, by altering their shape, adhering to the materials to which they are exposed, and undergoing a release reaction. This release reaction involves a surprisingly rapid movement of subcellular granules to the surface of the platelet where they degranulate and contribute to the prolongation and amplification of mural thrombus.

This analysis of platelet function has emphasized its role to promote hemostasis and its participation in thrombogenesis. In addition, platelets are also important for other functional characteristics:

- Platelets and leukocytes tend to accumulate around each other; this interaction frequently leads to mural thrombosis.
- Adhesion of platelets nearly always occurs when blood contacts any artificial polymer, possibly leading to the activation of the intrinsic coagulation sequence.

Table 1. Substances Released During Platelet Release Reaction

Subcellular Localization	Constituent	Reference
Dense granules	5-Hydroxytryptamine (5-HT)	9
	Adenosine DiPhosphate (ADP)	10
	Adenosine TriPhosphate (ATP)	11
	Calcium	12
	Pyrophosphate	13
	Antiplasmin	14
α-granules	Acid hydrolases	15
	Potassium	16
	Fibrinogen	17
	Permeability factor	18
	Chemotactic factor	19
Unknown	Growth factor	20
	Bactericidal factor	21
	Platelet Factor 4	22

- Platelets recognize microscopic imperfections in the surface of "smooth" biomaterials, forming a microthrombus, which may embolize, and reform in a perpetual cycle.
- Platelet clumps form spontaneously in areas of blood stasis. These clumps are periodically carried downstream, resulting in a continuous shower of platelet microemboli.

The successful design of hemocompatible biomaterials demands detailed knowledge and understanding of platelet function and activity. Among the most important functions of platelets is their role in hemostasis; another crucial function is the participation of these elements in the coagulation sequence, particularly in the activation of the intrinsic pathway of coagulation, a subject to be discussed in detail subsequently.

VI. INTRINSIC COAGULATION SEQUENCE

We now turn our attention to blood plasma. The liquid phase of blood is composed of an aqueous medium, which normally contains a bewildering array of special substances. For the sake of simplicity, we will only discuss certain substances known to interact with implanted biomaterials, and which lead to the phenomenon of thrombosis. Of special interest are three families of proteins: the immunoglobulins, albumin, and the blood coagulation proteins. The blood coagulation proteins are a series of enzymes that work in a sequential fashion, with the final outcome being the polymerization of a blood-circulating monomer (fibrinogen) to form a crosslinked, stable biological macromolecule known as fibrin, depicted in Figure 3. Table 2 lists the individual coagulation factors according to the description established by the International Nomenclature Committee, which utilizes a roman numeral for each individual coagulation constituent. The numbers were assigned on a historical basis, with fibrinogen being the first factor purified and identified. Because of a mistake, Factor VI was assigned to a substance initially thought to be unique, but which later turned out to be merely an impure factor; since by that time Factor VII had already been discovered, it was decided to drop Factor VI from the list to avoid possible confusion. Arabic numbers 3 and 4 are assigned to mixtures of platelet membrane phospholipids, and a protein, that are known to contribute to the coagulation process; but to date the phospholipids have not been identified biochemically in a manner com-

Table 2. Nomenclature of Blood Coagulation Factors

Factor	Other Common Names
I	Fibrinogen
II	Prothrombin
III	Tissue Thromboplastin
IV	Calcium
V	Proaccelerin, Plasma Ac-globulin, Labile factor
-	Omitted
VII	Proconvertin, Stable factor, SPCA
VIII	Antihemophilic factor, AHF, antihemophilic globulin, AHG, Antihemophilic factor A
IX	Plasma thromboplastin component, PTC, Christmas factor, antihemophilic factor B
X	Stuart-Power factor
XI	Plasma thromboplastin antecedent, PTA
XII	Hageman factor, HF
XIII	Fibrin stabilizing factor, Fibrinase, Laki-Lorand factor
3	Platelet phospholipid clotting activity
4	Platelet antiheparin activity

Note: The coagulation factors are all proteins, with the exception of Factor IV (Ca^{++}). Factors VI, 1, and 2 are omitted since they were found to be impure macromolecules.

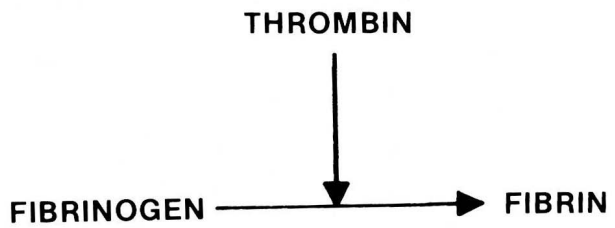


Figure 3. Formation of a fibrin meshwork by the proteolytic activity of thrombin on its substrate, fibrinogen. A fibrin meshwork is the final outcome of the coagulation cascade, and is an essential structural component of thrombus.

parable to other coagulation factors. Until they are identified in an unequivocal fashion, they are merely referred to as "phospholipids." A comprehensive glossary of known blood coagulation components is presented in Appendix B.

The coagulation of blood is mediated through a series of complex reactions involving the interaction of several enzymes, lipids, and ions. In the predominant reaction mechanism, an inactive circulating proenzyme is converted to an active proteolytic enzyme which has as its

substrate another proenzyme. This principle is illustrated in Figure 4, which depicts the formation of fibrin monomer from its precursor fibrinogen, accomplished by the proteolytic proenzyme thrombin.

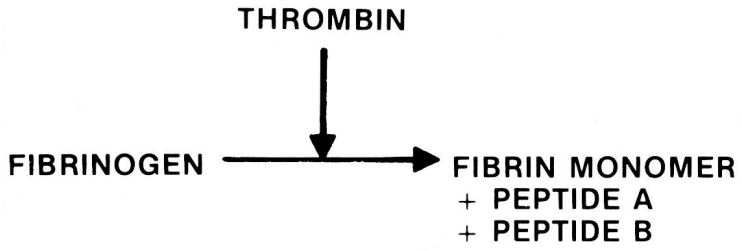


Figure 4. Formation of Fibrin Monomer by Cleavage of Peptides A and B.

Fibrinogen is a plasma protein that is synthesized in the liver parenchymal cells [23]. Its plasma concentration ranges between 200 and 400 mg/dl. Its biologic half-life is around 3 days. The fibrinogen molecule has a molecular weight of 330,000 Daltons, and consists of three pairs of disulfide-linked polypeptide chains designated $(A\alpha)_2$, $(B\beta)_2$, and γ_2 [24]. Since it circulates in dimerous form, [25] it is composed of a pair of alpha, beta, and gamma chains, shown in Figure 5. Peptides A and B represent the two fibrinopeptides cleaved from fibrinogen in the transformation of soluble fibrinogen into the spontaneously polymerizing fibrin monomer [26,27]. Covalent structure changes resulting from thrombin proteolysis reduce the molecular mass by an extremely small amount, approximately 6000 out of 340,000 mass units, but expose the polymerization sites of the fibrin monomer and permits gelation to occur.

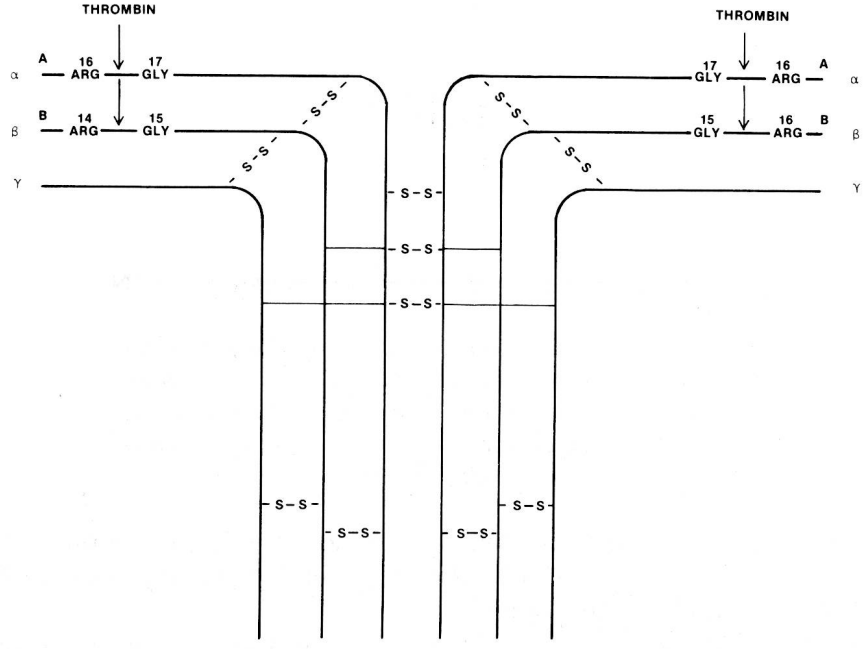


Figure 5. Molecular architecture of fibrinogen dimer. Peptides A and B are cleaved from the N-terminal portions of the α or β chain by the proteolytic action of thrombin.