

THE FORMATION AND FATE
OF THROMBI
WITH PARTICULAR EMPHASIS ON THE ROLE
OF THE PLATELET

A THESIS
SUBMITTED FOR THE DEGREE OF
DOCTOR OF MEDICINE
IN THE UNIVERSITY OF ADELAIDE
BY
N. G. ARDLIE, M. B., B. S., ADELAIDE, 1961

JANUARY, 1967.

CONTENTS

	Page
Acknowledgements	i
Declaration (of personal composition of thesis)	iii
 <u>PART I - HISTORICAL SURVEY</u>	 1-93
<u>Introduction</u>	1
 (1) <u>The vessel wall and thrombosis</u>	 3-28
Observations in the nineteenth century	3
The contribution of the electron microscope	5
Platelet adhesion to an injured vessel wall	8
Platelet aggregation	13
Mechanism of thrombin-induced aggregation	15
Mechanism of ADP-induced aggregation	16
Inhibition of ADP-induced aggregation	20
Stabilisation and growth of the platelet aggregate	22
Thrombosis and the atherosclerotic plaque	26
 (2) <u>The composition of the blood and thrombosis</u>	 28-83
A generalised change in the circulating blood	29
Blood coagulation and its relation to thrombosis	31
The concept of hypercoagulability	33
The role of lipids in the coagulation system	43
Blood lipid levels and coagulation	46
Fatty acids, coagulation and thrombosis	54
Fibrinolysis	58
Effect of plasma lipids on fibrinolysis	61
Diabetes mellitus and occlusive vascular disease	63
Dietary carbohydrate and ischaemic heart disease	65
Lipid metabolism in diabetes mellitus	68
Fibrinolytic activity in diabetes mellitus	68
A qualitative abnormality in platelets predisposing to thrombosis	69
Lipids, platelet aggregation and thrombosis	71
Adrenaline, platelet aggregation and thrombosis, and stress	75
Smoking and thrombosis	79
Diabetes mellitus, glucose and platelet aggregation	80
Post operative changes in platelet behaviour	80
The influence of red blood cells on platelet behaviour	81
Disseminated intravascular thrombosis	82
 (3) <u>Blood flow and thrombosis</u>	 83-88
<u>The fate of thrombi</u>	88-91
<u>Aims of the present study</u>	91-93

	Page
<u>PART II - THE FATE OF THROMBI</u>	94-151
<u>Experiment 1: The organisation and fate of autologous pulmonary thrombo-emboli in normocholesterolaemic rabbits, and a comparison with the fate of plasma thrombi in the anterior eye chamber</u>	94-124
1. Materials and Methods	94
2. Results	97
3. Discussion	112
<u>Experiment 2: The influence of hypercholesterolaemia on the organisation and fate of autologous pulmonary thromboemboli</u>	124-138
1. Materials and Methods	124
2. Results	125
3. Discussion	133
<u>Experiment 3: Electron microscopical observations on the organisation of artificial plasma thrombo-emboli in the pulmonary artery of the rabbit</u>	138-147
1. Materials and Methods	139
2. Results	139
3. Discussion	143
<u>Summary of Part II</u>	147
<u>PART III - STUDIES OF PLATELET AGGREGATION</u>	152-174
<u>Experiment 4: The influence of catecholamines on nucleotide-induced platelet aggregation</u>	152-158
1. Materials and Methods	153
2. Results	153
<u>Experiment 5: Inhibition and reversal of platelet aggregation by methyl xanthines</u>	159-161
1. Materials and Methods	159
2. Results	159
<u>Experiment 6: Fatty acids and in vitro platelet aggregation</u>	161-164
1. Materials and Methods	162
2. Results	162
<u>Summary of Part III</u>	165

CONTENTS (continued)

	Page
<u>Discussion of Part III</u>	166
<u>PART IV - IN VITRO THROMBOSIS AND PLATELET AGGREGATION IN VARIOUS CLINICAL CONDITIONS</u>	175-214
<u>Experiment 7: In vitro thrombosis and platelet aggregation in myocardial infarction</u>	175-189
1. Materials and Methods	175
2. Results	181
3. Discussion	185
<u>Experiment 8: Platelet behaviour and in vitro thrombosis in diabetes mellitus</u>	190-205
1. Materials and Methods	190
2. Results	193
3. Discussion	199
<u>Experiment 9: In vitro thrombosis and platelet behaviour after operation</u>	205-214
1. Materials and Methods	206
2. Results	207
3. Discussion	211
<u>PART V - SUMMARY</u>	
(including an indication of wherein the thesis is considered to have advanced medical knowledge)	215-221
<u>APPENDICES</u>	222-238
<u>REFERENCES</u>	239-310



Introduction

According to Robb-Smith (1955) the word "thrombosis" was coined by the Greek physician Claudius Galen, but not used in its modern sense until the nineteenth century. Prior to this, it was probably employed in much the same manner as the Anglo-Saxon word "clott". However, after a significant series of experiments in the five years 1882 to 1887, which clearly established the important role of the platelet in the development of a solid mass of blood in an injured vessel, the term thrombosis began to take on a more precise meaning, while the words clotting and coagulation came to be restricted to the solidification of blood under static conditions.

The word clot is now used to describe the solid material which is formed when blood solidifies under static conditions, in the body after death, or when shed from the body. This solid material is soft, shiny and gelatinous in appearance, and is dark red in colour and homogeneous, unless sedimentation has occurred before clotting is complete, when cell-rich and plasma-rich zones are formed. Microscopy shows a homogeneous mass of red cells enmeshed in a fibrin network. Discrete white cells and platelets are present in the proportions in which they normally occur in the blood and are therefore inconspicuous.

On the other hand a thrombus is "a solid mass or plug formed in the living heart or vessels from constituents of the blood". By employing this definition Welch (1889), in a monumental review of the subject,

wished to indicate the important role of blood cells, and in particular platelets, in thrombosis. Microscopically a thrombus is not homogeneous. It consists of masses of platelet aggregates encrusted with white cells and fibrin, and between these aggregates, red cells enmeshed in fibrin.

Grossly thrombi are firmer than clots, and are dry, friable and mottled in appearance, pale and dark red areas being visible.

Many thrombi, especially venous thrombi, consist of two different parts. The first part or head is formed in flowing blood by the aggregation of platelets and has a pale appearance. This either reduces or completely stops the blood flow, thereby facilitating the development of the second part or tail of the thrombus. This latter is red in colour and structurally is a blood clot.

Thus, although synonymous from an etymological point of view, we have come to distinguish between a clot and a thrombus. Regrettably this utopian view still lacks many adherents and even today there is a considerable intermingling of these two terms. The distinction has been and should be made for two reasons. Firstly there is the morphological difference. The importance of determining at necropsy whether a solidified mass of blood formed prior to, or after death is vital. For this reason alone a distinction is justified. In this circumstance it is obviously desirable to employ different terms for two solid objects whose clinical significance is so extremely different. Secondly, it has been proposed that the mode of formation is different. Subsequently evidence will be presented in this review that a thrombus might arise

together with a clot, and that a thrombus might arise from a clot.

in two different ways, depending on the presence or absence of vascular injury. The initial stages of thrombosis in an injured vessel appear to be independent of the coagulation mechanism. Alternatively, in the absence of preceding vascular injury thrombosis may represent a process of intravascular coagulation.

In 1846 Virchow outlined the principal factors influencing thrombosis. These were:

- (1) the vessel wall
- (2) the composition of the blood
- and (3) the blood flow.

Although there have been considerable recent advances in our understanding of the mechanisms of thrombosis, this classic triad still remains valid today, and thrombogenesis can best be discussed under these three headings. Some overlap in discussion must result, but nevertheless this division is convenient.