WOUND HEALING - A RE-APPRAISAL

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PART ONE

"The History of Wound Healing reflects the history of the development of Surgery", states Boyd in his textbook of Surgical Pathology. (1). Recent developments in the field of surgical research have indicated startling possibilities for developments which will necessitate the evolution of completely new concepts.

Since the subject of Wound Healing is more than adequately dealt with in textbooks, it is proposed to cover only those concepts which have arisen in the last few years, although any essay dealing with Wound Healing without a brief recall of the historical milestones would be incomplete.

HISTORICAL EVOLUTION OF WOUND HEALING

> "In the first place the lips of the wound are debrided, then the wound should be, and all about the wound should be abraded, completely cleaned of fuzz, hair and anything else, and let it be wiped dry with fine lint soaked in warm wine and wrung, and thus the lips of the wound may be reunited as well as possible in accordance with their original healthy state; and having made compresses of fine and clean lint soaked in warm wine and placed upon the wound so as to fit, let it be bound with a light bandage in such a way as reapproximation of the edges cannot be disturbed at all, and just as we have said before, do not undo the dressings until the third, fourth or fifth day if no pain occurs".

This vivid description with, perhaps, alcohol replacing the warm wine, could be as relevant today as it was when written in 1266 by Theodoric of Cervia (1205-1298), a treatment which he attributed to his father, Hugo of Lucca. (2). Such has been the development of surgical techniques that at one time this description was forgotten. Some surgeons argued that pus in the wound intimated that wound healing was undergoing its natural course, a contention strongly disputed by Theodoric.

Man's increasing search for power led to the invention and use of more and more powerful destructive weapons which presented special problems to doctors throughout the ages since the first use of gunpowder in the battle of Crecy in 1364.

Guy de Chauliac gave us a clear description of how to treat penetrating wounds of the abdomen, methods of removing foreign bodies (saying "Nature expels all that is strange to her"), and various kinds of sutures.

Some doctors poured hot oil onto gunshot wounds until that memorable day when Ambroise Pare ran out of oil and found that those wounds left untreated healed better and faster than those that had been traumatized iatrogenically. He claimed: "I dressed the wound, but God healed it".

Even the great John Hunter (1728-1793) who differentiated between healing by first intention and healing by second intention, advocated against probing and for leaving bullets in gunshot wounds since he was convinced that suppuration around the wound would extrude the bullet. Neither did he believe in debriding wounds and left blood clots in them.

Sir John Pringle coined the term "antisepsis" in 1750 and is now regarded as the father of modern day military surgery.

In the American Civil War (1861-1863) wounds were cleaned with dirty sea sponges which, when not in use, were kept in buckets.

In the 1840's Oliver Wendell Holmes and Semmelweiss published articles on the spread of puerperal infection. Pasteur and Koch's great work in bacteriology, closely linked with Lister's epoch-making treatise swept away for ever the black magic era of surgery.

In 1867 Lister published his articles on the antiseptic treatment of wounds.

In 1874 Sir John Erichsen prophesied that the abdomen, chest and brain would be forever closed to the surgeon's knife!

Halstead (1889-1922), Professor of Surgery at Johns Hopkins University, brought a new meticulousness into the handling of tissues at operation and emphasised the danger of dead space in the wound. He treated wounds by allowing a blood clot to form and suturing over the blood clot. He claimed good results and endowed the blood clot with almost magical properties.

From the battlefields of attrition of World War 1 with their mangled bodies came notably the Carrel-Dakin treatment of wounds, the use of immobilisation and the gentle surgery preached by Halstead being embodied by Moynihan's "caressing of tissues". A great breakthrough was the pioneering of plastic surgery. The use of antisepsis was not favoured by the majority of surgeons who instead minimized wound infection by debridement.

Table of Pathological Changes

EPITHELIUM		DERMS & S/Cut. Fat		DAYS
Intraincisional	Peri-incisional	Intraincisional	Peri-incisional	
I: LAG PHASE (Catabolic or Proliferative Phase)				
EPITHELIALIZATION		INFLAMMATORY RESPONSE		
1. Two thickening, advancing epithelial sheets.	1. Hyperplasia. No. of mitotic figures increases	**A. Acute or Exudative Phase**		
(after 12 hours)	2. Epithelium and rete pegs thicken.	1. Small round cells accumulate in injured dermis	1. Vasodilatation 2. Diapedesis of r. b. c.'s and polymorphs	1
2. These unite within 24 hours.				
3. Invasion by primary "spur" into line of incision.				
A. Invasion along incisional line due to:			1. Mononuclears appear to stream into wound space	2
a) Phagocytosis by epithelial cells b) Proteolytic enzymes released c) Collagenase		1. Vertically orientated strands of fibrin appear	2. Mitotic figures now abundant	3
B. Short secondary epithelial "spurs" may invade laterally		**B. Proliferative Phase**		
		1. Perivascular cuffing with small round cells	1. Perivascular cuffing	
		2. Collagen precursors (reticulin) seen	2. Capillary sprouts	4
II: FIBROBLASTIC PHASE (Anabolic Phase)		3. Mitotic figures noticeable in PHASE OF COLLAGENIZATION		5
		4. Fibroblasts seen orientated vertically		
		PHASE OF COLLAGENIZATION		
		Tensile strength of wound increase rapidly.		6
		Perpendicularly		7
		arranged collagen		8
		fibres seen.		9
		Progressive Collagenization.		10
		New elastic fibres	Cellularity of	11
		fine, wavy and	loose connective	12
		parallel to	tissue diminishes	13
		surface.		14
III: PHASE OF CICATRIZATION AND DIFFERENTIATION				
			Pseudo rete pegs removed by connective tissue	15
			foreign body reactions.	16
		Scar may widen	Devascularization and no. of new cells diminishes	

Zones of Repair occur in peri-incisional space

In 1944 the first use of penicillin revolutionized the whole field of management of wounds and infections, heralding the antibiotic era with its attendant dangers.

In World War II mortality from penetrating wounds of the abdomen and chest decreased dramatically due to quicker action, better surgery, the use of blood transfusions and the introduction of sulphonamides and later penicillin.

PATHOLOGY OF WOUND HEALING

Studies of wound healing have disclosed the need for investigation in **four** dimensions: length, breadth, depth and time.

The pathological changes that occur in healing of incised wounds are summarised in the table. Diagrammatic representations of the main features are shown in Figures 1 and 2.

Since the factors governing the various stages are complex and much work has been and is being carried out to determine them, it is proposed only to venture briefly into some important explanations that exist at the present time.

Gillman, in his magnificent treatise published in the Archives of Surgery 1966, states that "certain basic problems concerning repair in general still remain unresolved" and "Failure to resolve these problems is in large measure due to the fact that the nineteenth century descriptions of the healing of cutaneous wounds seem to persist unquestioned in the literature". He stresses the need to consider the healing of wounds three-dimensionally in order to understand the histogenesis of complications associated with the repair of a simple incision, eg. epidermoid cysts and abcesses. "The empirically derived surgical conviction of the importance of meticulous haemostasis or effective drainage of blood to promote rapid uncomplicated healing is contradicted by the pathologist's insistance on the role of fibrin as a scaffold permitting fibroblast invasion into incisions".

Gillman forcibly pointed out that the first tissue link between the two sides of the wound is epithelium and not connective tissue. He stressed that the processes of repair in the healing of incised and excised cutaneous wounds are fundamentally similar. Blood clots play no role in repair other than in promoting haemostasis, acting transiently as a wound glue, and serving as a wound dressing. Evidence indicates that the intraincisional blood clots usually retard healing for as long as it takes the macrophages to remove the blood from the site of the injury.

Gillman goes on to attack the traditional suturing method and talks of the Suture Needle Puncture Wound (S.N.P.W.) and Suture Track (S.T.) which cause the interdermal injuries. A 6" long incised wound with the edges coapted with strips has a volume of 8 cu. mm. By contrast, a 6" long wound sutured conventionally becomes converted into an excised wound with a volume of 88 cu. mm.

HOW DOES THE INITIAL PROCESS OF EPITHELIALIZATION TAKE PLACE?

Epithelialization is the process by which the surface of a wound is restored and approaches more closely the ideal of regeneration — *restituto ad integrum* — than does any other tissue; as opposed to the process of repair, which is a process of healing in which a new structure — a scar — is formed.

Medawar has pointed out that cell movement is fundamentally important in regeneration and in embryonic development. (4). Some authors claim that the epithelial cells move by amoeboid movement of the advancing edge of skin. It appears more likely that (5) the advancing cells are elongated and overlap one another. The cells then seem to roll over one another and are successfully implanted on the wound surface, — roughly analgous to the movement of a caterpillar track.

Individual vertebrate cells of almost all kinds have the ability to crawl along any solid substance to which they can adhere to an appropriate degree at the rate of 50-100*u* an hour (a yard a year) compared to a white blood cell which can move ten times as fast.

In 1915 Rand stated "Epithelium will not tolerate a free edge". Friedenwald and Buschke in 1944 observed cell migration in the cornea. Abercrombie in 1966 considers that the processes of mitosis and mobilization are promoted by products of cell damage and death, and keeps in mind the "old-fashioned" idea of a Wound Hormone. (6).

What is clear is that the epithelial cells that are "on the march" are capable of cutting through collagen and blood clot, due partly to their power of phagocytosis (7) and partly to the proteolytic enzymes that are released at the moving edge.

Professor Debono (8) wrote in 1949 "It appears further that the processes of regeneration have a priority over the maintenance of the body. Thus the salamander will reform its tail even if starved of food by utilising its own body tissues. In this way also may be explained the rapid wasting in cases of extensive wounds. Further, in Malta, during the Siege, the healing of wounds occurred normally even though the diet was very near the indispensable minimum".

There is much evidence that dedifferentia-

FIGURE 1

Cross Section through an incised wound :-

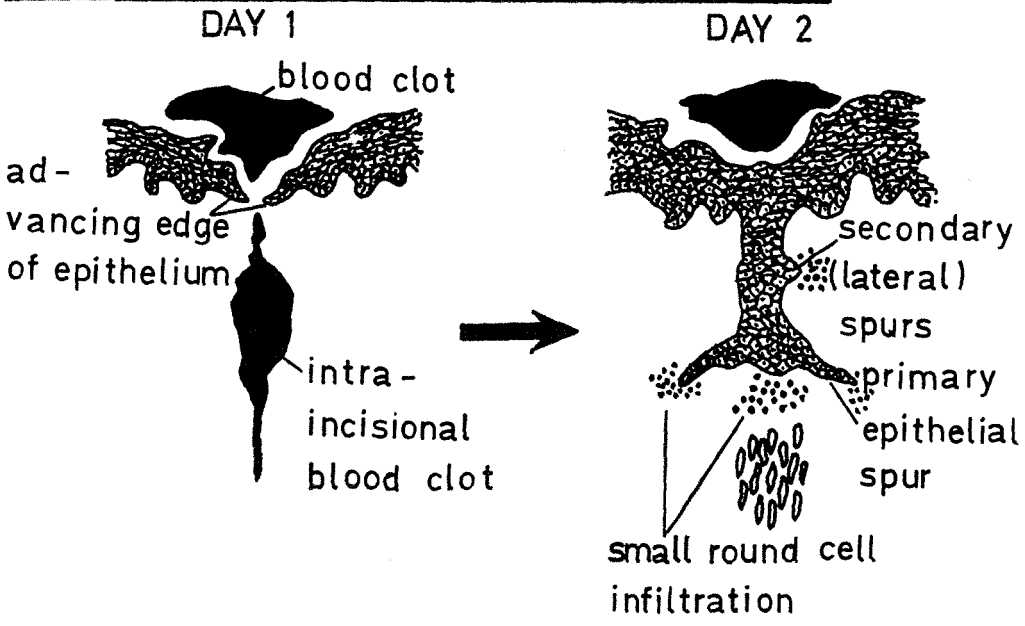


FIGURE 2

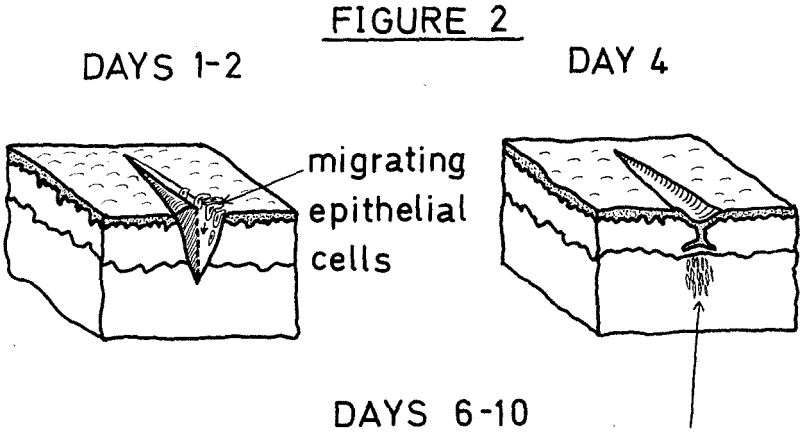
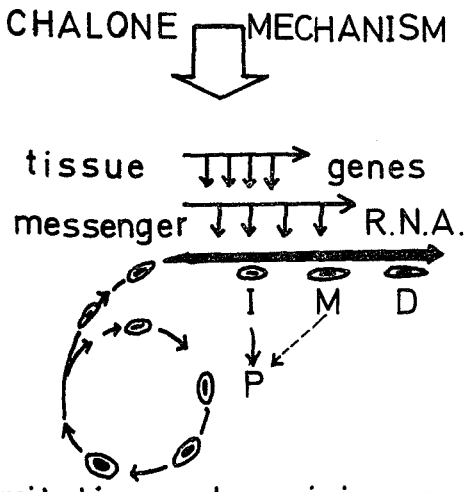


FIGURE 3

A Representation of life of tissue cell :-



mitotic cycle : minimum duration of 12 hours

B: Control mechanism of cell :-

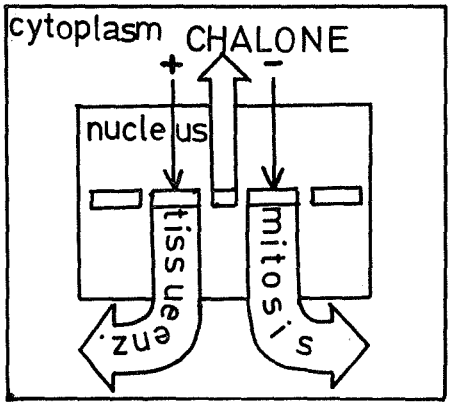
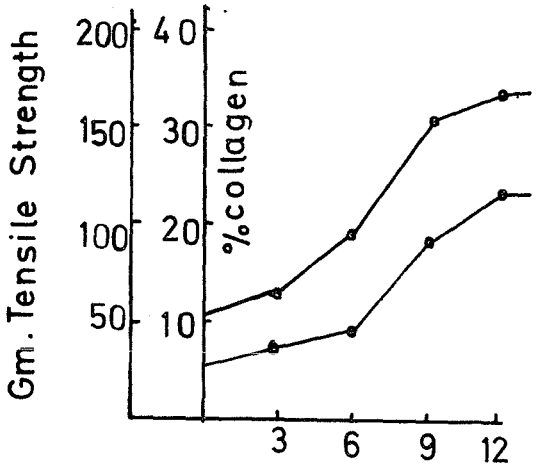
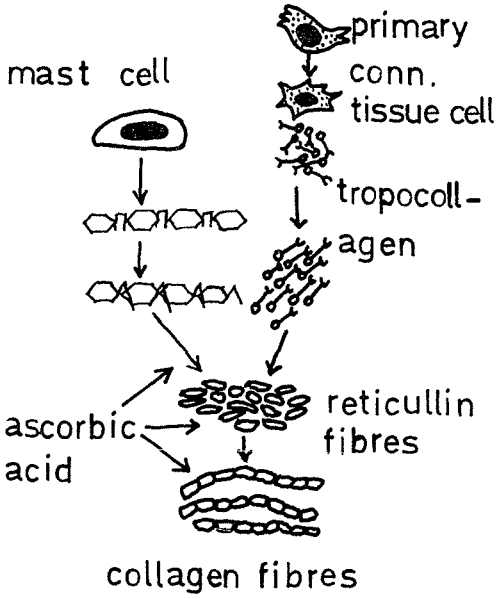


FIGURE 4

A: Correlation between Tensile Stength and Collagen content of incised wounds :-



B: Theory of collagen formation :-



tion to a more embryonic state is a general prelude to successful regeneration. Young individuals regenerate most rapidly and the power of regeneration declines with age. It is known that regeneration competes with sexual reproduction and it is often poor during the breeding seasons, even in animals which regenerate well at other times.

From such and other observations emerged a theory which has widespread implications. No essay on wound healing would be complete without its delineation.

THE CHALONE THEORY OF BULLOUGH (9)

Immediately after injury mitosis is seen first in those cells which have remained static on the original edges of the wound; it is rarely seen in cells that are actually migrating, and continues for some time after the wound has been closed and all movement has ceased. Bullough stresses that any one living cell at any one time may indulge in only one of three processes, i.e. movement, mitosis or manufacture of specialised cell products. Those cells involved in mitotic cycles he labels P cells (Progenitor cells), those preparing for tissue function he calls I cells (Immature), those potentially or actually functional (M cells), and the D cells those which are functional but which are moving irreversibly towards death. (See Figure 3).

Cells in the groups I and M can, when necessary, revert to the mitotically active state whereas cells of group D (eg. mature R.B.C., granulocytes and cells of epidermal stratum granulosum) cannot.

The range extends from duodenal mucosa where the I cells predominate to the epidermis where D cells predominate.

It now appears probable that within the epidermis the functionally active cells produce some substance which acts in the manner of a negative feed-back mechanism to suppress mitotic activity selectively in epidermis. Such a substance is called chalone. A high chalone concentration results in inactivation of those genes on which mitotic actively depends.

In cases of simple wounds the highest rate of mitotic activity occurs in a narrow zone about 0.3 mm. wide immediately adjacent to the edges of the wound at a rate 20 times that of mitotic activity in normal tissue at sleep (10). The length of the normal mitotic cycle of P cells is reduced to a minimum of 12 hours. I cells revert to the P form as may some M cells, but never D cells. Bullough says that the local mitotic reactions are more reasonably explained in terms of local loss of mitotic inhibitor synthesised in normal tissue than by the old theory of a Wound Hormone.

When varying degrees of liver are destroyed (from 10% to 70%) the strength of the mitotic reaction is in direct proportion to the amount of tissue lost. When one kidney is removed cleanly with no damage, the other kidney grows to double the size. This can be explained by depletion by half of the total body kidney chalone. During starvation and also during shock, mitotic activity around a wound may be powerfully suppressed, thus the avoidance of these two factors leads to better wound healing.

CHALONES AND CARCINOGENSIS

Since lack of mitotic control is a distinctive feature of tumours, it has been suggested that they may fail to synthesise or release an adequate concentration of tissue specific chalones (11).

INFLAMMATORY RESPONSES IN WOUND HEALING

"The morbid process designated by the term Inflammation, being one by which every organ and probably every tissue of the body is liable, possesses a deeper interest for the physician or surgeon than any other material subject which could be named... It is upon the **first** deviations from health that the essential character of the morbid state will be most unequivocally stamped and it is therefore to the early changes of Inflammation that attention must be chiefly directed" (12). This view expressed by Lister 111 years ago is as pertinent today as it was then.

The Inflammatory Response in Wound Healing may be divided into 2 phases (see Table of Pathological Changes).

A. THE ACUTE OR EXUDATIVE PHASE

For the first two or three days after injury this exudative inflammation is devoid of mitotic activity and is characterized by the exudation and emigration into the wound of firstly plasma proteins and mucoproteins, then of polymorphonuclear cells, and later by small round cells, all derived from the circulating blood.

Pappenheimer (13) estimated that the amount and composition of normal transudate could be accounted for by uniform porosity of the vessel walls. Landis proposed the name Fenestellae for the holes that vere postulated to exist mostly in the venular ends of the capillaries and in venules. Membrane porosity is thought to increase after injury. The mediators of this inflammatory response are not as yet identified completely.

Monocytes arriving at the scene are able to transform themselves into histocytes or macrophages. The appearance of kininogenases and

kinins is indisputable, but whether they have initiated the phase of exudation, or have been formed as the result of the exudation is still a matter of speculation.

B. THE PROLIFERATIVE PHASE

After the third or fourth day the Inflammatory response changes its character. This change is heralded by the enlargement and increasing basophilia of the small round cells and possibly of neighbouring resident fibroblasts. These changes invariably lead to some degree of fibrosis. Capillary endothelium thickens and forms buds which become canalized and transformed into capillary loops.

PHASE OF COLLAGENIZATION

Among the most important unsolved problems of Wound Healing is the origin of the fibroblastic cells population, and its ultimate fate. Twenty years ago (8) it was thought that the fibroblasts arose from fixed tissue cells in the area of the wound by returning to embryonic state and then beginning to proliferate. It is now generally thought that the fibroblasts are derived from the circulating blood. (3). The origins of the collagen fibres have been summarized by Dunphy (14). There is no doubt that with increase in collagen deposition the tensile strength (load per unit area at the point of disruption) of the wound increases and is especially marked on the 6th and 7th days after injury. (See Figure 4).

After day 20 there is a decrease in the number of new cells seen in the incision. After day 30 the number of new vessels diminishes.

Douglas, Professor of Surgery in Dundee, records in his paper of 1969 (16) how young collagen matures and the wound continues to become stronger. Human skin has very great tensile strength which varies from 20 — 44 Kg., whereas wounded skin is very much weaker, varying from 8 — 80% of intact skin.

There appear to be two main points of differences between Dermal Collagen and Wound Collagen:—

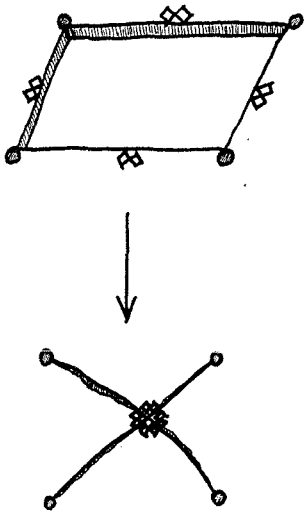
1. Dermal Collagen is arranged in much larger bundles than Wound Collagen.

2. Dermal Collagen bundles are birefringent (refractile under polarized light) whereas Wound Collagen is not, or only weakly so. After 52 days from injury there is no noticeable difference between the two. Hence, since at 50 days the wounds are still much weaker than intact skin, other factors such as orientation and arrangement of the fibres may be involved.

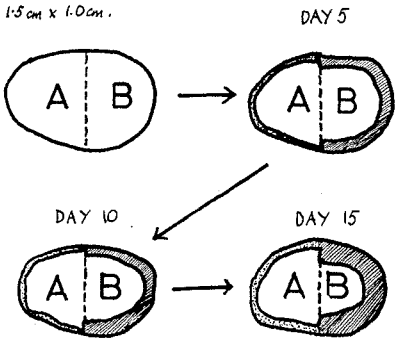
Douglas states that in man restitution to normal does not occur after wounding — a

FIGURE 5

A Rectangular wound :-



B Effect of removing scab :-



Observations 1. Side B had scab removed each day as it formed : A left intact. At present time scar is still faintly visible : measuring 1·3 cm x 0·9 cm.

2. Rectangularly shaped wounds have a greater power of contraction than do circularly shaped wounds

scar on the face is detectable for the rest of an individual's life —. A fracture of a long bone is permanently discernable on radiological examination, and repeated injury to the brain in professional boxers is associated with notable changes in intellectual performance.

MECHANISM OF WOUND CONTRACTION

The repair of full thickness excised wounds of the skin is not accomplished simply by the production of a scar whose dimensions are those of the original deficit. Instead, the intact skin bordering the wound is drawn centripetally with the consequence that the eventual scar area is considerably less than that of the wound. (See Figure5). The mechanism of wound contraction remains yet another unsolved problem.

Abercrombie (17) believes that the cells that have recolonised the wound (fibroblasts) have a motive power.

REFERENCES

1. Boy, W. *Textbook of Surgical Pathology 1955*
2. Meade, R.H. *An Introduction to the History of General Surgery. Saunders, W.B. & Co. 1968*
3. Ordman, L.J. & Gillman, T. *Archives of Surgery Dec. 1966*
4. Medawar, P. *Brit. Med. Bulletin 1945, 3:70*
5. Winter, G. *Advances in Biology of Skin 1964 p. 113*
6. Abercrombie, M.H. & James, D.W. *Wound Healing Pergamon Press 1961*
7. Platt, *J. Path. and Bact. 1963, 86: 113*
8. Debono, P.P. *Chestpiece 1949 Vol. 1 No. 3*
9. Bullough, W.S. *Lister Centenary Symposium, Wound Healing.* 1966 p. 43
10. Epstein, W.L. and Sullivan, D.J. *Ad. Bio. of Skin 1964*
11. Leading Article *Lancet 1968 Nov. 23rd.*
12. Lister, J. *The Early Stages of Inflamation* 1868
13. Pappenheimer et al. *Am. J. of Physiol. 1951 167, 13*
14. Dunphy, J.E. *Ann. of Roy. Coll. of Surg. 1960, 26, 29*
15. Howes et al. *J.A.M.A. 1929, 92, 42*
16. Douglas, D.M. *Brit. J. Surg. 1969 Vol. 56 No. 3*
17. Abercrombie, M.H. et al *J. Anat. 1960, 94. 170.*

Part two will be included in the next issue of Chestpiece.