ATHEROSCLEROTIC METAMORPHOSIS OF AUTOLOGOUS PULMONARY THROMBOEMBOLI IN THE RABBIT

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Since the recent revival of Rokitansky's concept[1] of the thrombotic origin of atherosclerosis,[2-4] many investigators[5-8] have attempted the experimental induction of atherosclerotic plaques from emboli composed of different blood elements. These studies have been concerned with the injection of fibrin clots or whole blood clots of various types into the ear veins of rabbits and the observation of their subsequent degeneration and organization within pulmonary arteries. The experiments have proved the capacity of arteries to incorporate intimal deposits within themselves; however, two critical features of these investigations require consideration. The lesions thus produced were mainly fibrous intimal thickenings which lacked the proportions of lipids, foam cells, cholesterol clefts, calcium, and collagen present in natural atherosclerosis. Secondly, the emboli used for injection were clots, not thrombi; these are two entirely different entities.[9]

Clots and thrombi differ in structure and in the dynamics of formation. Perhaps the only significant feature they share is that both represent nonliquid states of the blood. A thrombus is built principally from platelets in a flowing stream of blood.[10,11] Certainly this dynamic mechanism finds no parallel in the coagulation of stagnant blood or plasma in a test tube. In order to appreciate the lack of structural resemblance between a thrombus and a clot, one should compare the haphazard entanglement of erythrocytes, leukocytes and platelets in a fibrin clot (Fig. 1) with the fine, orderly arrangement of a thrombus (Figs. 2 to 4). The structure of the latter, with its coralline platelet columns rimmed by borders of fibrin, was described in lucid detail by Aschoff.[10] Yet, although many 19th century investigators[12-15] recognized thrombosis and blood clotting as separate phenomena, there is now a widespread practice of using the words "clot" and "thrombus" interchangeably and attributing the features of one to the other.[16]

These considerations prompted the thought that it would be logical

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to inject true thrombi into the pulmonary arteries of rabbits to determine whether or not the lesions produced would resemble natural atherosclerosis more closely than those which follow the injection of clots. The recent development of a simple method for the production of thrombi *in vitro*[17,18] provided a technique for the manufacture of autologous thrombi. Thrombi are produced *in vitro* in a flowing column of blood and have a striking similarity to natural intravascular thrombi (Figs. 2 to 4), even under the scrutiny of electron microscopy.[19]

In principle the experiment described in detail below consisted of introducing autologous thrombi into the pulmonary arteries of rabbits and observing their organization and degeneration at serial intervals.

METHOD

Forty-eight young, full-grown rabbits of both sexes, weighing 2 to 4 kg. and fed regular Purina Rabbit Chow and water, were used.

Each rabbit was anesthetized by intravenous sodium pentothal and subcutaneous infiltration of the neck with procaine. Under aseptic conditions a phlebotomy of the external jugular vein was made. A 50 cm. polyethylene tube (Intra-Medic®, Clay-Adams; internal diameter, 0.125 inch; external diameter, 0.157 inch) was inserted cephalad into the incised vein. Blood (5 ml.) was allowed to flow into the tube under the control of ligatures placed loosely about the vein. The tube was then withdrawn, and from it, 1 ml. portions of blood were run into each of 3 or 4 polyethylene tubes (20 cm. long) filling about half of each tube. The ends of each tube were joined by plastic collars to form closed circles (Text-fig. 1). Each circle of tubing was then rotated on a turntable inclined at 45°, causing the blood to flow round the tube as it revolved. After 5 to 7 minutes a cylindric thrombus with an attached tail of coagulated blood formed at the advancing edge of the column of blood. Each thrombus measured approximately 6 by 3 mm.

In every instance one thrombus was fixed in buffered formalin and examined histologically as a control. The remaining 2 or 3 thrombi, from which the tails of coagulum had been removed, were quickly suspended in physiologic saline and injected via the jugular vein toward the right side of the heart and ultimately into the pulmonary arteries. The jugular vein was ligated after this procedure.

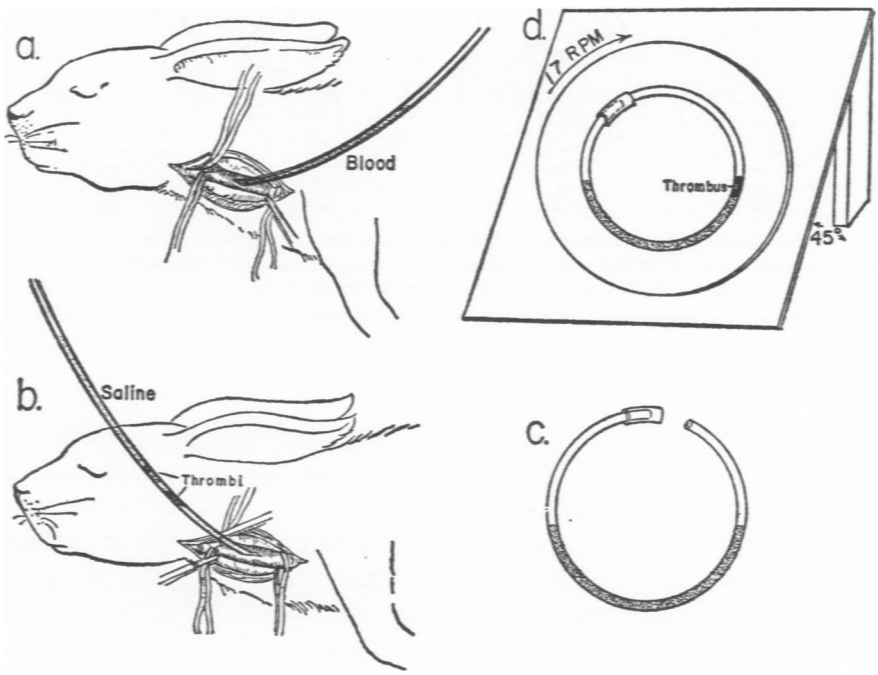
In order to obtain greater use of the animals, each rabbit underwent a second venotomy, in the opposite jugular vein, 5 to 7 days after the first. Shorter intervals were avoided to prevent difficulty in distinguishing between the thromboemboli of different ages in the same animal.

The rabbits were killed at intervals after operation by the intravenous injection of sodium pentothal. The times of sacrifice were calculated to provide for the examination of emboli of different ages, from 1½ hours to 12 months after introduction.

The lungs were removed with the heart *en bloc* and fixed in buffered formalin for two hours. Each lung was then removed from the block and the individual lobes separated from each other at the hilum. Each lobe was cross-sectioned, beginning at the hilum and progressing peripherally, the slices being approximately 2.5 mm. apart. Every cut surface was examined with a dissecting microscope; all tissues containing thrombi or plaques were embedded after fixation in buffered formalin for an additional 24 hours. The superior vena cava, the jugular veins, and the heart were also dissected and scrutinized for the presence of thrombi. The blocks of lung were serially sectioned and mounted on large lantern slides. The following staining methods were used: hematoxylin and eosin, phosphotungstic acid hematoxylin, Sudan IV, resorcin-fuchsin, Prussian blue, von Kossa, and Gomori trichrome.

Results

Ninety-two thromboemboli were identified within the pulmonary arteries. Since two or more thrombi occasionally became stuck together during injection, all thrombi were not separately identifiable at the time of dissection. However, at least one thrombus was found in the lungs of each rabbit at necropsy.



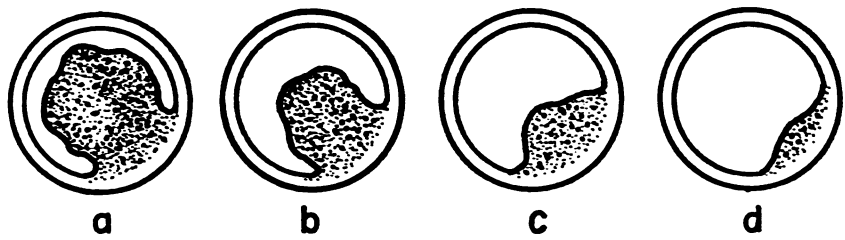
Text-figure 1. Schematic representation of technique. (a) Blood is withdrawn via a tube inserted cephalad into the jugular vein. (c and d) A 1 ml. sample of blood is put into a length of tubing, the ends of which are joined with a plastic collar to form a circle, which is then rotated on an inclined turntable until a thrombus forms at the leading edge. (b) The thrombi are suspended in saline and injected into the jugular vein toward the right side of the heart.

Gross Appearance During the First 3 Weeks

For the first 3 days the emboli retained the appearance of fresh white thrombi. By the third day marginal channels filled with fresh blood had been formed by retraction of the emboli (Fig. 5). Thrombi examined after 5 or 6 days were exceptionally friable as a result of a loss of fibrin binding. By the end of the second week further organization had made the thrombi quite firm. In the subsequent 4 weeks the emboli underwent further shrinkage, thereby assuming eccentric positions on the arterial walls (Fig. 7). In most cases the sites of mural attachment

were broad (approximately one fourth of the circumference), and the shrinking thrombi gradually assumed the appearance of eccentric mural plaques (Fig. 13).

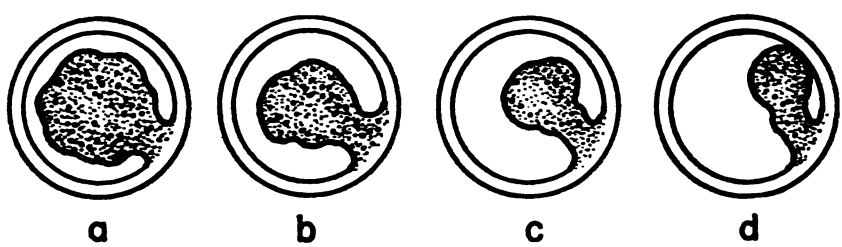
In other instances the sites of attachment after initial retraction were smaller (approximately one eighth of the circumference), and these became longitudinal ridges which appeared polypoid in cross section (Fig.



TEXT-FIGURE 2. The sequence of retraction of a thromboembolus with a broad base of attachment. (a) Two days after embolization, ¾ of the circumference of the thrombus has retracted from the arterial wall. The embolus is partially covered by endothelium at this stage. (b) After 6 days the embolus is smaller and completely endothelialized (see Fig. 7). (c) Further shrinkage flattens the thrombus against the arterial wall. (d) At 3 weeks the thromboembolus is a fibrofatty atherosclerotic plaque.

8). Such polypoid ridges fell to one side against the adjacent wall and were incorporated within it, thus also forming eccentric mural plaques during the second to fourth weeks.

Changes in Color. Alterations in the color of the emboli at various intervals correlated well with histologic features. Control thrombi were white, with circumferential margins of pink blood clot. This was also the appearance of emboli up to 3 days after introduction into the pul-



TEXT-FIGURE 3. The sequence of retraction of a thromboembolus with a narrow base of attachment. (a) Two days after embolization only approximately ⅛ of the circumference of the thrombus remains attached. (b) Seven days after embolization, shrinkage gives the embolus a polypoid appearance in cross section. (c) After two weeks the weak pedicle has allowed the thromboembolus to fall to one side (see Fig. 8). (d) The embolus becomes incorporated into the wall, resembling the plaque depicted in Text-figure 2.

monary arteries. Thereafter they became golden tan for 6 more days as hemosiderin was derived from degenerating erythrocytes. Finally a butter yellow hue appeared and deepened as the thrombus became an

eccentric plaque. This color reflected the presence of great numbers of lipophages within the plaque.

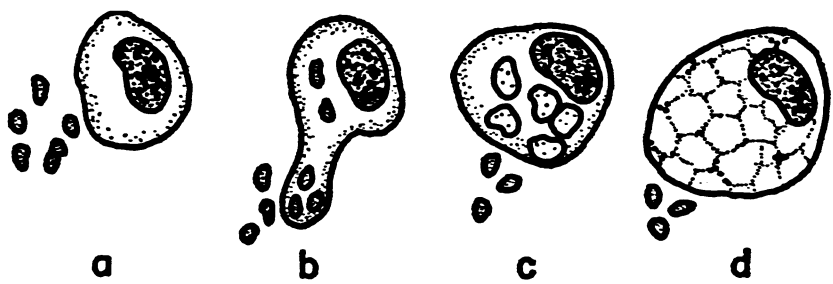
Histologic Features During the First 3 Weeks

For the first 24 hours the embolus maintained a histologic appearance not unlike that of control thrombi. During the second and third days, however, evidence of degeneration became prominent (Fig. 6). Granulocytes underwent lysis and were reduced to deposits of nuclear debris; erythrocytes were also beginning to disintegrate, but many remained intact for a week. The sparse fibrin strands underwent lysis, thus reducing the bulk of the thrombus and accounting in part for its retraction from the arterial wall (Fig. 7). By the third day the endothelium beneath the attachment of the thrombus had already become flattened and involuted and was no longer identifiable; but the mural endothelium at the attachment margins had acquired abundant basophilic cytoplasm and formed a syncytium which grew out upon the thrombus. On the fourth day the endothelium had completely covered the lumen surface of the shrinking thrombus, incorporating it into the wall of the artery. Inflammatory response to the presence of the thrombus was manifested on the fourth and fifth days by marked adventitial edema, engorgement of vasa vasorum and adventitial capillaries and by scattered granulocytes in the intima beneath the mural endothelium.

On the fifth day fibrinolysis had progressed to such a degree that the spaces left in its absence gave the thrombus a distinctly spongy appearance. Further endothelial proliferation was indicated by numerous mitotic figures, with strands of cells continuous with the surface endothelium insinuating deeply among the platelet columns (Fig. 8). Some of these invading cells formed small rosettes in which recanalized lumens appeared, but most became isolated fusiform cells, clearly identifiable as fibrocytes on the fifth day. At the end of two weeks abundant collagen was demonstrable within the thrombus.

Monocytes and Lipophages. A conspicuous feature in the metamorphosis of thromboemboli was the phagocytic activity of monocytes subsequent to their failure to undergo lysis. As mentioned above, the granulocytes disintegrated and were identifiable only as nuclear debris by the second and third post-embolic days. The monocytes, however, did not diminish in numbers, but rather increased by amitotic division and were scattered liberally throughout the thrombic mass, many cells having 2 and 3 nuclei. Cytologic changes in these were appreciable on the third day: their nuclei became hyperchromatic, and the cytoplasm increased to approximately 3 times its former volume. On the third and fourth days the monocytes actively phagocytized the debris of fragmented granulocytes and erythrocytes (Fig. 6).

On the fifth day monocytes began to engulf platelets (Fig. 9) from the loose margins of platelet columns where platelets were discrete membrane-bound entities. (Iseri and Benditt[19] investigated natural thrombi *in vivo* and *in vitro* with the electron microscope and reported that platelets were intact within both types of thrombi.) Within phagocytic monocytes the lipid-rich platelets became greatly swollen and approached the size of erythrocytes, from which they were nevertheless distinct by virtue of recognizable dense granules within them and by their lack of the porcelain-like homogeneity of erythrocytes (Fig. 10). On the eighth day, as the platelets underwent dissolution and were digested by monocytes, they became cytoplasmic vacuoles of stainable lipid within these cells, now identifiable as lipophages (Fig. 11). Some of the lipophages contained birefringent material which gave the characteristic pattern (Maltese cross) considered specific for cholesterol and phospholipids.[20] The relatively high neutral fat, lipoprotein, phospholipid and cholesterol content of blood platelets apparently had provided the substrate from which these vacuoles were derived (Table I).



TEXT-FIGURE 4. Phagocytosis of intact platelets by monocytes, which become lipophages. (a) Monocytes are present at the margins of the platelet columns during the first 6 days. (b) After the fifth day enlarged monocytes (macrophages) engulf these platelets. (c) Within the cytoplasm the platelets become swollen during the second week after embolization. (d) After digestion by the macrophages, the platelets become fat vacuoles, justifying the name lipophage for these phagocytic cells.

Later Changes: Gross and Histologic Features

At the end of 2 weeks, foam cells were closely packed together and were the most conspicuous cells in the changing thromboembolus. On the endothelialized surface of the embolus and also at its mural attachment, fibrocytes laid down coarse stratums of collagen. Contraction of these fibrous elements probably occurred and contributed to further shrinkage of the thrombic mass. At 3 weeks the thrombus had shrunken to an eccentric position and was grossly recognizable as an atherosclerotic plaque by its configuration and by its butter yellow color (Fig. 13). Beneath the intimal structure the muscular wall of the artery was abnormally thin and atrophic (Fig. 14). Histologically the plaque exhibited

all the features normally associated with atherosclerotic plaques (Fig. 14) except cholesterol clefts. Another interesting finding at 3 weeks was the deposition of calcium salts (stained by the von Kossa technique) among masses of degenerated platelets which had remained unphagocytized (Fig. 12).

After 3 weeks alterations occurred very slowly in the plaque and consisted largely of increased fibroplasia, further deposition of calcium salts and further reduction in the size of the thromboembolic plaque. Two plaques were identified at 2 months; both were small fibrous intimal plaques with a thin stratum of foam cells but no cholesterol clefts.

Four animals, into which a total of 19 thrombi had been injected, were sacrificed 12 months after embolization, and their pulmonary arteries examined for thromboemboli and plaques. Five thin mural plaques were identified at secondary pulmonary arterial bifurcations. These lacked the butter yellow hue seen in plaques developing at 3 weeks to 2 months. Instead they were fibrous, crescentic thickenings of the arterial wall. In serial sections of these, there were numerous lipophages containing histochemically demonstrable fat and hemosiderin. In two plaques cholesterol clefts were thought to be present; this could not be proved since the tissue had been processed in alcohol for paraffin embedding (Fig. 15). Calcification of unphagocytized platelets was still conspicuous; in several areas this was of such advanced degree as to constitute bony metaplasia (Fig. 15).

Additional Observations and Comment

Infarction. Only 3 hemorrhagic pulmonary infarcts were found. In all but one of these there was evidence that the affected animal had been in some degree of congestive failure. In one, the rabbit had been anesthetized too deeply during operation; there had been subsequent signs of pulmonary edema and a difficult post-operative recovery. Another developed an infarct following the second surgical procedure, the previous injection of 3 large thrombi having no doubt been a significant impairment to the pulmonary circulation. In a third case, although only one operative procedure had been done, an embolus completely occluded the principal artery to the upper lobe of the right lung, thus causing infarction of the entire lobe. These findings confirmed the observations by Karsner and Ash[21] who contended that pulmonary infarction did not occur in the absence of passive pulmonary congestion, except in the case of complete obstruction of a lobar pulmonary radicle.

Recanalization. Five thrombi which had neither retracted nor become plaques or longitudinal ridges were found, instead, to remain occlusive and to be extensively recanalized. Lipophages were abundant in all recanalized thrombi.

Recanalization seemed to be related to the site of embolization. Examination with the dissecting microscope and of serial histologic preparations indicated that emboli which became plaques had lodged at secondary bifurcations of pulmonary arteries. Others, which had undergone extensive recanalization rather than retraction, were not at bifurcations but had lodged tightly where the diminishing arterial lumen was of insufficient caliber to permit passage. It is reasonable to consider that the thrombi which had lodged at bifurcations, forming "saddle emboli," were not necessarily larger in diameter than either of the two secondary arterial branches. They would not, therefore, have required much, if any, shrinkage to provide a free lumen surface for endothelialization. On the other hand, those thrombi which had become tightly packed into arteries too narrow to permit passage became organized and adherent to the arterial wall throughout its inner circumference before sufficient shrinkage could occur to allow retraction.

Discussion

Blood Platelets in Thrombosis and Atherosclerosis

One of the earliest descriptions of blood platelets was published by Donné[22] in 1842. Their existence was confirmed by the classic work of Bizzozero,[12] who also established their importance in thrombosis. Further experimental evidence indicating the participation of platelets in thrombosis was contributed by Eberth and Schimmelbusch,[13,14] who induced thrombosis in the mesenteric vessels of dogs, and by Osler,[15] who investigated thrombi made by incising the femoral arteries of dogs. Welch[23] and other pathologists[24,25] emphasized, as many others do today,[9,10,11,16,26,27] the importance of platelets in the pathogenesis and histologic composition of thrombi.

Rokitansky,[1] writing long before the publication of Bizzozero's original observations,[12] introduced the concept that atherosclerosis is derived from intimal deposits by the circulating blood. This concept was revived by Duguid[4] and others[28–31] who, like Rokitansky, have stressed the role of fibrin; they have not, however, discussed platelets as a histologic feature or as a major source of lipids in atherosclerotic plaques. Much of the current and historical information concerning thrombosis and atherosclerosis was reviewed in 1956 in Morgan's extensive monograph,[32] but reference to "platelet" or "thrombocyte" does not occur in this volume although several illustrations depict formations closely resembling platelet columns (see Figures 76 and 101 in Morgan's monograph[32]).

Duguid's observations stimulated several experimental efforts to confirm the thrombogenic hypothesis. In 1948 Harrison[5] injected washed

human fibrin clots into the ear veins of rabbits, producing fibrous thickenings in their pulmonary arteries. This approach was modified in 1952, by Heard,[6] who produced similar lesions by injecting fragmented autologous "worm clots" into the veins of rabbits. The same basic approach has been employed by Barnard[7] and by Thomas, O'Neal and Kyu.[8] In each of these experiments the authors' observations have illustrated the incorporation of fibrinous material into arterial walls and its organization into fibrous mural plaques. But these plaques have lacked the proportion of lipid, cholesterol clefts and calcification which are found in natural atherosclerosis. Since fibrin is a protein and contains no lipid, it could not account for the fat in thrombi or in atherosclerotic plaques. The blood platelets, on the other hand, are rich in lipid content, including cholesterol.[33-36]

TABLE I

THE LIPIDS OF HUMAN ERYTHROCYTES AND PLATELETS EXPRESSED AS A PERCENTAGE OF DRY WEIGHT[34,35]

Fraction	Per cent of dry weight		
	Erythrocytes (Barkhan *et al.*[35])	Platelets (Barkhan *et al.*[35])	Platelets (Maupin[34])
Total lipids	1.26	17.0	19.0
A. Phospholipids	0.85	13.3	13.8
B. Nonphospholipids	0.35	4.2	5.2
Cholesterol (free)			3.2
Cholesterol (esterified)			0.7

The Origin and Development of Lipophages

Our observations have indicated that lipophages may arise from blood monocytes within thrombi and that the lipid-filled vacuoles in them are derived principally from the blood platelets. The phagocytosis of lipid-rich platelets by monocytes and their subsequent digestion, a phenomenon which has been observed also in human thrombi and in tissue culture,[37] can logically account for the development of fat vacuoles within these cells. It has been observed by Mustard, Downie, Murphy and Roswell[36] that in human subjects the cholesterol content of washed platelets tends to be directly proportional to serum cholesterol levels. Fragments of erythrocytes are also phagocytized, but because of their relatively lower lipid content (Table I), they are not likely to be the major source of these lipids, as Duguid[2] has suggested. The experiment by Harrison[5] indirectly lends support to the hypothesis that lipid vacuoles are derived principally from platelets (and to a lesser extent erythrocytes) since no fat was stainable in the pulmonary arterial plaques he produced by injecting washed human fibrin clots, containing very

few erythrocytes and platelets, into the ear veins of rabbits. In a similar experiment by Thomas and co-workers,[8] whole blood clots were injected into the marginal ear veins of rabbits, in some of which hyperlipemia was produced. However, their investigations were consistently unable to find more lipophages in the embolic lesions of animals with hyperlipemia than in those fed low-fat diets.

It is of interest that although human venous thrombi are characteristically totally occlusive and undergo recanalization rather than plaque formation, they also exhibit thrombophagocytosis and contain lipophages,[37] as in the occlusive pulmonary thromboemboli of the rabbit (*vide supra*).

The existence of many intermediate forms between typical monocytes and equally typical lipophages, with no suggestion of derivation from endothelium, is at variance with the views of others.[38] It seems likely that lipophages are derived from the blood and are overgrown by endothelium, as Rannie and Duguid[39] postulated, and that they are blood monocytes, not Kupffer cells, as Leary[40] contended. Mitotic division of lipid-laden subendothelial macrophages was not observed in our experiments, but had been described by others.[41] Our studies of lipophages in thromboembolic plaques suggested that such cells divided by amitotic division. Transendothelial penetration by foam cells, illustrated in beautiful photographs of formalin-fixed specimens by Poole and Florey,[42] was not seen.

Cyclic and Progressive Atherogenesis

Our observation that some thromboembolic atherosclerotic plaques developed 3 weeks after embolization and then steadily underwent regression suggested that atherosclerotic lesions of this type might not always be progressive and irreversible. Instead, they could often be relatively transient structures, constantly developing and subsequently regressing, being supplanted by the occurrence of fresh lesions. Perhaps the original structure, a thrombus, may first become a plaque and may then undergo spontaneous regression. However, if factors exist which increase the rate of production of these plaques or impede and prevent their regression (hyperlipemia or hypercholesteremia could be such factors[36,43–45]), then they might in effect accumulate, producing the advanced atherosclerosis seen in cases of natural disease.

SUMMARY

In this experiment adult rabbits of both sexes were injected with multiple thromboemboli via the jugular vein; the emboli were autologous thrombi, prepared *in vitro*. Rabbits were sacrificed at various intervals,

and the fate of the emboli within pulmonary arteries was investigated in detail.

In the pulmonary arteries the thrombi became eccentric, fibrofatty atherosclerotic plaques. During the first week, a vascular inflammatory reaction and lysis of erythrocytes and granulocytes were conspicuous; the retracting thrombi became endothelialized. In succeeding weeks, fibrocytes derived from endothelium laid down coarse layers of collagen. Lipophages appeared and increased in numbers. These were derived from monocytes within the thrombi, principally by phagocytosis and digestion of blood platelets. Calcification of unphagocytized platelets was demonstrable after 3 weeks; and at 12 months bony metaplasia had taken place. The plaques fulfilled the gross and histologic criteria for atherosclerosis.

The experiment differed from previous efforts to demonstrate the thromboembolic origin of atherosclerosis experimentally. True autologous thrombi were produced *in vitro* and used as emboli; these were not fibrin clots or whole blood clots. Fibrofatty plaques produced contained abundant lipophages and calcium. No dietary manipulations to produce hyperlipemia were employed.

There appeared to be experimental evidence that fibrofatty atherosclerotic plaques may be produced by a natural process of degeneration and organization of pulmonary arterial thrombi or emboli.

Addendum

Since the submission of this manuscript, the work of Clark, Graef and Chasis[46] in 1936 has come to our attention. These investigators concluded from their studies of human aortas and coronary arteries that the fibrin-staining or "fibrinoid" material in atherosclerotic plaques represented "compressed and hyalinized remnants of organizing and frequently repeated surface deposits of fibrin." Clark and co-workers gave prior credit for this concept not to Rokitansky, but to Mallory[47] whose references to this mechanism were brief and lacked the detail of their own discussion. Although their publication clearly antedated Duguid's works in the 1940's, their efforts neither received the recognition nor had the same stimulating effect on research that Duguid's did.

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LEGENDS FOR FIGURES

Unless otherwise specified, histologic preparations used for photography were paraffin sections stained with hematoxylin and eosin.

FIG. 1. A fibrin clot from the coagulated tail of the thrombus depicted in Figure 4. The clot is composed of a random arrangement of leukocytes, erythrocytes and platelets in a fibrin meshwork. Phosphotungstic acid hematoxylin (PTAH) stain. × 160.

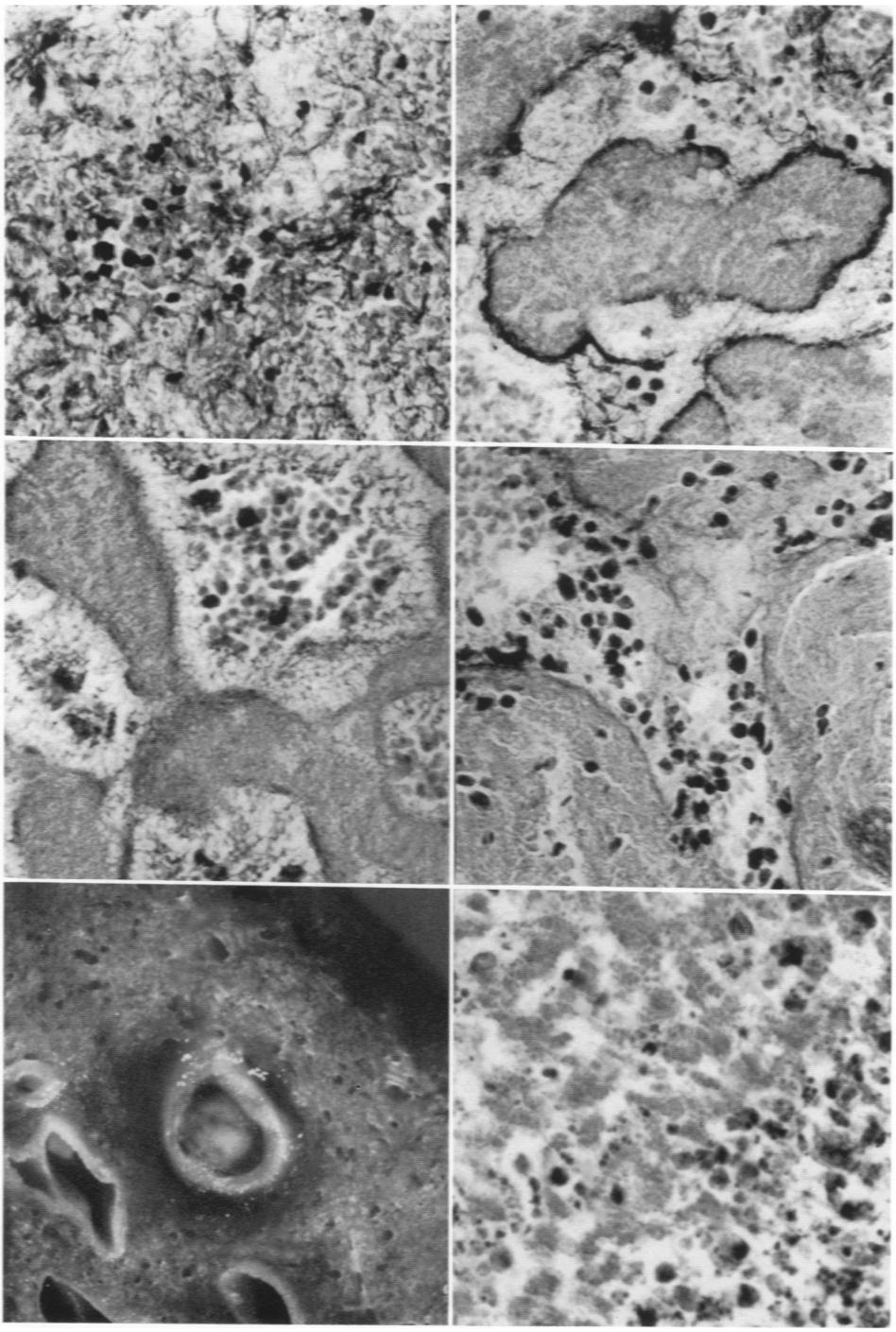
FIG. 2. A natural thrombus from the coronary artery of a patient who died 5 days after an acute myocardial infarction. Columns of discrete platelets are sharply delineated by dark borders of fibrin, among which scattered leukocytes and erythrocytes are seen. PTAH stain. × 160.

FIG. 3. A natural thrombus from a needle tract within the lung of a rabbit whose pulmonary artery was inadvertently punctured during cardiocentesis at the time of sacrifice. The platelet columns and the borders of fibrin are clearly seen. PTAH stain. × 160.

FIG. 4. A control thrombus, produced *in vitro* from rabbit's blood and withheld from injection. The arrangement of platelet columns, fibrin borders and scattered leukocytes is as shown in Figures 2 and 3. PTAH stain. × 160.

FIG. 5. A gross photograph of an autologous thromboembolus recently lodged in a rabbit's pulmonary artery. A central white area of thrombus with a slightly darker periphery of coagulated fibrin clot may be seen. The embolus is 3 days old. Buffered formalin fixation.

FIG. 6. Three days after embolization the thrombus exhibits extensive lysis of granulocytes. These persist only as "nuclear dust," while the monocytes remain viable. Lysis of fibrin strands gives the thrombus a porous, spongy appearance at this time, and the thrombus is extremely friable. × 100.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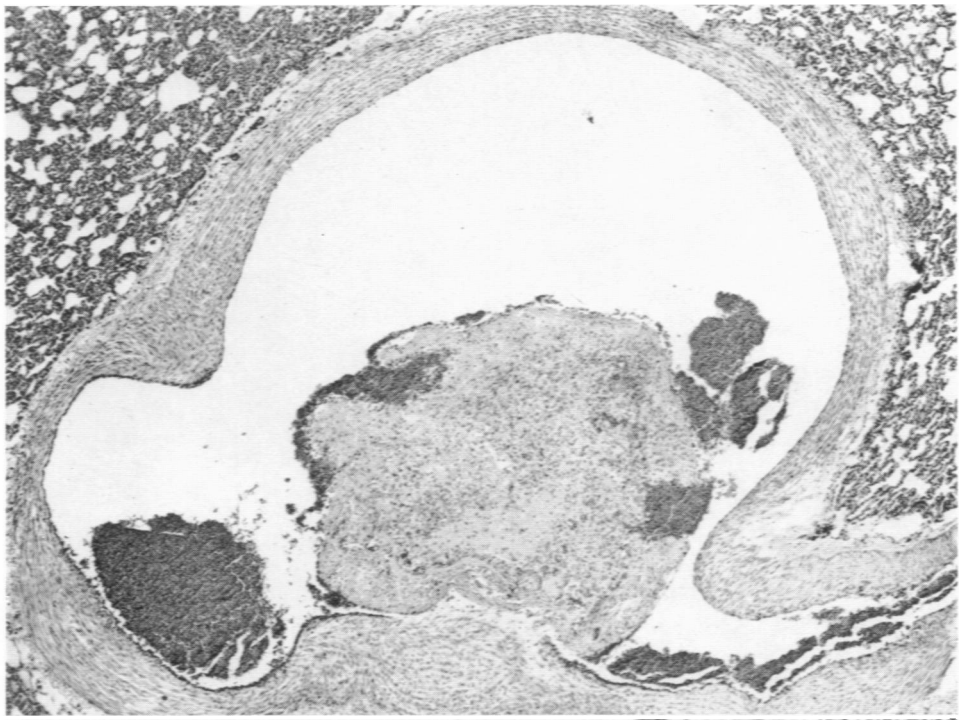

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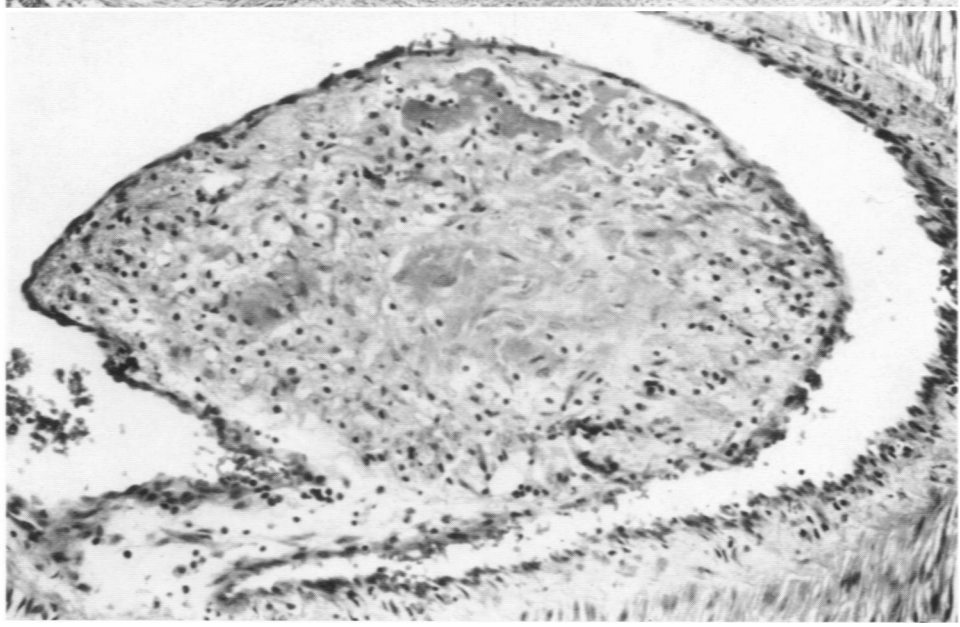


FIG. 7. Four days after embolization, an autologous thrombus has retracted, leaving a broad site of attachment at the arterial bifurcation. Endothelium has partially covered the embolus, which is easily distinguished from adjacent postmortem blood clot. × 10.

FIG. 8. Another retracting thromboembolus, completely endothelialized 8 days after embolization. This has a polypoid appearance in cross section because of its narrow mural attachment. All granulocytes in the thrombus have been lysed, but many large monocytes with prominent nuclei remain. A few platelet columns may be recognized in the upper portion of the thrombus. × 40.

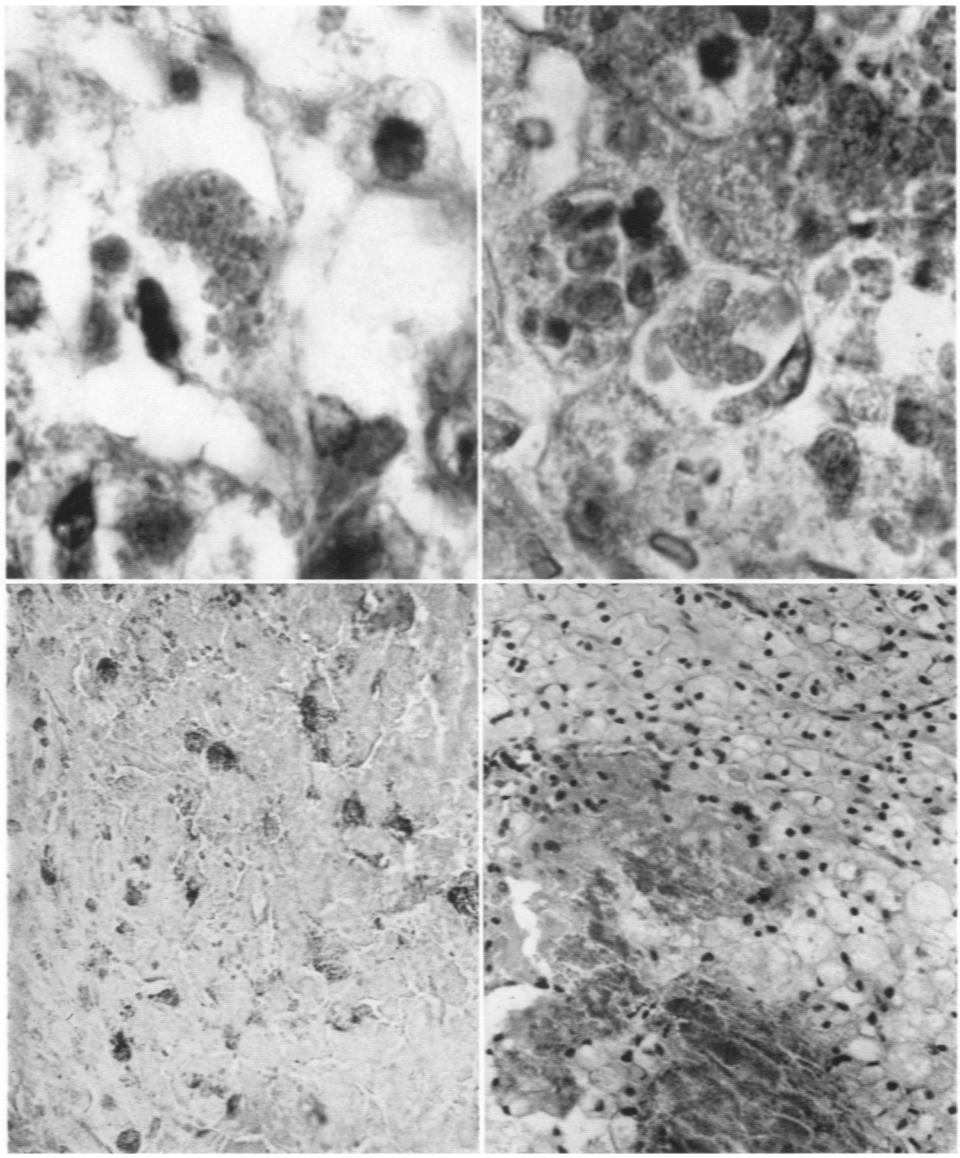
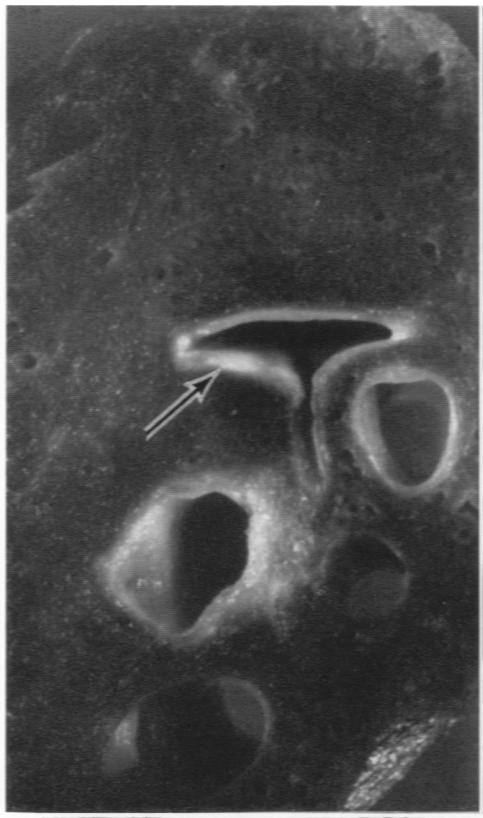


FIG. 9. Macrophages within an embolus 6 days old. The macrophage with eccentric nucleus occupying the center of the field contains numerous phagocytized platelets. Above and to the right, a lipophage contains a large cytoplasmic vacuole. × 400.

FIG. 10. Macrophages within an embolus 8 days old. The phagocyte at the center of the field contains several swollen but intact platelets. × 400.

FIG. 11. A thrombus 8 days after embolization. Partially digested platelets and cytoplasmic vacuoles within several lipophages stain brightly for fat. Sudan IV stain, frozen section. × 160.

FIG. 12. Foam cells (lipophages) in a thromboembolus 3 weeks old. The darkly stained, granular material adjacent to the foam cells is composed of unphagocytized, calcified platelets. These are also stainable by the von Kossa technique. × 160.



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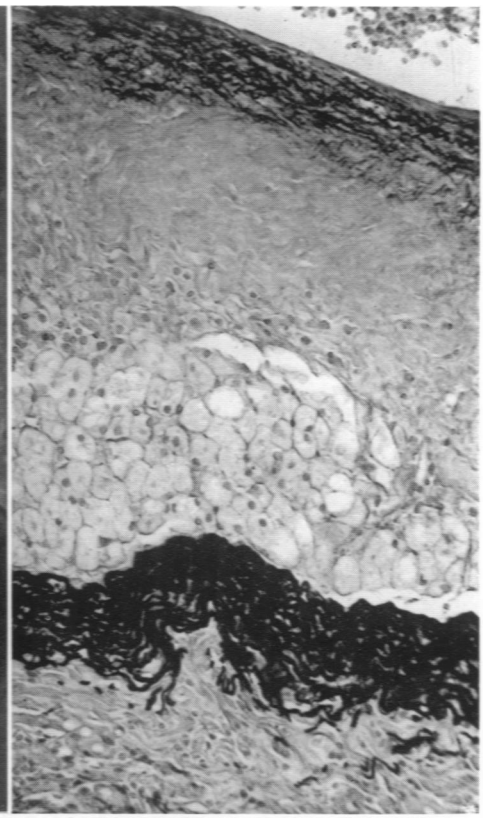

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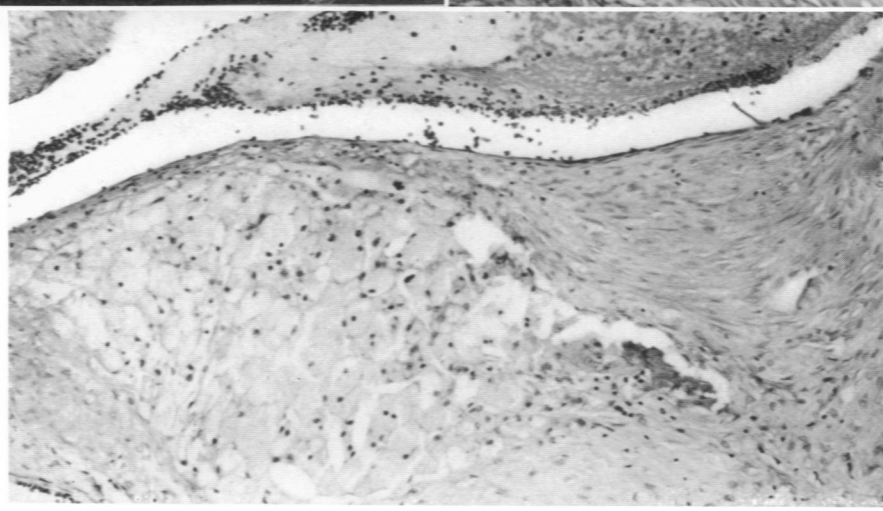

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FIG. 13. A gross photograph of a rabbit's lung, containing a pulmonary arterial thrombus 3 weeks after embolization. The original thrombus has become a fibrofatty atherosclerotic plaque (arrow), identified grossly at the T-shaped arterial bifurcation as an eccentric, butter yellow thickening of the intima. Buffered formalin fixation.

FIG. 14. A section of the plaque depicted in Figure 13. Characteristic layers of endothelium, elastica, collagen, lipophages and minute capillary channels comprise the plaque, beneath which the arterial media is atrophic. Resorcin fuchsin stain. × 160.

FIG. 15. Another atherosclerotic plaque derived from a thrombus injected 12 months before sacrifice. Bony metaplasia has taken place, and among the lipophages are fusiform spaces suggestive of cholesterol clefts. This plaque also exhibits the features seen in Figure 10. × 100.