DEGRADATION OF SOLUBLE FIBRIN COMPLEXES, FIBRINOGEN, AND FIBRIN BY MACROPHAGE ENZYMES.

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Stimulated macrophages are known to hydrolyze fibrinogen (F) and fibrin (f) through the elaboration of plasminogen (P) activator. We report here on the degradation of soluble fibrin complexes (SFC), F, and noncrosslinked f by rabbit alveolar macrophages (RAM) via a nonplasmin-mediated pathway. 125 I SFC in P-containing and P-free systems adhering to intact RAM were continuously degraded at equal rates into less than 30,000 Daltons, 60-80% trichloroacetic acid-soluble peptides, as evidenced by Bio Gel A-5m gel chromatography of cell-free supernatants. SFC degradation was blocked by 10^{-3} M iodoacetamide. P-free F, incubated with enzymes from lysed RAM, rapidly became incoagulable and potentially inhibited normal F-to-f conversion. SDS gel electrophoresis (SDSGE) (nonreducing conditions) demonstrated only a modest reduction of the MW of P-free F and f during initial incubation; peptides released were too small to be visualized on 6% SDS gels; after prolonged incubation, SDSGE demonstrated no discrete peptides. SDSGE (reducing conditions) revealed the initial proteolytic attack to involve F-f(A) α chains, followed by proteolytic cleavage of (B) β and γ chains. RAM enzymes hydrolyzed f more rapidly than F. Thus, RAM enzymes, in contrast to plasmin, degraded SFC, F, and f extensively, leaving no demonstrable "core fragments" resistant to further proteolytic attack. Since SFC have been shown to be rapidly removed from the circulation by the reticulo-endothelial system, the catabolic pathways delineated here may have pathophysiological significance.

THE STRUCTURAL CHARACTERISTICS OF CHICKEN FIBRINOGEN. J. Pindyck, M.W. Mosesson, D. Bannerjee, D. Galanakis.

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The structure and subunit composition of chicken fibrinogen (ϕ) have been investigated. Dodecyl sulfate gel electrophoresis of unreduced specimens revealed a single ϕ band with a molecular weight of approximately 320,000. ϕ and fibrin specimens were also electrophoresed after reduction with dithiothreitol, and after crosslinking of unreduced specimens in the presence of Factor XIIIa. Chromatographically separated S-sulfo chains were also studied after reptilase or thrombin treatment, and certain samples were stained with periodic acid Schiff reagent (PAS). Chicken A α chains were smaller than human A α chains (54,500 vs. 70,900, respectively) but, like mammalian A α chains, they possessed a reptilase and thrombin sensitive site, were PAS negative, and undergo Factor XIIIa catalyzed α -polymer formation. The sizes of chicken B β and γ chains were nearly the same as their mammalian counterparts, (i.e. 60,000 and 49,000 respectively); both types of chains were PAS positive. Chicken B β chains possessed a slowly reactive thrombin sensitive site apparently corresponding to the one in human β ; the chicken β chains, like mammalian β chains, did not undergo Factor XIIIa catalyzed cross-linking. Like mammalian γ chains, chicken γ chains could undergo Factor XIIIa catalyzed γ - γ dimerization and did not possess thrombin or reptilase sensitive sites. These findings indicate that the chicken fibrinogen molecule is composed of three pairs of disulfide-bridged chains corresponding in most respects to mammalian fibrinogen chains.

COMPARATIVE STUDIES ON FETAL FIBRINOGEN FROM FULL TERM AND PREMATURE INFANTS. D.K. Galanakis and M.W. Mosesson.

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Cord plasma or fibrinogen from full term (FT) infants, when compared with that from adults, displays delayed fibrin aggregation under conditions of high ionic strength (0.29); this phenomenon serves as an indicator of a fetal form of fibrinogen. In studies to be reported we compared adult fibrinogen with that from pooled premature (P) (24-35 weeks gestation) or FT (38-42 weeks) infant plasma. The thrombin and arvin clotting times of P fibrinogen (fraction I-2) were even more prolonged than those of FT fibrinogen. Dodecyl sulfate gel electrophoresis disclosed no significant differences in the amounts of coagulable fibrinogen catabolites in P, FT, or adult plasma. The thrombin time of P or FT plasma did not change following incubation at 37 $^{\circ}$ for 8 hours. Furthermore, there was no change in the thrombin (citrate) or Reptilase (heparinized) clotting time of P or FT plasma to which had been added sufficient adult fibrinogen to normalize the fetal fibrinogen effect prior to incubation at 37 $^{\circ}$. Thus, functional differences between adult and fetal fibrinogen cannot be attributed to the fetal plasma environment per se. The results suggest that plasma from infants born during the early part of the third trimester of pregnancy is relatively enriched with fetal fibrinogen; that from FT infants, by comparison, contains sufficient amounts of adult fibrinogen to result in shortening of the clotting times.