HIGHLY IODINATED FIBRINOGEN: A NEW THROMBUS-LOCALIZING AGENT

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We have examined radioiodinated fibrinogen prepared at high levels of iodination as an agent for improved in vivo thrombus detection. Fibrinogen containing 25, 50, and 100 atoms of iodine per molecule is prepared by an electrolytic procedure and is compared with conventional radiolabeled fibrinogen (<0.5 iodine atom per molecule) prepared by the iodine monochloride method. The level of iodination has little effect on the isotopic clottability of the product, but its degree of aggregation and its rate of blood clearance in experimental animals is strongly dependent on iodination level. Isotopic thrombus:blood ratios obtained in recently induced thrombi with the 25 atom per molecule preparation average about 50:1, twice as high as the ratios obtained with conventionally labeled fibrinogen.

Considerable effort has been devoted in recent years to the study of radioiodinated fibrinogen. Labeled fibrinogen is being used extensively and with increasing frequency in the detection of deep vein thrombosis. The fibrinogen uptake test has been shown to be an accurate method of detecting thrombi forming in the deep venous system of the lower extremities (*1–4*). This technique cannot be used for detecting thrombi in the upper thigh or pelvis, however, because in these regions insufficient differentiation is obtained between activity in the thrombus and background activity in the large vessels (*2,4*). This disadvantage of the fibrinogen uptake test is due to slow clearance of the labeled fibrinogen from the blood. In the present study, to develop an improved thrombus-localizing agent which may be useful in areas other than the lower extremities and which may be applicable to the detection of preformed thrombi, we have investigated fibrinogen iodinated extensively, with considerably greater than 0.5 atom per molecule. This is reported to be the maximum level at which fibrinogen can be iodinated by the iodine monochloride method without a significant increase in clearance rate (*5*). It has also been reported that iodine monochloride (ICl) iodination of fibrinogen with up to 10 atoms per molecule does not alter clottability (*6*). Our preliminary evidence indicated that a faster clearance rate combined with relatively unchanged clottability were probably responsible for the higher thrombus:blood ratios obtained with highly iodinated fibrinogen (*7*). Under these conditions, thrombi might be more readily detected clinically by imaging with a scintillation camera.

This paper describes an electrolytic procedure for the preparation of highly iodinated fibrinogen. The physicochemical properties, biologic clearance, and thrombus:blood ratios of canine fibrinogen iodinated electrolytically with 25, 50, and 100 atoms per molecule are reported and compared with ICl-labeled preparations.

MATERIALS AND METHODS

Electrolysis. Canine fibrinogen is separated from pooled plasma as the Blombäck I-2 fraction (*8*) by the procedure of Mosseson and Sherry (*9*). It has a spectroscopic clottability ranging from 96–98% (*10*). The procedure for electrolytic iodination is based in part on that of Rosa, et al (*11,12*) and Katz and Bonorris (*13*). The electrolytic cell consists of a 15-ml platinum crucible anode, a 1-cm platinum wire cathode, and a saturated calomel reference electrode. The cathode is contained within a small dialysis bag to prevent direct contact with the anolyte. Prior to each iodination, the surfaces of the platinum electrodes are treated by the method of Adams (*14*) to insure removal of organic contami-

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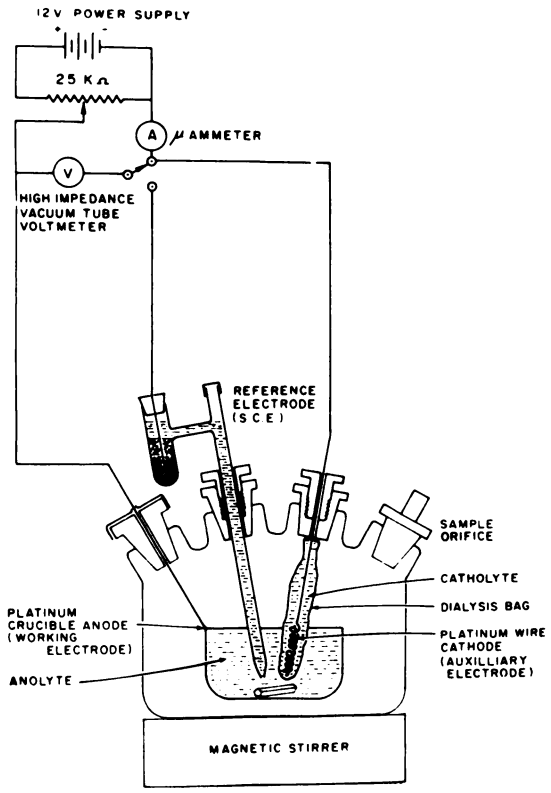

FIG. 1. Electrolysis cell assembly and associated electrical circuit employed in electrolytic iodination reactions.

nants and adsorbed oxygen. The entire cell assembly is contained within a 125-ml Bantam-Ware resin flask to maintain a closed system. The various inlet and outlet tubes and wires are passed through the standard taper joints in the head of the resin flask. A drawing of the cell assembly and a schematic of the electrical circuit are shown in Fig. 1.

Several buffers were employed as reaction media including phosphate, acetate, tris, and barbital. The optimum reaction medium is 0.02 *M* sodium barbital — 0.20 *M* NaCl buffer at pH 7.4. The anolyte consists of 8.0 ml of this buffer containing 3 mg fibrinogen, the desired level of $Na^{125}I$ or $Na^{131}I$ activity, and sufficient carrier $Na^{127}I$ to achieve a 150:1 molar ratio of iodide:fibrinogen. The catholyte consists of 0.5 ml of the barbital buffer. The radioiodide (Industrial Nuclear Corp., St. Louis, Mo.) is of high purity, free of reducing agents, and is supplied in 0.1 *N* NaOH at a concentration of approximately 1 mCi/μl. Iodination is performed at an anode potential of +0.4 to +0.5V measured against the reference electrode. The potential between the anode and cathode ranges from approximately 0.75 to 1.5V while the observed current varies from 10 to 30 μA.

The progress of the reaction (labeling efficiency) is followed by withdrawing 10-μl aliquots of the anolyte at regular intervals and precipitating the fibrinogen with 12% (w/v) trichloracetic acid (TCA) after addition of albumin carrier. The precipitate and supernate are counted in a simple gamma scintillation well counter (Picker Corp.) to determine the relative amount of radioactivity associated with the protein. The reaction is allowed to proceed to the point at which the desired average number of iodine atoms per molecule of fibrinogen have been incorporated as determined from the labeling efficiency and the initial molar ratio of iodide:fibrinogen. Labeling normally proceeds at a rate of 15–20%/hr, and typical reaction times vary from 1½ to 4½ hr, depending on extent of iodination desired. Following electrolysis, the anolyte is withdrawn and dialyzed three times for 1 hr each against 500-ml portions of 0.02 *M* tris — 0.15 *M* NaCl buffer (pH 7.4). At this stage, <5% free iodide remains. If not used immediately, the labeled fibrinogen is stored in polyethylene tubes at —20° after addition of albumin (2–3% w/v).

Iodine-monochloride labeling. Iodine-monochloride labeling of fibrinogen with approximately 0.5 atom per molecule is effected by standard procedures (*15–17*). Briefly, the $Na^{125}I$ or $Na^{131}I$ is mixed with ^{127}ICl to allow exchange of the radioiodine and the carrier iodine. This mixture is then added to the fibrinogen. The amount of ICl is adjusted to give a 1:1 molar ratio of ICl:fibrinogen. The actual iodination level is determined from this initial molar ratio and the labeling efficiency. Iodine-monochloride labeling of fibrinogen with 25 atoms per molecule is performed in a similar manner except that a 50:1 molar ratio of ICl:fibrinogen is used with a labeling efficiency of approximately 50%. Some of the ICl preparations with 0.5 atom per molecule were further subjected to the same conditions of buffer, electrode potential, reaction time, etc., as those used in the preparation of highly iodinated products.

Physicochemical tests. For calculations based on radioactivity measurements, the samples are counted for a sufficient time to accumulate on the order of 10^5 counts per sample. Isotopic clottability is determined by the method of Regoeczi (*18*). The stability of the fibrinogen–iodine bonds is determined by the rate of hydrolytic deiodination of the labeled fibrinogen in normal saline (pH 7.4) at 37°C. Aliquots are withdrawn at regular intervals, and the protein is precipitated with TCA. The precipitable activity at any given time is expressed as percent of the initial value. The relative amount of iodine bound to the sulfhydryl groups of cysteine residues of the fibrinogen is estimated by adding cysteine to the labeled fibrinogen and determining the amount of free iodide liberated as reflected by the decrease in TCA precipitable activity. Molecular weight profiles of the

labeled fibrinogen preparations are determined by chromatography on Sepharose 4B columns, 0.9 cm × 45 cm or 1 cm × 24 cm. The columns are eluted at 4°C with 0.02 *M* tris–0.135 *M* NaCl buffer (pH 7.4) containing 0.05 *M* ε-aminocaproic acid.

Animal studies. The in vivo studies of the labeled fibrinogen include determination of its blood clearance and measurement of its uptake by experimentally induced thrombi. For the clearance studies, 200 μCi of ^{131}I-labeled fibrinogen at the desired iodination level are injected intravenously into mongrel dogs weighing 15–25 kg and anesthetized with sodium pentobarbital. In each case, 150 μCi of ^{125}I-labeled fibrinogen with 0.5 atom per molecule are injected at the same time for comparison. Serial blood samples are withdrawn to EDTA tubes and centrifuged. The clottable activity of the plasma is determined and expressed as a percentage of the initial (3 min) value. For these radioactivity measurements, an automatic four-channel gamma scintillation well counter (Searle Radiographics) is employed. The raw counts obtained are automatically corrected for background, decay, and scatter from the ^{131}I channel into the ^{125}I channel with a Wang Model 600 programmable calculator. The percent initial activity cleared at any given time and the half-life of the fibrinogen are calculated directly from the clearance curves. At various times the relative amount of activity that remains protein-bound is determined by the TCA precipitability of the corresponding plasma sample.

Thrombus uptake determinations are performed as previously reported (*19*). Briefly, 100 μCi of ^{131}I-labeled fibrinogen at the desired iodination level and 100 μCi of ^{125}I-labeled fibrinogen at the 0.5 atom per molecule level are injected 4 hr after femoral vein thrombosis is induced by a locally applied electric current. Twenty-four hours after tracer injection the thrombus is removed, weighed, and counted. A weighed blood sample is obtained at the same time and counted. The thrombus:blood activity ratio is calculated as (cpm/gm thrombus) ÷ (cpm/gm blood).

RESULTS AND DISCUSSION

Nature of the electrolytic reactions. Several factors in the electrolytic reactions are critical including the nature of the buffer, anode potential, and pH. In phosphate, acetate, and borate buffers the electrolytic iodination either does not occur or proceeds at a very slow rate. This result is probably due to electrode adsorption and inhibition effects arising from the small size and high mobility of the anions of these buffer salts and the presence of the polarizable hydroxyl group (*20*). With barbital buffer, such problems do not occur. Furthermore, use of tris buffer as an electrolysis medium leads to erroneous electrode potential measurements, probably due to junction potential effects with the saturated calomel electrode (*21*).

TABLE 1. SOME COMMON HALF-CELL REACTIONS AND STANDARD ELECTRODE POTENTIALS FOR IODINE

Reaction	E^0, Volts versus saturated calomel electrode
$I^- + 6OH^- \rightleftarrows IO_3^- + 3H_2O + 6e^-$	−0.02
$I^- + 2OH^- \rightleftarrows IO^- + H_2O + 2e^-$	−0.25
$2I \rightleftarrows I_2 + 2e^-$	−0.30
$I^- + H_2O \rightleftarrows HIO + H^+ + 2e^-$	−0.75

The standard electrode potentials (E^0) for some common half-cell reactions of iodine are shown in Table 1. The potentials listed refer to a standard state of unit molarity. Calculations based on the Nernst equation show that at the concentration of iodide and the pH employed in this study, only the oxidation of iodide ions to elemental iodine will occur at the controlled anode potential of +0.4–+0.5V relative to the saturated calomel electrode (*20*). At working potentials below this range, the labeling reaction is too slow to be useful. At potentials above this range, a considerable amount of the fibrinogen precipitates from the anolyte during electrolysis and cannot be redissolved.

The optimum pH range is 7.0–7.4. At lower pH values the labeling rate drops significantly. At higher pH values there is a possibility of side reactions (Eqs. 1 and 2, Table 1). The concentration of iodide, $1.5 \times 10^{-4}M$, is slightly above the minimum value, $5 \times 10^{-5}M$, needed to insure that the neutral iodine radicals formed at the anode will encounter each other and form diatomic iodine prior to reaction with the fibrinogen (*12*). The concentration of fibrinogen, $1.0 \times 10^{-6}M$, is the minimum possible. At lower concentrations no labeling occurs. The resulting ratio of iodide:fibrinogen is 150:1 at which the desired extent of iodination can be achieved in a reasonable amount of time.

These electrolytic reactions are simple, mild, and readily controlled. At the level of 25 iodine atoms per molecule, these reactions yield a product which compares favorably in vitro with fibrinogen labeled with low levels of iodine by the ICl or lactoperoxidase methods, the methods reported to be the most satisfactory (*22*). In general, electrolytic procedures are often the mildest means of effecting a particular reaction (*23*), a principle which has been recognized in relation to the iodination of fibrinogen (*11,13*).

TABLE 2. PROPERTIES OF FIBRINOGEN IODINATED BY THE ELECTROLYTIC METHOD*

Iodination level (iodine atoms per molecule Φ)	Isotopic clottability (%)	Thrombus:blood ratio	Initial activity cleared in first 24 hr in vivo (%)	TCA precipitability after first 24 hr in vivo (%)	$T_{1/2}$ of late component of blood clearance curve (hr)
25 (n = 3)	73 ± 3	50 ± 7	64 ± 3	80 ± 4	37 ± 2
50 (n = 3)	65 ± 5	15 ± 5	91 ± 2	61 ± 4	22 ± 3
100 (n = 3)	60 ± 4	3 ± 2	97 ± 2	25 ± 6	10 ± 2

* Results expressed as group mean ± 1 s.d.

TABLE 3. PROPERTIES OF FIBRINOGEN IODINATED BY THE ICL METHOD*

Iodination level (iodine atoms per molecule Φ)	Isotopic clottability (%)	Thrombus:blood ratio	Initial activity cleared in first 24 hr in vivo (%)	TCA precipitability after first 24 hr in vivo (%)	$T_{1/2}$ of late component of blood clearance curve (hr)
0.5† (n = 6)	80 ± 2	23 ± 3	52 ± 2	82 ± 3	52 ± 2
0.5‡ (n = 2)	78 ± 5	24 ± 3	56 ± 2	75 ± 6	52 ± 2
0.5‖ (n = 2)	75 ± 3	——	61 ± 2	75 ± 5	52 ± 2
25 (n = 3)	71 ± 5	33 ± 3	79 ± 4	71 ± 4	28 ± 3

* Results expressed as group mean ± 1 s.d.
† No further treatment following labeling.
‡ Following labeling, subjected in the absence of additional iodine to the electrolytic conditions employed in the labeling of fibrinogen with 25 atoms per molecule.
‖ Following labeling, subjected in the absence of additional iodine to the electrolytic conditions employed in the labeling of fibrinogen with 100 atoms per molecule.

Previous work on electrolytic labeling of fibrinogen in this laboratory, however, was rather unsuccessful and resulted in considerable sulfhydryl group labeling and aggregation of the labeled molecules (*24*). The improved results in the present study are due to careful control of the anode potential and thus the nature of the anodic reactions. When performed in this manner, electrolytic iodination may be of greater general value than indicated by the earlier results.

The present work also demonstrates the superiority of the electrolytic method over the ICl method in the preparation of highly iodinated fibrinogen. Attempts to iodinate with ICl at levels over 25 atoms per molecule result in immediate irreversible precipitation of the protein upon addition of the ICl solution. This result is probably due to the strongly oxidizing environment which results from the requisite high concentrations of ICl. Iodine-monochloride iodination at the level of 25 atoms per molecule is achievable, but the properties of the product indicate that the labeling process produces greater alteration of the fibrinogen than does the electrolytic labeling. Reaction of fibrinogen with molecular iodine, $^{127}I_2$, has been used to incorporate up to 270 iodine atoms per molecule of fibrinogen in studies not involving labeling with radioactive isotopes (*25*). However, applicability of this technique to the radiolabeling of fibrinogen for in vivo use is doubtful.

Chemical properties. The properties of the highly iodinated fibrinogen preparations produced electrolytically are given in Table 2. Additionally, other electrolytic reactions as well as some ICl reactions were performed to further evaluate the preparations listed in Table 2. These additional reactions were designed to investigate the relative utility of the electrolytic and ICl methods in achieving the same level of fibrinogen iodination and the effect of the electrolytic conditions per se on the properties of the labeled fibrinogen. The data for these reactions are presented in Table 3.

The isotopic clottability of the labeled fibrinogen shows little change with increasing extent of iodination (Tables 2 and 3). Even at the level of 100 atoms per molecule, the clottability is 60%, while, as noted in subsequent sections, the product containing 100 atoms per molecule has a considerably different molecular weight distribution and clearance rate than the preparations with lower numbers of iodine per molecule. These results demonstrate again that in vitro clottability is not a satisfactory measure of the quality and in vivo behavior of radioiodinated fibrinogen (*26*).

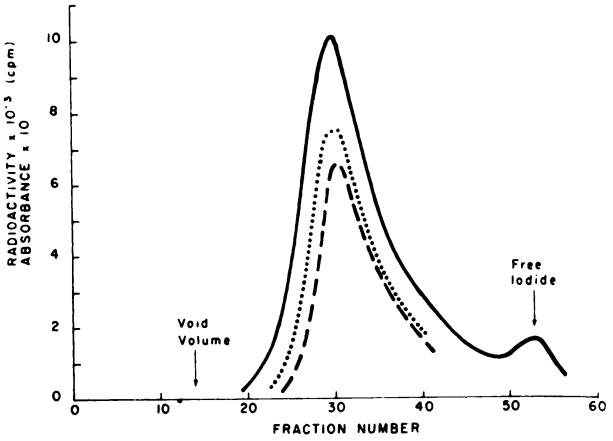

FIG. 2. Molecular weight profiles obtained on Sepharose 4B gel column for authentic normal fibrinogen (- - - -); fibrinogen iodinated with 25 iodine atoms per molecule by electrolytic method (——); and fibrinogen iodinated with 25 iodine atoms per molecule by ICl method (. . . .).

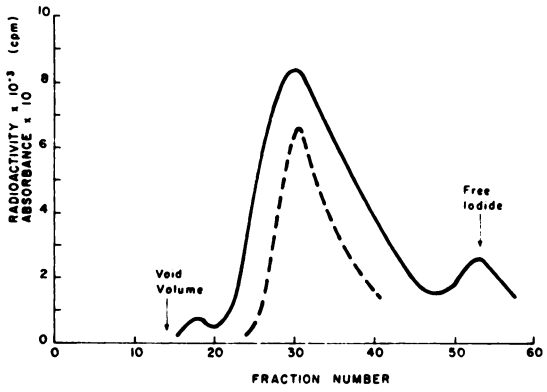

FIG. 3. Molecular weight profiles obtained on Sepharose 4B gel column for authentic normal fibrinogen (- - - -) and fibrinogen iodinated with 50 iodine atoms per molecule by electrolytic method (——).

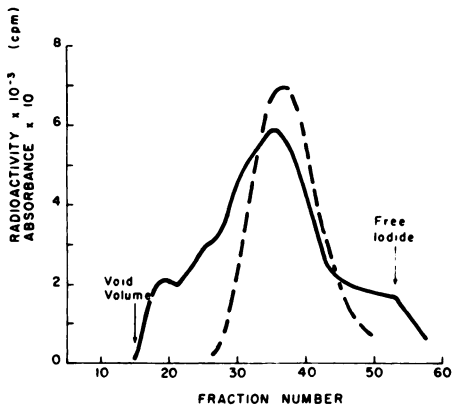

FIG. 4. Molecular weight profiles obtained on Sepharose 4B gel column for authentic normal fibrinogen (- - - -) and fibrinogen iodinated with 100 iodine atoms per molecule by electrolytic method (——).

Addition of cysteine to the labeled fibrinogen results in losses of TCA-precipitable activity ranging from 0 to 10%. This contrasts sharply with the results of previous electrolytic studies in this laboratory in which from 40 to 70% of the iodine was bound to sulfhydryl groups (*27*). Furthermore, hydrolytic deiodination rates of approximately 2%/day are observed in the present study compared with 20%/day in the previous work (*24*). These results, too, may reflect different degrees of sulfhydryl binding. These marked differences are an indication of the extent to which the present electrolytic reactions have been modified.

Molecular weight profiles. Molecular weight profiles of the labeled fibrinogen preparations are presented in Figs. 2, 3, and 4. In each case, the chromatogram of a sample of authentic normal fibrinogen is included for comparison. At the iodination level of 0.5 or 25 iodine atoms per molecule of fibrinogen, the profiles are indistinguishable from that of normal fibrinogen. At the level of 50 iodine atoms per mole-

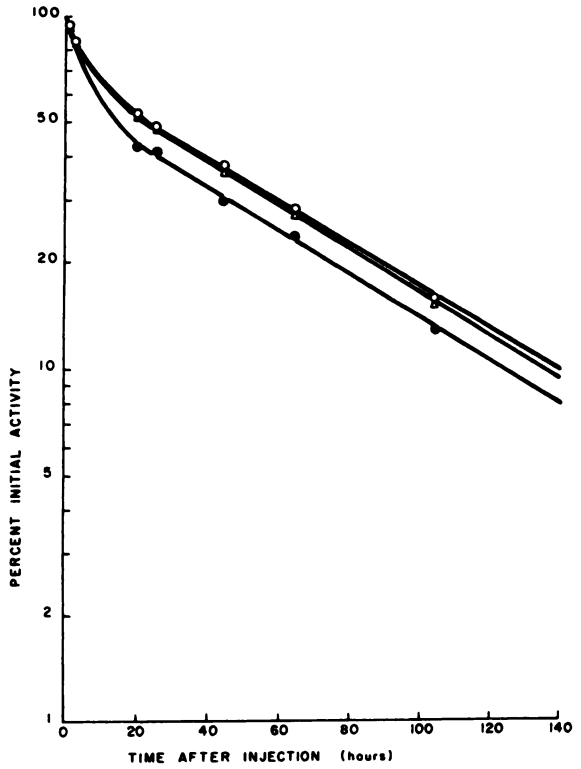

FIG. 5. Blood clearance curves for fibrinogen iodinated with 0.5 iodine atom per molecule by ICl method, with no subsequent treatment (○); fibrinogen iodinated with 0.5 iodine atom per molecule by ICl method, then further subjected in absence of additional iodine to electrolytic conditions employed in labeling of fibrinogen with 25 atoms per molecule (△); and fibrinogen iodinated with 0.5 iodine atom per molecule by ICl method, then further subjected in absence of additional iodine to electrolytic conditions employed in labeling of fibrinogen with 100 atoms per molecule (●).

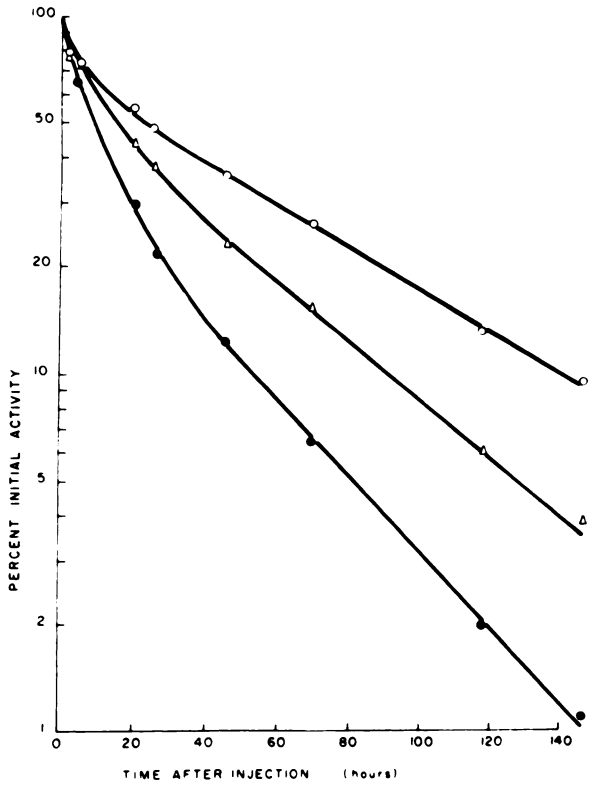

FIG. 6. Blood clearance curves for fibrinogen iodinated with 0.5 iodine atom per molecule by ICl method (○); fibrinogen iodinated with 25 iodine atoms per molecule by electrolytic method (△); and fibrinogen iodinated with 25 atoms per molecule by ICl method (●).

cule, a small peak appears before the fibrinogen peak, near the void volume. At the level of 100 iodine atoms per molecule, a significant early peak and a great deal of front tailing on the fibrinogen peak are evident. The two preparations iodinated by the ICl method and then further subjected to electrolytic conditions exhibit chromatograms indistinguishable from that of the 0.5 iodine per molecule material which has not undergone further treatment.

The high molecular weight components cannot simply consist of individual fibrinogen molecules with large numbers of iodine atoms attached. Even if labeled with 300 atoms of iodine, the fibrinogen would increase in molecular weight by only about 10%. This is not a sufficient molecular weight increase to cause the appearance of an early peak or shoulder with the columns employed. Instead, the early components of the chromatograms probably consist of aggregates of the labeled fibrinogen with sufficiently large molecular weight to be eluted before the nonaggregated material but not so large as to be incapable of penetrating the gel. In the case of the 25 and 50 iodine per molecule preparations, most of the radioactivity applied to the column is recovered in the effluent. With the 100 iodine per molecule material, however, only about 45% is recovered. When the column is scanned vertically with a portable detector, a great deal of activity is found on top of the gel. Evidently, macroaggregates present in the applied sample do not penetrate the gel. This phenomenon has been observed previously with other labeled fibrinogen preparations (*24*).

Clearance curves. Blood clearance curves for the various preparations are presented in Figs. 5, 6, and 7. The clearance of fibrinogen labeled with 0.5 iodine atom per molecule by the ICl method is presented for comparison in each case. At the level of 25 iodine atoms per molecule produced electrolytically, the clearance curve differs only slightly from the curve for the 0.5 iodine per molecule material. At the level of 50 iodine atoms per molecule, a considerably faster clearance is observed while at the level of 100 iodine atoms per molecule the clearance rate is still faster. Fibrinogen iodinated with 25 iodine atoms per molecule by ICl exhibits a faster clearance rate than the electrolytic preparations at the same iodination level. The clearance curves for the two products labeled by ICl and then further treated electrolytically do not differ significantly from that of the 0.5 iodine atom per molecule product.

The rates of clearance of the various preparations during the first 24 hr are summarized in Tables 2

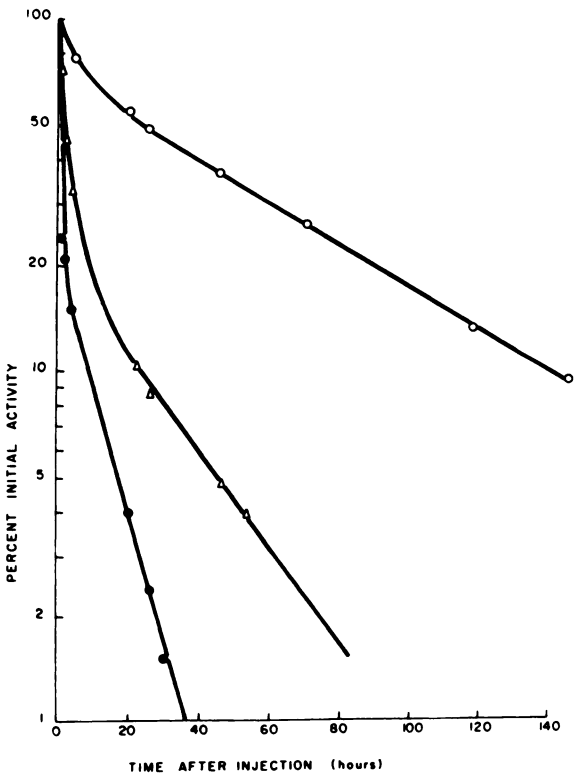

FIG. 7. Blood clearance curves for fibrinogen iodinated with 0.5 iodine atom per molecule by ICl method (○); fibrinogen iodinated with 50 iodine atoms per molecule by electrolytic method (△); and fibrinogen iodinated with 100 iodine atoms per molecule by electrolytic method (●).

and 3. During this early period, clearance occurs by several mechanisms including filtration of aggregates by the liver, intravascular-extravascular exchange, and hydrolytic deiodination (*28*). Thereafter, the clearance curves reflect predominantly the catabolism of the labeled fibrinogen remaining in circulation. While there are differences in this late component of the clearance curves, the major differences among the labeled fibrinogen preparations lie in the amounts rapidly cleared in the early part of the curves. This correlates with the amount of aggregation observed for the various iodination levels. The higher the level of iodination, the greater the amount of material that is rapidly removed from circulation by the liver. Furthermore, oxidation of the aggregates may occur in the liver with the release of free iodide into the blood stream. This latter phenomenon may account for the differing levels of free iodide (differing TCA precipitability) in vivo which is observed with the various preparations (Tables 2 and 3).

Thrombus uptake. Thrombus:blood ratios obtained with the various labeled fibrinogen preparations are presented in Tables 2 and 3. Fibrinogen iodinated electrolytically with 25 atoms per molecule exhibits the best thrombus:blood ratio, 50:1. This value is higher than those obtained either with fibrinogen iodinated electrolytically with 50 or 100 atoms per molecule or with fibrinogen iodinated by ICl with 25 atoms per molecule. This value is also superior to the 23:1 ratio obtained with fibrinogen iodinated by ICl with 0.5 atom per molecule, which is the material normally used in the fibrinogen uptake test for detection of deep vein thrombosis (*2*).

Among the factors that will affect these thrombus:blood ratios are the innate ability of the radiolabeled fibrinogen to be incorporated into a developing thrombus and the rate of clearance of the fibrinogen from the blood. Direct assessment of the ability of the fibrinogen to be incorporated into a thrombus in vivo is difficult although, as noted earlier, the alteration of structure at the higher iodination levels may be associated with reduced biologic activity. On the other hand, the rate of clearance of the fibrinogen is readily determined and shows significant variation among the various levels of iodination. The rate of clearance exerts two opposing effects. Ideally, the rate should be slow enough to allow the fibrinogen to circulate in the blood for a sufficient time to become incorporated into the thrombus but fast enough to allow the unincorporated fibrinogen to be removed from the blood to reduce background activity. Apparently, the rate of clearance of the 25 atoms per molecule fibrinogen produced electrolytically gives the optimum balance between these two opposing effects and the best thrombus uptake value is obtained. At the higher iodination levels, the aggregates are very rapidly removed from circulation by the liver. As a result, the length of time that the fibrinogen circulates in the blood is not sufficient to produce significant thrombus uptake values.

In addition, the preparations with 50 and particularly 100 atoms per molecule suffer from lower in vivo TCA precipitability due to release of considerable free iodide into the blood. This is in contrast to in vitro deiodination, which is very slow. Higher levels of free radionuclide in the blood contribute to higher background activity. Although some of this background activity could perhaps be eliminated through renal excretion, this may not be a desirable or even feasible procedure. Finally, at the higher iodination level the degree of alteration of the fibrinogen appears to be considerable and to vary from one iodination reaction to another. Accordingly, the fibrinogen with 100 atoms per molecule tends to behave in a somewhat random manner compared with the fibrinogen with lesser iodination. Although one fairly high thrombus uptake value was observed for a preparation with 100 atoms per molecule (*7*), other studies at this iodination level produced much lower values, averaging approximately 3:1, as shown in Table 2.

In summary, we have prepared a new thrombus-localizing agent that has an improved thrombus:blood ratio over conventionally labeled fibrinogen in 4-hr-old thrombi. This preparation may have considerable advantages over other agents for thrombus detection in areas of the body other than the lower extremities where blood background is a problem. In this regard, we are developing techniques for preparing highly iodinated fibrinogen with ^{123}I as the radioactive isotope. Because of the short physical half-life of ^{123}I relative to ^{125}I and ^{131}I, and because the 159-keV photon of ^{123}I is ideal for imaging with a scintillation camera, this isotope is a more suitable label for highly iodinated fibrinogen. The radiation dose resulting from ^{123}I would be considerably lower than from ^{125}I and ^{131}I. With the short physical and shorter biologic half-lives, the ^{123}I-radiopharmaceutical should be a superior agent for the localization of recently formed thrombi.

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