Coating Membranes for a Sorbent-Based Artificial Liver: Adsorption Characteristics

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ABSTRACT

Techniques are described for the coating of sorbents to be used in an artificial liver support system based on mixed sorbent bed hemoperfusion. Activated charcoal has been coated with cellulose acetate (CA) by solvent evaporation. With Amberlite XAD-4, the Wurster technique was used for coating with CA. XAD-4 has also been coated with a synthetic polyelectrolyte with anticoagulant activity by adsorption and fixation by gamma radiation-induced crosslinking. Activated charcoal, XAD-4, and a cation exchange resin, all in powdered form, were encapsulated in agarose gel beads. Adsorption characteristics onto the sorbents are described. The results are in agreement with a theoretical model presented. In general, adsorption onto XAD-4 is limited by film diffusion. With activated charcoal, pore diffusion limitation is generally observed. Blood compatibility is improved by coating.

Key words: coatings, sorbents, adsorption kinetics theory and practice, blood compatibility

INTRODUCTION

Treatment of severe acute and chronic liver failure remains a serious medical problem. The encephalopathy and coma associated with fulminant hepatic failure is generally attributed to the accumulation of water-soluble as well as protein-bound and fat-soluble toxic compounds in the systemic circulation.¹ Although the exact identity of the etiological factors is still unknown, several agents such as ammonia, fatty acids, mercaptans, phenols, biogenic amines, some

amino acids, and probably middle molecules are reported to be involved.¹ In addition, synergistic effects are observed between several of the toxins listed above.² Removal of these toxins from the blood seems to be a rational approach. Hemodialysis, however, does not improve survival.³ This is likely due to protein binding and water insolubility of some of the toxins involved. Hemoperfusion through coated activated charcoal improves the survival of rats with grade II hepatic coma;⁴ however, the effect in clinical studies has yet to be proven.⁵ Activated charcoal is very effective in the removal of water-soluble compounds, but is less suited for protein-bound substances.⁶ Both neutral and charged resins are more appropriate.⁷ Therefore, hemoperfusion through a combination of activated charcoal, neutral (XAD) and cation exchange resins at an early stage of the disease may be worthwhile to try in the treatment of acute liver failure in humans.^{6,8} The application of this kind of treatment is hampered by serious blood compatibility problems. Notably, resins show very severe thrombocyte losses,⁷ and, especially with liver patients, this cannot be accepted since these patients already have an increased bleeding tendency. In order to reduce the loss of platelets, coating of the sorbents with thin, permeable, blood-compatible membranes is required.

Tijssen et al.⁹ have described the coating of charcoal with an ultrathin membrane of cellulose acetate (CA), making use of a simple solvent evaporation technique. In vivo platelet losses were of the order of 15% after four hours of hemoperfusion of intoxicated patients. This technique was adopted in this work for the coating of activated charcoal with CA. The procedure failed in the coating of resins because of strong agglomeration of resin particles. Avoiding this, Amberlite XAD-4 has successfully been coated with CA by means of the Wurster technique.¹⁰ In this fluid-bed technique, partial drying of resin particles occurs. For that reason, only neutral resins could be coated, since ion exchange resins show too strong an expansion

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upon rewetting, which causes the coating membrane to rupture. Furthermore, XAD-4 has been coated with a synthetic polyelectrolyte (PLE), which has anticoagulant activity, by adsorption followed by fixation of the adsorbed PLE by gamma radiation-induced crosslinking.¹⁰ Since no adsorption of PLE takes place onto cation exchange resin, this procedure proved to be not applicable for this sorbent, while a PLE coating applied to activated charcoal appeared to be unstable.

Brunner et al.¹¹ have described a method for the encapsulation of sorbents in agarose gel. Using this technique, activated charcoal, Amberlite XAD-4 and a cation exchange resin, all in powdered form, were encapsulated. The mechanical strength of these capsules is not very high and sterilization is difficult since the capsules have a melting point of 90°C and are not resistant to gamma radiation. To overcome these problems, a crosslinking procedure has been developed by the reaction with 1,3-dichloro-2-propanol at strong alkaline conditions. After crosslinking, the compression strength is increased by a factor of 4, while the capsules can be autoclaved at 122°C for several hours without changes in properties.¹²

A logical consequence of these procedures is to investigate to what extent these coatings affect the adsorption characteristics of the sorbents. We have, therefore, studied this influence and the effects of sorbent particle size and of hydrodynamic conditions on the adsorption kinetics. This has been related to a theoretical model. The in vitro blood compatibility with regard to thrombocyte losses, using both citrated and heparinized fresh human blood, was examined.

THEORETICAL CONSIDERATIONS

Adsorption is a complicated process in which molecules are transported from the well-mixed bulk of the solution across a thin, stagnant, liquid film enveloping every particle, through the pore system, to the interior of the sorbent. Depending on concentration, diffusion constants of the adsorbate, and hydrodynamic conditions, the rate of adsorption can either be limited by diffusion through the pores of the sorbent or by diffusional transport across the stagnant film. For both extremes, simple approximative relations can be derived, expressing the concentration in the bulk of the solution as a function of the recirculation time. The derivations were based on those of Boyd et al.,¹³ but were corrected for a recirculation system with decreasing concentration.

Film Diffusion Limitation

When a linear concentration gradient across the film and equilibrium at the surface of the sorbent are

assumed for all times of contact, combination of the mass balance and the first law of Fick leads to Equation 1:

$$(C_t - C_\infty)/(C_0 - C_\infty) = \exp(-RSt) \quad (1)$$

In this relation the rate constant is

$$R = 3D/r\delta\alpha \quad (2)$$

$$\text{and } S = C_0/C_\infty = m\alpha/\rho V + 1 \quad (3)$$

The distribution coefficient α , defined as $\alpha = C_\infty^{\text{sor}}/C_\infty^{\text{liq}}$, and the diffusion constant D are assumed to be independent of the concentration. When the concentration at infinite time, C_∞/C_0 , approaches zero, $m\alpha/\rho V \gg 1$, and Eq. 1 reduces to Eq. 4.

$$C_t/C_0 = \exp(-R't) \quad (4)$$

$$\text{In this relation, } R' = 3mD/\rho r\delta V \quad (5)$$

Pore Diffusion Limitation

Equation 6 is derived by following the same reasoning and combining the mass balance with the second law of Fick.

$$C_t/C_0 = 1 - Kt^{1/2} \quad (6)$$

This represents an alternative expression of the well-known "parabolic" diffusion law in which the pore diffusion rate constant K is defined as:

$$K = 6m\alpha(D/\pi)^{1/2}/\rho Vr \quad (7)$$

The symbols employed are:

C = concentration	mg/L
D = diffusion constant	m ² /min
K = pore diffusion rate constant	min ^{-1/2}
m = mass of sorbent	kg
r = radius of sorbent particles	m
R = film diffusion rate constant	min ⁻¹
t = recirculation time	min
V = recirculating volume	m ³
α = distribution coefficient	—
δ = thickness of the film	m
ρ = density of the sorbent	kg/m ³
superscripts: liq = liquid, sor = sorbent	

MATERIALS AND METHODS

Materials

The sorbents employed were: Norit RBX-1, extruded granular activated charcoal, obtained from Norit (Amersfoort, Netherlands); Amberlite XAD-4, neutral polystyrene-based resin (0.3–1.0 mm), obtained from Serva (Heidelberg, Germany); Imac C-12,

strong acidic cation exchange resin (0.25–0.85 mm), donated by Akzo-Chemie (Amsterdam, Netherlands).

The coating materials employed were: cellulose acetate (40.1% acetyl) with an average molecular weight of 53,000 daltons, obtained from Fabelta (Tubize, Belgium, Type TV-20); polyelectrolyte synthesized from cis-1,4-polyisoprene;¹⁴ agarose powder (Type C) obtained from Pharmacia Fine Chemicals (Uppsala, Sweden), crosslinked after encapsulation with 1,3-dichloro-2-propanol (Aldrich, Beerse, Belgium).

Coating Techniques

Cellulose Acetate Coating

Activated charcoal was coated with CA (4 gm CA/kg charcoal) by the solvent evaporation technique, using a solution of CA in acetone (4 gm/L) as described by Tijssen.¹⁵ Amberlite XAD-4 was coated in a laboratory-scale Wurster coating device (Uni-Glatt, Haltingen, Germany). A solution in acetone (7 gm/L) was atomized onto the fluidized particles by a pneumatic nozzle to a total amount of 6 gm CA/kg XAD-4. Air-dried particles were rewetted in ethanol, which was replaced by water.¹⁰

Polyelectrolyte Coating

A net weight of 250 gm XAD-4 was placed in 150 ml of PLE solution (25 mg/ml) in phosphate-buffered saline (pH = 7.4). The adsorbed PLE (3 mg/gm) was crosslinked by means of gamma irradiation with a total dose of 4 Mrads as described previously.¹⁰

Encapsulation in Agarose Gel

The method used was based on the procedure described by Brunner et al.¹¹ with the following modifications. All sorbents were ground in a vibrating disc mill (Siebtechnik, Mühlheim/Rhur, Germany). Fractions able to pass a 140-mesh sieve were used for encapsulation. A suspension of 4% w/w of agarose powder was used with an amount of sorbent to yield a wet sorbent weight content of 33%. The fractions with a diameter from 0.85 to 3.35 mm were used for further experiments, unless otherwise indicated.

These capsules were crosslinked by reaction with 1,3-dichloro-2-propanol in a twofold molar excess with regard to agarose hydroxyl groups and an equimolar amount of NaOH with regard to dichloropropanol (DCP) at a concentration of 0.4 mol/L. A typical formulation was: agarose capsules, 200 gm; alkaline solution, 600 ml, 0.4 mol/L; DCP, 9 ml. The reaction was allowed to proceed for four hours at 50°C. A detailed description and the optimization will be presented elsewhere.¹² After crosslinking, the capsules were washed with water and stored wet.

Adsorption Kinetics

The rates of adsorption of test compounds with a molecular weight from 94 to 5200 daltons onto coated and uncoated sorbents were measured in a recirculating system at 37°C. In all cases, 1 liter of solution in phosphate-buffered saline (pH = 7.4, 0.02 mol KH_2PO_4 /L, 9 gm NaCl/L) was recirculated at a flow rate of 34 ml/min through a small column (17 mm in diameter) containing 6.25 gm of sorbent. When agarose capsules were tested, either the sorbent volume/recirculating volume ratio or the sorbent weight/recirculating volume ratio was kept equal to the ratio for the native sorbent by adjusting the recirculating volume.

Phenol ($C_0 = 150$ mg/L); creatinine (150 mg/L); methyl orange (50 mg/L); BSP (100 mg/L); vitamin B_{12} (50 mg/L); and inulin (100 mg/L) were used as test compounds. Samples were drawn over a total time of 180 minutes at 30-minute intervals. The concentrations of phenol, methyl orange, BSP (after adjusting the pH to 12), and vitamin B_{12} were determined photospectrometrically at wavelengths of 271 nm, 467 nm, 581 nm, and 363 nm, respectively. With creatinine the extinction was read at 490 nm after color reaction with picric acid, while inulin was determined at 635 nm after color reaction with diphenylamine and

FIG. 1. Film diffusion-limited adsorption of methyl orange onto activated charcoal. Native and CA-coated granular charcoal. Powdered charcoal encapsulated in agarose, equal volume (ag(v)) and equal weight (ag(w)). C_t/C_0 on a logarithmic scale versus time.

FIG. 2. Film diffusion-limited adsorption of BSP onto XAD-4. Native and CA- and PLE-coated bead-form resin. Powdered resin encapsulated in agarose, equal volume (ag(v)) and equal weight (ag(w)). $(C_t - C_\infty)/(C_0 - C_\infty)$ on a logarithmic scale versus time.

hydrochloric acid hydrolysis.¹⁵ The influence of flow rate and particle size on the adsorption kinetics was studied with methyl orange and vitamin B₁₂ onto activated charcoal encapsulated in agarose, using flow rates of 10, 34, and 70 ml/min and capsule diameters of 5.6–3.35 mm; 3.35–2.0 mm; 3.35–0.85 mm, and 2.0–0.85 mm.

Blood Compatibility

In vitro thrombocyte losses caused by the various coated and uncoated sorbents were measured in the recirculation system described by Chamuleau et al.⁸ Fresh human blood (250 ml) was recirculated through 100 gm of sorbent at a flow rate of 100 ml/min at a temperature of 37°C. The system was primed with physiological saline prior to use. From a solution of disodium citrate (3%) and glucose (2.5%), 65 ml were added to 435 ml of blood. The citrated blood was divided into two equal portions for simultaneous experiments. When heparin was used as the anticoagulant, the system was primed with 2500 U in 250 ml of phys-

FIG. 3. Pore diffusion-limited adsorption of vitamin B₁₂ onto activated charcoal. C_t/C_0 versus time on a square root scale.

iological saline for 30 minutes. In the blood a dose of 5000 U/L was employed. Empty columns were used for blank experiments.

RESULTS

Adsorption Kinetics

The adsorption kinetics of methyl orange onto coated and uncoated activated charcoal are presented in Figure 1. Since a linear relation is obtained between $\ln(C_t/C_0)$ and time, the rate of adsorption is limited by diffusional transport across the liquid boundary film. The concentration at infinite time approaches zero, thus the adsorption process can be characterized completely by the rate constant R' , which is the slope of the graph as indicated in Figure 1.

The same mechanism but with a concentration at infinite time C_∞/C_0 of 0.145 is observed for the adsorption of BSP onto coated and uncoated Amberlite XAD-4, as shown in Figure 2. A linear relation is ob-

TABLE I
CHARACTERISTICS OF ADSORPTION ONTO AMBERLITE XAD-4

Coating	Phenol			Methyl Orange			BSP			Vitamin B ₁₂			Inulin		
	Q_{3h}	R	C_∞/C_0	Q_{3h}	R'	K	Q_{3h}	R	C_∞/C_0	Q_{3h}	R	C_∞/C_0	Q_{3h}	R	C_∞/C_0
native	52.9	22.6	.647	48.4	19.2		67.3	1.25	.145	36.3	1.69	.155	18.4	4.06	.713
CA	52.9	22.4	.647	47.9	17.6		65.4	1.17	.145	28.0	.93	.155	7.7	1.24	.713
PLE	52.9	21.6	.647	47.5	16.6		49.2	.70	.145	28.6	.97	.155	7.0	1.10	.713
ag(v)	23.3	21.4	.845	18.3		27.9	33.7	1.15	.326	11.1	.88	.355	3.4	1.67	.881
ag(w)	52.9	21.4	.647	50.0		83.7	65.1	1.15	.145	27.0	.88	.155	9.9	1.67	.713

In this table, the total amount adsorbed after three hours (Q_{3h}) is expressed in mg. The rate constants R and R' are expressed in 10^{-3} min^{-1} . The rate constant K is expressed in $10^{-3} \text{ min}^{-1/2}$.

Native and CA- and PLE-coated bead-form XAD-4. Powdered XAD-4 encapsulated in agarose compared on the base of equal volume (ag(v)) and on the base of equal weight (ag(w)).

TABLE II
 CHARACTERISTICS OF ADSORPTION ONTO ACTIVATED CHARCOAL

Coating	Creatinine			Methyl Orange		BSP			C_{∞}/C_0	Vitamin B ₁₂		Inulin			C_{∞}/C_0
	Q_{3h}	K	R'	Q_{3h}	R'	Q_{3h}	K	R		Q_{3h}	K	Q_{3h}	K	R	
native	132.1	65.7		47.8	17.4	39.7	29.8			11.1	16.3	19.1	14.2		
CA	119.1	59.7		41.7	9.9	24.3	18.8			5.9	8.9	10.0	7.5		
ag(v)	24.4		0.96	38.3	7.9	34.0		3.71	.530	5.3	8.3	8.4		3.84	.849
ag(w)	74.2		3.79	50.0	31.3	73.9		3.71	.222	22.1	32.8	28.7		3.84	.588

In this table, the total amount adsorbed after three hours (Q_{3h}) is expressed in mg. The rate constants R' and R are expressed in 10^{-3} min^{-1} . The rate constant K is expressed in $10^{-3} \text{ min}^{-1/2}$. Native and CA-coated granular activated charcoal. Powdered charcoal encapsulated in agarose, compared on the base of equal volume (ag(v)) and equal weight (ag(w)).

tained between $\ln\{(C_t - C_{\infty})/(C_0 - C_{\infty})\}$ and time. In this case the adsorption rate is characterized completely by the constants R and $C_0/C_{\infty} = S$. The slope in the graph refers to the product of R and S , as indicated in Figure 2.

A completely different situation is met in the adsorption of vitamin B₁₂ onto activated charcoal (Fig. 3). A linear relation is observed between the concentration C_t/C_0 and the square root of time. This indicates that the rate of adsorption is limited by diffusional transport through the pores of the sorbent. The adsorption kinetics are completely characterized by the rate constant K , which is the slope of the graph as indicated in Figure 3.

All data obtained are presented in Table I for the adsorption onto Amberlite XAD-4, and in Table II for the adsorption onto activated charcoal. The appropriate rate constants R' , R , and C_{∞}/C_0 , and K are presented. In all cases the 99% confidence limit for the rate constants is of the order of 5%. All measurements

showed correlation coefficients of 0.998 or better. In addition the total amount of test compound adsorbed within three hours (Q_{3h}) is presented. No adsorption was found of creatinine onto Amberlite XAD-4.

The permeability of the coatings for the various test compounds, expressed as the ratio of the total amount adsorbed after three hours over the total amount adsorbed onto the uncoated native sorbent in the same time ($Q_{3h}/Q_{3h,\text{native}}$) as a function of molecular weight, is presented in Figure 4 for the adsorption onto XAD-4. A sudden decrease is observed when the molecular weight is over 1000 daltons. No large deviations are observed for the various coatings. Only agarose capsules, compared on the basis of volume (ag(v)), show a decreased adsorption.

Figure 5 shows the same relation for adsorption onto activated charcoal. The decline in $Q_{3h}/Q_{3h,\text{native}}$

FIG. 4. Effect of coating on the relative adsorbed amounts after three hours onto Amberlite XAD-4 as a function of molecular weight. Native and CA- and PLE-coated bead-form resin; powdered resin encapsulated in agarose.

FIG. 5. Effect of coating on the relative adsorbed amounts after three hours onto activated charcoal as a function of molecular weight. Native and CA-coated granular charcoal and powdered charcoal encapsulated in agarose.

FIG. 6. Effect of capsule radius on the film diffusion rate constant R' of methyl orange (○) and on the pore diffusion rate constant K of vitamin B_{12} (●) onto activated charcoal encapsulated in agarose.

FIG. 7. Effect of flow rate on the film diffusion rate constant R' of methyl orange (○) and of the pore diffusion rate constant K of vitamin B_{12} (●) onto activated charcoal encapsulated in agarose.

for the CA coating occurs at a lower molecular weight, compared to CA-coated XAD-4. Another striking observation is that agarose capsules, compared on the base of volume, show an adsorption comparable to CA-coated charcoal. When equal charcoal weights are compared (ag(w)), an increase in adsorption is observed.

The effect of agarose capsule size on the adsorption rate onto activated charcoal for a typical film diffusion-limited compound (methyl orange) and a typical pore diffusion-limited compound (vitamin B_{12}) is shown in Figure 6. Both the film diffusion rate constant R' and the pore diffusion rate constant K appear to be inversely proportional to the mean capsule radius. The effect of flow rate, using the same com-

pounds, is presented in Figure 7. The film diffusion rate constant R' appears to be proportional to the flow rate. On the contrary, no significant influence of flow rate can be observed on the pore diffusion rate constant K .

Blood Compatibility

The in vitro thrombocyte losses caused by the various coated and uncoated sorbents are presented in Table III. The very preliminary results for most of the agarose capsules are presented only for the sake of completeness. It is obvious that coating has a beneficial effect on blood compatibility. In all cases the platelet losses with heparinized blood are much more pronounced than with citrate as anticoagulant.

TABLE III
IN VITRO BLOOD COMPATIBILITY OF SORBENTS

Sorbent	<i>n</i>	Citratated Blood			<i>n</i>	Heparinized Blood		
		60 min	120 min	180 min		60 min	120 min	180 min
blank	5	91 (8)	85 (9)	85 (15)	8	87 (5)	78 (11)	70 (15)
charcoal								
native	4	46 (17)	48 (17)	49 (19)	6	29 (13)	25 (12)	20 (13)
CA	4	81 (15)	73 (12)	71 (15)	8	48 (8)	42 (17)	39 (17)
agarose	2	87 (1)	90 (1)	86 (2)	1	61	52	46
XAD-4								
native	4	15 (12)	19 (16)	20 (15)	4	15 (8)	21 (10)	18 (13)
CA	4	70 (16)	63 (18)	56 (19)	5	58 (19)	49 (18)	35 (16)
PLE	8	81 (12)	73 (12)	72 (16)	7	25 (13)	27 (11)	27 (12)
agarose	1	64	70	58	1	57	38	23
Imac C-12								
native	1				1	41	34	27
agarose	4				4	57 (16)	35 (20)	27 (9)

Percentage of initial count after 60, 120, 180 minutes of recirculation time; mean (SD) Citratated and heparinized blood.

DISCUSSION

From the preparative point of view, the methods employed for the coating of the various sorbents form a satisfying set of techniques which can all be scaled up for industrial production. The CA-coated activated charcoal is already manufactured by Organon Teknika for use in the Hemopur 260 hemoperfusion cartridge.⁹ The completeness of the coatings and the methods used to investigate this have been published previously.¹⁰ The crosslinking of agarose capsules is a major improvement because the sterilization problem has been solved since autoclaving at 122°C is possible. In addition the mechanical strength is increased by a factor of 4. This implies that the risk of release of microparticles, generated by mechanical causes, is greatly reduced.¹²

The conditions of the *in vitro* adsorption rate experiments are directly comparable to the clinical situation, since both the recirculating volume and the amount of sorbent were equally adapted by scaling down normal hemoperfusion conditions by a factor of 40. The flow rate of 34 ml/min gives rise to the same linear flow velocity (in m/s) as is obtained with a flow rate of 250 ml/min using the Hemopur 260 column, thus operating at the same hydrodynamic conditions. This is important as flow rate can influence the adsorption rate (Fig. 7).

The adsorption kinetic theory presented in this paper describes the process of adsorption as measured in our recirculation system with remarkable accuracy. As can be observed from Equations 2, 5, and 7, the theory predicts an inverse proportionality between the radius of the sorbent particles and the appropriate rate constant R' , R , or K . Figure 6 shows that this prediction turns out to be correct. Furthermore it is predicted that, in the case of film diffusion limitation, the rate constants R' and R are inversely proportional to the "thickness" δ of the stagnant fluid film. Since δ is inversely proportional to the linear flow velocity,¹³ the rate constants should be proportional to the flow rate. Because δ does not appear in the formulation of K (Eq. 7), no influence of flow rate is predicted in the pore diffusion limitation mechanism. This turns out to be correct as well, as can be concluded from Figure 7.

In general, film diffusion limitation is the slowest process. It can be compensated for by increasing the flow rate, until pore diffusion limitation becomes the rate-limiting process. With Amberlite XAD-4, only film diffusion limitation was observed (Table I), with the exception of the adsorption of methyl orange onto agarose capsules. Therefore only minimal effects of the coating on the adsorption rate of low molecular weight compounds have been observed. Middle molec-

ular weight compounds (from 1000 daltons) show reduced adsorption rates. This is probably caused by concentration polarization. Upon grinding the resin and encapsulation in agarose, the hydrodynamic diameter increases when compared to the native resin, while the sorbent content reduces by a factor of 3. Therefore both constants R and S decrease. This results in a decreased adsorption rate when agarose capsules are compared on the base of volume. From Figure 4, it can be concluded that grinding of Amberlite XAD-4 to a fine powder and encapsulation in agarose offers no advantages with regard to adsorption kinetics, but merely a disadvantage because larger amounts of capsules are required to obtain the same capacity. Therefore larger columns are also needed.

A completely different situation is encountered in the adsorption onto activated charcoal. With the exception of methyl orange, all compounds studied showed a pore diffusion-limited adsorption rate onto the granular charcoal (Table II). The reduction in adsorption ($Q_{3h}/Q_{3h,native}$) caused by the CA coating is more pronounced, although not dramatically, in the low molecular weight region when compared to CA-coated XAD-4, this notwithstanding the fact that the latter coating is 100 times as thick (3 nm for charcoal versus 400 nm for XAD-4). Grinding of the charcoal, thus offering shorter diffusion routes through the charcoal, results in an increase in adsorption rate, especially when the same amounts of sorbent are compared. In several cases this is accompanied by a change in mechanism to film diffusion limitation (Table II). It is concluded that grinding of activated charcoal to a powder and encapsulation in agarose offers advantages with regard to adsorption kinetics. This advantage will be even more pronounced when the flow rate in clinical use can be increased to 500 ml/min as was proposed by Brunner.¹¹ The compounds used in this adsorption study are only model compounds. No significant differences in adsorption rate were measured for the adsorption of amino acids, bile acids, fatty acids, and octopamine onto coated and uncoated activated charcoal and Amberlite XAD-4.^{8,10}

Coating with CA markedly improves the blood compatibility of activated charcoal and XAD-4, with both heparin and citrate as anticoagulant. Encapsulation in agarose tends to show an improvement even when activated charcoal is encapsulated, but not when XAD-4 or cation exchange resin is encapsulated. The number of experiments however is too small to draw final conclusions from the agarose measurements as yet. Even coating with PLE appears to be an improvement compared to CA when citrated blood is used. With heparin the sequence is reversed, a phenomenon which is difficult to explain. In agreement with the

results published by several authors,¹⁶ use of citrate as anticoagulant resulted in a reduced loss of thrombocytes as compared to heparin. The thrombocyte losses in these in vitro experiments are naturally very high as the conditions of experiment are quite drastic. The amount of sorberent is five times the amount used in normal clinical conditions. The blood is recirculated approximately 75 times through the column in every experiment.

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