ventricle. pulmonary conus or the proximal part of the pulmonary artery, no evidence of A-V shunts was ever seen with the injection of Thorotrast. However, with deliberate "jamming" of the catheter into a branch of the pulmonary artery, A-V shunts were seen in 3 of 4 dogs. It is suggested that the irritation produced by the catheter may cause the pulmonary vessels to constrict around the catheter as well as peripherally and thus force the injection material through A-V shunts. Another explanation is that this irritation may cause the A-V shunts to open without need of greater filling pressure.

Summary. A shunt between the pulmonary artery and vein was demonstrated in 3 of 4 dogs by injecting Thorotrast into a catheter inserted tightly into the pulmonary artery and following the circulation by cinefluorography. In one of these dogs the passage of glass spheres 200 $\pm$ 25 μ in diameter, through a cannula in the pulmonary artery, to and through a cannula in the aorta, together with the location of the shunt from plastic casts of the pulmonary vessels, help to confirm the presence of the shunt.

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Absorption Rates of Colloid and Milky Suspensions of Halogenated Fats for Hepatosplenography in the Rat. (19582)

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Success with intravenous injections of emulsified iodized oils for hepatosplenography has not been achieved, partially because of the pyrogenic effect of the fat and toxicity of the halogens(1). This report concerns attempts to lessen these toxic effects by the use of injection methods other than by the intravenous route.

Experimental. Milky and colloid suspensions of the following halogenated fats were used in these experiments: 40% iodine in poppy seed oil (Lipiodol LaFay), 27% iodine in peanut oil (Iodochloral Searle), bromiodized linseed oil, iodized linseed oil, brominized linseed oil, bromiodized lecithin and hexabromostearic acid. Owing to the fact that the reaction between the halogen and the nonsaponifiable portion of fat seems to produce toxic substances, recrystallized hexabromostearic acid was tried. This was prepared by dissolving 2 g (M.P. 179-180°C) in 10 ml of boiling dioxane which was then injected as a capillary stream into 15 ml boiling distilled water rotated in a micro Waring blendor. The resulting sol was bluish by reflected and reddish by transmitted light. After stabilizing with 1% polyoxyethylene sorbitan-fatty esters (Tween 80), it was dialyzed free of

dioxane. In other experiments, Lipiodol as well as bromiodized and brominized oils were emulsified with 4 volumes 1% Tween 80 in a 2-stage recirculating homogenizer at a pressure of 3000 lbs/sq in. These emulsions contained droplets of 1 to 3 μ and probably invisible particles of colloid size. In some experiments, we used colloid sols of Lipiodol showing the Tyndall phenomenon, obtained by using larger amounts and higher concentrations of Tween 80.

Most of the emulsions used in these experiments were injected either into the subcutaneous, muscular, perivenous lymphatic spaces, or stomach. In some experiments, a combination of these injections with the intravenous route was tried. For the perivenous lymphatic injections, the loose areolar tissues around such large veins as the jugular or femoral veins were selected. Absorption from these spaces was watched in preliminary experiments by using emulsions of corn oil stained with Sudan Black B. By sacrificing animals at various periods, these emulsions could be traced into the regional lymph nodes and then into the general circulation at 4 hours to 5 days after injection. A few experiments were done after intrapleural and intraperitoneal injections. An ampule of hyaluronidase 150 TR units (Widase) was mixed with each 10 ml of emulsion to speed absorption of all the subcutaneous and intramuscular injections. In the emulsions given by stomach tube, 10 mg procaine were added to open the pylorus. Rats weighing 200 g were used in all experiments.

Results. Successful hepatosplenography was not obtained with a single dose of the tested emulsions given subcutaneously, intramuscularly or by stomach tube alone. A slight accentuation of the liver and spleen shadows was obtained if the emulsions were given repeatedly by stomach tube. The absorption of halogenated sols from the pleural and peritoneal cavities was too slow to be of any value(1). In Table I are recorded the results obtained with the intralymphatic and intragastric injections alone and combined with intravenous injection.

Successful hepatosplenography with any emulsion was never obtained with less than 3 ml. With this dose, the hepatic and splenic shadows were well intensified within 1 to 2 hours after intravenous injection. These shadows faded within 5 to 7 days; but if then another injection were given either by the gastric or intralymphatic routes, the hepatic and splenic shadows again were potentiated, in spite of the inadequacy of the intragastric or intralymphatic dose given alone.

TABLE I. Hepatosplenography with Various Iodized, Bromiodized and Brominized 20% Emulsions in Tween 80 after Intralymphatic and/or Intragastric Injections or Combined with Intravenous Injections (in ml Doses).

No. of rats	Intralymphatic	Intragastric	Intravenous	Hepatosplenography
5	12	—		—
3*	—	40		+
5	3	5		—
50			3	++
5			1.5	—
5		5	1.5	+
5	3	5	1.5	++
3†		8	3	++

Accentuation of liver or spleen x-ray shadow: — none; + slight; ++ moderate.

* 10 ml doses at 9 a.m. and 3 p.m. for 2 days.

† Intravenous dose 5 days before intragastric dose.

With combinations of different routes of injections, it was found that good hepatosplenography was obtained 5 hours after intralymphatic and 24 to 30 hours after intragastric if an intravenous injection were given one hour before x-ray studies. Dibromindigo appears in the urine after intragastric injection of hexabromostearic acid, suggesting that the halogen is partially split off in the alimentary tract.

Discussion. The rapid subcutaneous absorption of fat emulsions observed by Shafiroff (2) seems not to apply to halogenated fats for purposes of splenography. Nor were we able to obtain the results of Olsson and Ekman(3) with single intragastric injections. The combination of routes with the intravenous injection, in spite of good results with smaller doses, does not seem practicable for use in the human. No studies of the storage capacity of our emulsions were done, but the darkening of halogenated fats in light is apparently hastened by the non-saponifiable portion and the emulsifier.

Boiling emulsions for sterilization was never

done without an increase in size or aggregation of droplets; but boiling for sterilization was found to be not necessary. Tests with broth and blood agar plate cultures for bacteria and Littman plate cultures for molds were found negative. Apparently, the halogen acts as a sterilizing agent.

Summary. 1. A colloid sol of hexabromostearic acid was used successfully for hepatosplenography in the rat. This sol could be given intravenously or by repeated intragastric injections. Dibromindigo appeared in the urine after intragastric injection. 2. Half the minimum intravenous dose of several halogenated fat emulsions for hepatosplenography was effective when reinforced with injections by the intralymphatic and intragastric routes, but the total dose was greater than the minimum intravenous dose alone. 3. The emulsions were broken by boiling but this method of sterilization was not necessary since it was found that the halogens acted as sterilizing agents.

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Combination of Hydrochloric Acid and Sodium Hydroxide with Human Aortic Albuminoid.* (19583)

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Recent studies have revealed that the albuminoid (collagen and elastin) fraction of the human aorta undergoes significant quantitative aging and pathological changes. Though the percentage of elastin in this albuminoid fraction increases significantly only in markedly arteriosclerotic aortae(1), notable increases in the calcium content of aortic elastin have been demonstrated with progressive age(2). Likewise, important quantitative aging changes in the amino acid composition of elastin have been shown(3). It seemed of value, therefore, to investigate the acidic and basic properties of the human aortic albuminoid as part of an overall attempt to more fully evaluate the underlying pathological changes in this tissue.

Materials and methods. The albuminoids were prepared by previously described methods(4) with the exception that sodium chloride and dibasic sodium phosphate soakings were substituted for the calcium hydroxide treatments. The refrigerated tissues, subsequent to acetone and alcohol extractions, were soaked in 3 changes of 10% sodium chloride for a minimum of 24 hours during each extraction period. They were then treated with M/15 dibasic sodium phosphate for 2 soaking periods of at least 24 hours in the refrigerator. After five 24-hour washings in distilled water, the samples were dehydrated with several changes of absolute alcohol, dried in an oven at approximately 70°C, and desiccated for at least 24 hours. The 29 albuminoid specimens were each subdivided into 2 approximately equal samples ranging from 0.4 g to 2.6 g in weight. One aliquot of each tissue was treated with 30 ml of .10 N hydrochloric acid in saturated sodium chloride (25°C) per g of albuminoid, while the other fraction was soaked in 30 ml per g of tissue of .11 N sodium hydroxide in saturated sodium chloride (25°C) according to the method of Beek(5). After standing for 24 hours with occasional shaking,

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