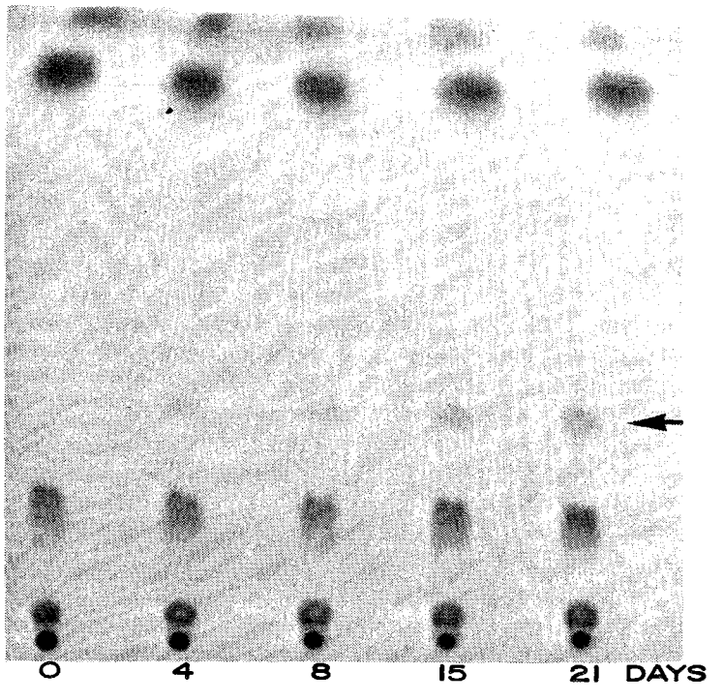

Thin-layer chromatogram demonstrating the presence of dihexyl phthalate in blood stored for different periods in plastic packs.

Arrow indicates dihexyl phthalate.

In order to determine for certain whether the dihexyl phthalate in lipid extracts of plasma came from the transfusion packs, the following experiment was performed. 1250 ml. of blood from compatible donors was collected in heparinised glass bottles and pooled. 250 ml. fractions were transferred to commercial plastic transfusion packs and kept in these at 4°C for periods of 4, 8, 15, and 21 days, while a 250 ml. fraction was stowed away in a glass bottle. At the end of the storage period each aliquot of blood was centrifuged and the plasma extracted with chloroform/methanol mixtures according to the method of Folch et al.[7]

The dihexyl phthalate content of the lipid extracts was enriched by chromatography on thick layers of silica-gel H,[8] and final purification was achieved by thin-layer chromatography on the same medium.[9] The concentrations of dihexyl phthalate (mg. per 100 ml. of plasma) were 0, 4, 7, 11·5, and 11·5 for storage periods in plastic packs of 0, 4, 8, 15, and 21 days. Such concentrations are high enough to allow dihexyl phthalate to be demonstrated by thin-layer chromatography of the total lipid extract on silica gel-H, developed with the solvent system hexane/diethyl-ether/acetic acid (90/10/1), as shown in the accompanying figure. This chromatogram shows the total lipid extracts of plasma following storage of the blood in plastic packs for 0, 4, 8, 15, and 21 days.

The toxicity of analogous plasticisers, such as dibutyl phthalate, has been investigated by Smith[10] in the rat. In this study, dibutyl phthalate was administered orally, intraperitoneally, and subcutaneously in various doses and for varied periods of time. The toxicity was found to be moderate, but high doses could be lethal. Because of the dissolution of phthalates in plasma during storage in plastic transfusion packs, the use of such blood or plasma for large-volume transfusions may be inadvisable.

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ORAL CONTRACEPTIVES AND LOW ANTITHROMBIN-III ACTIVITY

SIR,—Two independent investigations have shown that hereditary deficiency of antithrombin-III activity predisposes to thromboembolic disease in both males and females.[1,2] It is also known that patients with thrombosis and embolism often have low antithrombin-III activity[2] and that in individuals with repeated episodes of this type of intravascular clotting the antithrombin-III values tend to stay low over long periods of time. Low antithrombin-III activity is also found in severe liver disease. So far as we were able to determine, the antithrombin-III activity has not been assessed in any of the numerous coagulation studies which have tried to elucidate the possible correlation between the use of oral contraceptives and thromboembolism. Without specifically studying this question, we were struck by the observation that women using oral contraceptives have very low antithrombin-III values. The number of observations is small, but in view of the pronounced alteration of antithrombin-III activity in these women we feel justified in calling attention to our results at this early stage.

In our hands, antithrombin-III activity is expressed as antithrombin-III-time, with a short time indicating low antithrombin-III activity. The average value for normal individuals is 38·5 seconds, compared with average values in patients with thromboembolism of 18·5 seconds. Surprisingly, the average value in eleven women on oral contraceptives was only 17·7 seconds. Two of these women, found to have extremely low values of 13·8 and 14·9 seconds (indicating the near absence of antithrombin-III activity) subsequently developed thrombosis and embolism. If the oral contraceptives indeed suppress antithrombin-III activity, as these preliminary figures suggest, then they would certainly create an imbalance of the clotting system favouring intravascular clotting. This would be particularly true if additional events, such as a temporary rise of the platelet-count or of the activity of clotting factors, further increased the imbalance. A pre-existing reduced antithrombin-III activity, then further decreased by the Pill, is another possible explanation of the precipitation of intravascular clotting. One patient who tried oral contraception twice, had an episode of bilateral thrombophlebitis at each attempt. When first seen several weeks after withdrawal of the drug, the antithrombin-III time was only 16·8 seconds. In retrospect it would seem that this patient, who probably had a low antithrombin-III activity initially, should have never been started on the Pill.

The role of antithrombin-III or serum-antithrombin is not yet established beyond all doubt; the removal of thrombin traces from the circulation and/or a role as a heparin cofactor are the most likely functions of this normally occurring inhibitor. Antithrombin-III activity, in our opinion, must be included in any further investigation into the effect of oral contraceptives on human blood coagulation. Such studies are incomplete if the naturally occurring inhibitors are not tested.

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CADMIUM

SIR,—Your annotation (Dec. 20, p. 1346) says that the American Conference of Governmental Industrial Hygienists (A.C.G.I.H.) suggests a "maximal allowable concentration (M.A.C.) of 100 microgrammes per cubic metre of environmental air". The 1969 recommendation of the A.C.G.I.H. is in fact for a *threshold limit value* (T.L.V.) of

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