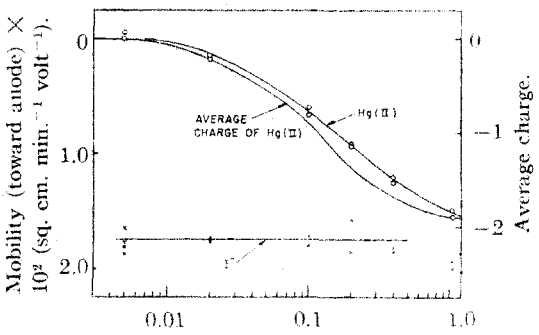

Fig. 1.—Filter paper electromigration of ions.

Hg(II) was practically zero at low M Cl^- and increased rapidly in the region 0.1 to 1 M Cl^-. The mobility of Hg(II) is roughly paralleled by the calculated values of the average charge of the Hg(II) ions using the data of Sillén.[3] Thus, if data on the relative mobility of complex ions of different charge can be obtained, this technique may become generally applicable for rapid determination of estimates of stability constants of complex ions.

Acknowledgment.—We are indebted to Dr. H. H. Shain for a number of valuable discussions and for informing us of the details of his work on the continuous separation of ions by filter-paper electromigration.

(3) L. G. Sillén, *Acta Chem. Scand.*, **3**, 539 (1949).

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RECEIVED JULY 20, 1950

PAPER CHROMATOGRAPHY OF STEROIDS[1]

Sir:

The separation of cholesterol and cholestenone has been achieved by using paper impregnated with "Quilon"[2] as the stationary phase and simple primary alcohols as solvents. In effect, the stationary phase consists of the stearic acid residues.

Use of paper impregnated with rubber latex,[3] silicic acid[4] and alumina[5] in paper chromatography has been reported. Of these, alumina paper was tried and found to give erratic results. The method of Zaffaroni and co-workers[6] for the paper chromatography of steroids using paper saturated with formamide or propylene glycol as the stationary phase and a hydrocarbon solvent was also tried. In these experiments the steroids were found to move with the front. Using ordinary paper (Whatman No. 1), cholesterol was found either to move with the solvent front or remain at the origin.

(1) The work described in this paper was sponsored by the Atomic Energy Commission.

(2) Stearato chromic chloride, generously supplied by E. I. du Pont de Nemours and Co., Inc.

(3) Boldingh, *Experientia*, **4**, 270 (1948).

(4) Kirchner and Keller, THIS JOURNAL, **72**, 1867 (1950).

(5) Datta and Overell, *Biochem. J.*, **44**, xliii (1949).

(6) Zaffaroni, Burton and Keutmann, *Science*, **111**, 6 (1950).

For ease of location, tritiated cholesterol was used and the material located by scanning the paper with a windowless counting tube designed to locate weakly radiating substances on paper.[7]

The presence of cholesterol at the points of high activity was confirmed by the red color developed after papers treated with a solution of silicotungstic acid were dried.[8] Cholestenone gave an olive green color with this reagent, but only when the steroid was present in relatively large amounts. Cholestenone was most easily detected by the yellow color obtained with a reagent consisting of a solution of iodine and potassium iodide in water.[9]

The most satisfactory solvents, to date, have been methanol, ethanol and ethanol–water 8:2. The latter solvent gives the best separation of cholesterol and cholestenone. The results are collected in Table I.

TABLE I

Solvent	Cholesterol (R_f)	Cholestenone (R_f)
Methanol	0.56	0.77
Ethanol	.92	.97
80% Ethanol	.52	.86

All experiments were carried out as descending chromatograms using 1.5 × 15 inch strips of the impregnated paper. The paper was usually wet to a distance of about 25 cm. from the origin. R_f values were measured from the farthest point of the origin and the foremost point of the colored or active zone.

Projected work includes widening the range of usable solvents, development of supplementary color reactions and extension of this method to other steroids.

(7) Gray, Ikeda, Benson and Kritchevsky, *Rev. Sci. Inst.*, in press.

(8) Montignie, *Bull. soc. chim.*, **51**, 690 (1932).

(9) Munier and Macheboeuf, *Bull. soc. chim. Biol.*, **31**, 1144 (1949).

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RECEIVED JULY 31, 1950

COLCHICINE. NATURE OF THE B-RING[1,2]

Sir:

The structure of deaminocolchinol methyl ether has been established by J. W. Cook[3,4] as 9,12,13,14-tetramethoxy-3,4,5,6-dibenzocycloheptatriene-1,3,5. This compound, together with isodeaminocolchinol methyl ether, may be ob-

(1) The work carried out at the University of Pennsylvania was aided by a Grant-in-Aid from the American Cancer Society recommended by the Committee on Growth of the National Research Council.

(2) This investigation was supported (in part) by a research grant from the National Cancer Institute, of the National Institutes of Health, Public Health Service.

(3) Barton, Cook and Loudon, *J. Chem. Soc.*, 176 (1945).

(4) Buchanan, Cook, Loudon and MacMillan, *Nature*, **162**, 692 (1948).