

The Semi-quantitative Determination of Cholesterol and Cholesteryl Esters by Paper Chromatography

In previous papers we described the one-dimensional separation of cholesterol and its esters¹. We mentioned the possibility of the semi-quantitative determination. Further details of this procedure are given here:

To 1 ml of blood serum 9 ml of ethanol-ether mixture 3:1 (v/v) were added. After 10 min standing, the mixture was centrifuged and 5 ml of clear supernatant were evaporated to dryness in a test tube. The residue having been extracted with 0.1 ml of chloroform, 0.04 ml of this solution were spotted on Whatman No. 3 paper impregnated with paraffin oil². The mixture of acetic acid-chloroform-paraffin oil 65:25:10 (v/v/v) was used as a mobile phase. After 12–15 h of ascending run, the chromatogram was dried and cut into strips 1 cm wide and these eluted with 3 ml of chloroform for 5–6 h at room temperature, in test tubes with occasional stirring. Then 1 ml of acetic anhydride and 0.1 ml of concentrated sulphuric acid were added and the density of the coloured solution was measured at 610 m μ after 10 min.

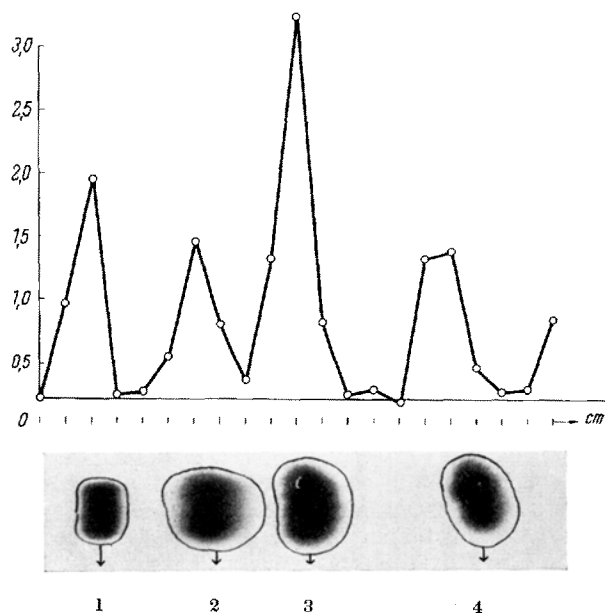


Fig. 1. — Mixture of synthetic cholesteryl esters. 1 Cholesteryl oleate; 2 Cholesteryl caproate; 3 Cholesteryl formate and acetate; 4 Cholesterol.

The calibration curve was prepared from the standard solution of cholesterol. The calculation was carried out by addition of all extinction values of every individual spot and subtraction of the blanks and then multiplication with the factor calculated from the calibration curve.

For the calculation of only relative percentage, it is possible to weigh the individual pieces of paper obtained by cutting out the individual peaks of the graph representing relation between the extinction values and the distances in cm.

The results are in good agreement with those obtained by other methods.

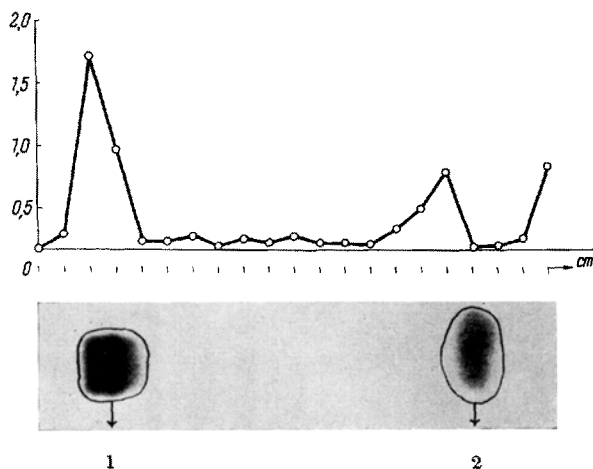


Fig. 2. — Human blood serum. 1 Cholesteryl esters with higher fatty acids (oleate, palmitate, linoleate); 2 Free cholesterol.

The present method is simple and permits a comparatively rapid determination of free and esterified cholesterol in blood serum and other biological fluids and organs.

Č. MICHALEK, V. JIRGL, and
J. PODZIMEK

Central Biochemical Laboratories of the University Hospital, Prague (Czechoslovakia), February 8, 1957.

Zusammenfassung

Eine papierchromatographische Methode zur semi-quantitativen Bestimmung von Cholesterin und Cholesterinester in biologischem Material wird beschrieben.

Application vaginale de cystamine et Radio-Protection locale

I. *Introduction.* Dans un travail antérieur publié dans cette même revue, l'un de nous¹ a pu montrer que l'administration de Becaptan par voie générale protège la muqueuse vaginale de la rate contre les effets d'une irradiation locale.

Les travaux expérimentaux consacrés aux radio-protecteurs en général et au Becaptan en particulier ont été le plus souvent réalisés en administrant la substance protectrice par voie générale; l'effet est alors étudié soit sur l'organisme *in toto* (courbes de mortalité, chute de poids) soit au niveau d'un tissu déterminé (ovaire, intestin grêle, foie, rate, thymus, lignée rouge, leucocytes, muqueuse vaginale etc.).

Peu de recherches ont été consacrées à l'action protectrice de l'application locale d'une substance déterminée au niveau de l'organe à protéger. Nous pouvons cependant citer:

¹ Č. MICHALEK, *Naturwissenschaften* 42, 509 (1955).

² Č. MICHALEK, *Biochim. biophys. Acta* 19, 187 (1956).

¹ L. DARCIS, P. HOTTERBEEX et C. ONKELINX, *Exper.* 12, 286 (1956).