fatto di fenolo/acqua, 80/20, (*p/v*), e per la seconda corsa (10 h) etanolo/NH_3/H_2O[9],60/20/10, o più raramente collidina/lutidina/acqua, 100/100/100. Sulla carta sono state individuate: la fosforilcolina, la fosforiletanolamina, la fosforilserina, la L-α-glicerilfosforilcolina, e la L-α-glicerilfosforiletanolamina.

La frazione *C* era ridisciolta in 0,2–0,3 ml di H_2O; da essa erano prelevati 0,005–0,008 ml e depositati su carta per cromatografia S. S. 2045/a precedentemente lavata con 2*N* ac. acetico. Per la prima corsa (10 h) è stato usato il solvente fatto di acetone/0,005 *N* ac. acetico, 50/50, e per la seconda corsa (20 h) il solvente di KREBS e HEMS[10] senza versene (ac. isobutirrico/*N* NH_3, 100/60). Sulla carta è stata finora identificata, fra gli altri nucleotidi, la citidindifosfatocolina.

I dati ottenuti possono essere così riassunti:

1° È stato possibile separare contemporaneamente da piccole quantità di tessuto nervoso (340–610 mg) un notevole numero di corpi fosforati liberi (alcuni già conosciuti, altri poco o non conosciuti), i quali partecipano verosimilmente al metabolismo di alcuni fosfatidi in qualità di precursori dei medesimi od a vicenda di prodotti di loro degradazione.

2° Essi sono: la fosforiletanolamina e la L-α-glicerilfosforiletanolamina (già conosciuti)[11], la fosforilcolina e la L-α-glicerilfosforilcolina (poco conosciuti)[12], la fosforilserina e la citidindifosfatocolina (non conosciuti). Inoltre sono state separate dallo stesso campione di tessuto la fosfatidil-colina, -etanolamina e -serina.

3° Tutto lascia supporre che altri corpi fosforati, specificatamente la glicerilfosforilserina, la citidindifosfato-etanolamina e -serina ancora non identificati siano presenti sui cromatogrammi.

4° Sulla base di prove collaterali sembra che il metodo di estrazione e di frazionamento dell'estratto acquoso (frazioni *B* e *C*) non alteri, nei limiti dell'errore sperimentale, qualitativamente e quantitativamente i composti fosforati ricercati.

5° La concentrazione media (76 animali) di alcuni composti fosforati nell'encefalo di ratti compresi fra il 3° ed il 13° giorno di vita è così risultata: fosforilcolina 76 ± 13 μg P/g di tessuto fresco; fosforiletanolamina 91 ± 7 μg P/g; fosforilserina 29 ± 8 μg P/g; L-α-glicerilfosforilcolina 42 ± 14 μg P/g; L-α-glicerilfosforiletanolamina 32,4 ± 11 μg P/g. Nel calcolo non è stato tenuto conto delle perdite che ciascun composto subisce durante la separazione cromatografica su carta.

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Summary

A method is described whereby the phosphorylcholine, -ethanolamine, -serine, the L-α-glycerylphosphoryl-choline, -ethanolamine, the phosphatidyl-choline, -ethanolamine, -serine and also the cytidinediphosphatecholine may be separated in the brain of a rat a few days old (340–610 mg).

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PRO EXPERIMENTIS

Electrophoretic Analysis of Clinical Dextrans

The clinical value of dextran depends on its molecular weight. The mean molecular weight, however, is a very unreliable index of the therapeutic properties unless supplemented by the analysis of the polydispersity of the dextran preparation.

We have recently shown that dextran dissolved in a borate buffer exhibits electrophoretic mobility and that a mixture of two homogeneous dextran fractions of different molecular weight can be partitioned in the electric field with a good recovery of each component[1].

It appears that electrophoresis of dextran may afford means of estimating both the mean molecular weight and the polydispersity of a dextran preparation.

Materials and Methods.—*Electrophoresis* was carried out in a Fokal-B apparatus (Strübin & Co., Basle) at + 2·5°C, using white light and a standard 85 mm Tiselius type cuvette[2]. The percentage composition of a mixture was evaluated by fitting Gaussian curves to the enlarged patterns.

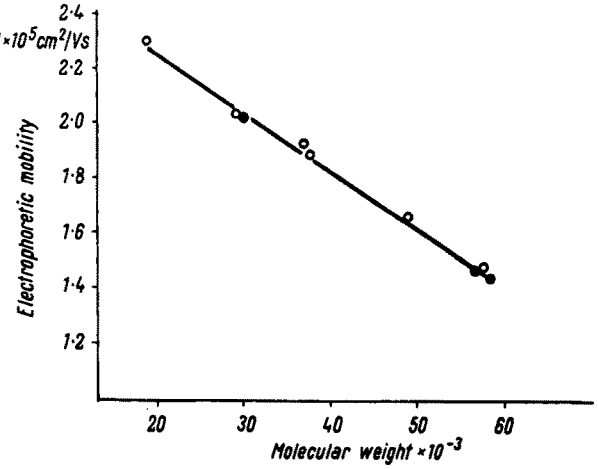

Fig. 1.

Borate buffer was prepared by adjusting a boric acid solution to pH 10·0 (glass electrode) with sodium hydroxide; the final concentration of borate ion was 0·05 *M* in all experiments.

Molecular weights were calculated from the combined diffusion constant, intrinsic viscosity and partial specific volume determinations[3].

Purified dextran fractions were obtained from acid-hydrolysed native dextran by repeated ethanol fractionation. The final lyophilised preparations were dissolved in the borate buffer and exhaustively dialysed against the same buffer.

Results.—Electrophoretic mobilities of purified dextran fractions were determined; the mean values, calculated from ascending and descending boundaries, are plotted against molecular weights of the fractions as shown in Figure 1. The data on 'Macrodex'-fractions, kindly donated by Pharmacia A.G., Uppsala, Sweden, are included in this graph and marked with crosses. It is evident that electrophoretic mobility is linearly de-

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Preparation	Intrinsic viscosity +25°, 0·16 *M* NaCl	Electrophoresis results						Pattern in Figure 2
		Component I			Component II			
		mobility $U \cdot 10^5$ cm²/V·s	molecular weight $\times 10^{-3}$	%	mobility $U \cdot 10^5$ cm²/V·s	molecular weight $\times 10^{-3}$	%	
‚Intradex' Benger Labs. England	0·22	1·14	> 60·0	21·8	1·76	46·6	78·2	*a*
‚Dextraven' Benger Labs. England; Nr. 18055 .	0·26	0·99	⩾60·0	65·0	1·56	53·8	35·0	*b*
‚Macrodex' Pharmacia AG., Sweden; A7673A .	0·24	1·41	61·0	79·2	2·36	16·0	20·8	*c*
‚Expandex' Com. Solv. Corp., U.S.A. Nr. 36610A	0·20	1·59	52·4	100·0	–	–	–	*d*
‚Gentran' Baxter Labs. U.S.A., Nr. 94090 . . .	0·22	1·11	> 60·0	100·0	–	–	–	*e*
‚Poliglukan' Experimental preparation, Poland .	0·19	1·49	57·0	100·0	–	–	–	*f*

pendent on molecular weight, at least within the molecular weight range of about 18,000 to about 60,000. This relationship has been made use of in determination of molecular weights of investigated dextrans described below.

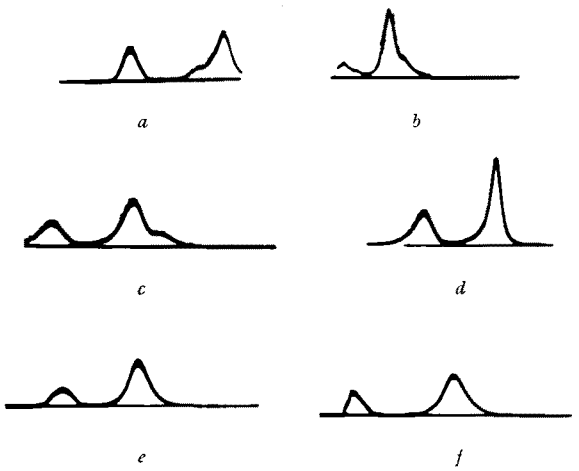

Fig. 2.

Samples of clinical dextrans of various manufacturers obtained from commercial sources, were analysed under the same conditions. Typical electrophoretic patterns are presented in Figure 2 *a–f*, and the results of corresponding calculations in the Table.

The above results clearly point to the usefulness of electrophoresis in the analysis of clinical dextrans.

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Department of Biochemistry, Institute of Haematology, Warsaw (Poland), July 26, 1957.

Résumé

On démontre que l'électrophorèse de dextrans cliniques peut être employée pour la détermination de leur poids moléculaire et de la polydispersité.

PRO LABORATORIO

Techniques for Making Capillary Microelectrodes[1]

The use of the LING-GERARD capillary microelectrodes[2] in electrobiological research has been rapidly growing. While microelectrodes can be made by hand and commercially made capillary pullers are available, a puller which can be assembled from parts found in a laboratory would have wide usefulness. A technique for rapidly filling capillaries is also described.

The principle of the puller is simple. A short length of soft glass tubing (about 1 mm o.d.) is held in the jaws of a tong-like holder. The tongs are so constructed that approximation of the handles causes the jaws to separate. An elastic band is stretched across the handles, but the jaws are prevented from separating by the capillary tubing. The tubing is heated with a microflame and the softened glass pulled to a fine capillary tip as the result of the restoring force of the elastic band. Simultaneously, with the sudden release of tension, the microflame is automatically pushed aside.

A detailed description of the component parts follows:

A tong-like holder (*A*) may be adapted from any tongs whose arms do not cross over at the pivot point, or may be fashioned from small stock. The ends of the tongs with the wider excursion (of about 4 inches) are fitted with alligator clamps (*B*) in the jaws of which are placed thin strips of rubber. One arm of the tongs is fixed by a clamp, while the other arm is free to move. With the capillary tubing (*C*) inserted, the alligator clamps are approximately 1–2 inches apart. A rubber band (*D*) is looped several times across the opposite ends of the tongs so as to be under stretch when the capillary tubing is in place.

The microburner (*E*) is made from a 13 gauge hypodermic needle flared at the point to simulate a wing tip. The wing tip is made by cutting off the bevel of the needle and pinching the ends with a pair of pliers. The microburner is clamped to a horizontally oriented rod

[1] This work was supported by a research grant from the National Institute of Neurological Diseases and Blindness, U. S. Public Health Service.

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