

Discussion and conclusion.—The present work has shown that while the decrease of serum inorganic P which occurs after casein hydrolysate administration is accompanied by an increase of serum labile phosphoric esters, this does not occur after glucose administration, although a sharp decrease of inorganic P is shown in this case also.

The behaviour observed after glucose administration eliminates the possibility that the increase of labile phosphoric esters of serum observed after casein hydrolysate administration, is to be attributed to the administration of compounds which by being oxidated by KREBS cycle, supply the necessary energy for labile phosphoric bonds synthesis.

The increase of labile phosphoric esters after casein hydrolysate administration may be explained if it is then for granted, according to LIPMANN, that the amino acids administrated are phosphorylated with formation of acylphosphates.

If this last hypothesis is accepted, we can understand how a part of the acylphosphate formed at tissues level and not utilized in proteosynthesis processes, may avoid the high acylphosphatase activity of tissues¹, and pass into the blood stream causing a temporary increase of labile phosphoric esters.

This would explain the lack of increase of labile phosphoric esters after glucose administration while inorganic P decrease, observed either after glucose administration or after amino acids administration, might be due, in both cases, to the utilisation of inorganic P for the resynthesis, at tissues level, of the A.T.P. largely involved in forming hexose-6-phosphate, in the first case, and acylphosphates in the second case.

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Riassunto

Alla introduzione endovenosa di un idrolisato di caseina, consegue un abbassamento del P inorganico del siero con aumento degli esteri fosforici. Gli autori pensano che tale reperto possa interpretarsi ammettendo che gli aminoacidi introdotti vengano fosforilati con formazione di acilfosfati prima di essere uniti in legami peptidici.

¹ F. LIPMANN, *Adv. Enzymol.* 6, 231 (1946). – E. SHAPIRO and E. WERTHEIMER, *Nature* 156, 690 (1945).

Studies on Arterio-Venous Anastomoses in the Lungs

It is now generally accepted that arterio-venous anastomoses exist in the lungs. As regards the conditions however in which they may act as haemodynamic shunts the literature is controversial¹. Further nothing is known as to the percentage amount of blood coming from the pulmonary artery that passes through these anastomoses and thus bypassing the lung capillary circulation. We therefore undertook the following studies to get quanti-

tative data regarding the functioning of these channels in various species of animals.

Methods. In principle the method consisted in injecting at a slow fixed rate a known quantity of radioactive spheres (P^{32})¹ with diameters of 25 to 30 μ into the jugular vein of narcotised animals and sacrificing the animals after 15 to 30 min to determine the radioactivity retained in the lungs. The difference in activity of the spheres injected and those retained in the lungs corresponds to the amount of the spheres that have passed through anastomotic channels. This difference may be considered to be a criterion of the percentage volume of blood from the pulmonary artery passing through these shunts. The studies were conducted on rats, guinea pigs, rabbits and cats.

Table I

Data sheet indicating the injected, residual and the total activity of ten specimens of spheres

Sample No.	Weight of spheres mg	Activity of the spheres cts./min		
		glass a (injected)	glass b (residual)	a + b (total)
1	5	7789	2602	10 391
2	5	7100	3361	10 461
3	5	7097	2936	10 033
4	5	7552	2850	10 402
5	5	7650	2855	10 505
6	5	7479	3114	10 593
7	5	7566	2657	10 223
8	5	7614	2684	10 298
9	5	7948	2650	10 598
10	5	7841	2257	10 098
Mean				10 360
Standard Deviation*				± 1,9%

$$* \text{ Standard Deviation} = \sqrt{\frac{1}{N-1} \cdot \sum (x - \bar{x})^2}$$

Rats, guinea pigs and rabbits were narcotised with 2 g/kg urethane subcutaneously and cats with 40 mg/kg sodium pentothal intraperitoneally. The suspensions of the spheres were prepared freshly each time just before the injection. 5 mg of spheres were put into a flask containing 2 ml of normal saline with 4 mg of Dioctyl-Sodium-Sulfosuccinate (aerosol) for the rats and double the above quantities for the other species. The aerosol was used to prevent adhesion of the spheres and to give a homogenous suspension. The suspensions were put into a syringe and injected for 2 min at a uniform speed. The spheres left over in the syringe and flask were carefully collected and their activity determined. $\frac{1}{4}$ to $\frac{1}{2}$ h after the injection the animals were bled to death, chest opened and lungs removed. The organs were washed free of blood and the wet weight recorded. They were then emulsified thoroughly and 1 g of the homogenate was wet ashed and taken for determining radioactivity.

Since in our method the percentage flow in the anastomoses is calculated by noting the difference in the activity of the spheres injected with those retained in the lungs, accurate knowledge of the exact amount injected is of primary importance. In order to assess the accuracy and the degree of variation in our technique the following experiments were carried out:

¹ H. EMMENEGGER, A. HÜRLIMANN, and K. BUCHER, *Helv. physiol. Acta* 9, 224 (1951).

¹ M. CLARA, *Die arterio-venösen Anastomosen* (J. A. Barth, Leipzig, 1939). – H. V. HAYEK, *Z. Anat.* 112, 221 (1943). – A. HÜRLIMANN, *Arch. int. Pharmacodyn.* 80, 99 (1949). – K. PRINZMETAL, E. M. ORNITZ, Jr., B. SIMKINS, and H. C. BERGMANN, *Amer. J. Physiol.* 152, 48 (1948). – H. RAHN, R. C. STROUD, and C. E. TOBIN, *Proc. Soc. exp. Biol. Med.* 80, 239 (1952).

A known weight of spheres were put into a small flask containing a few glass beads. The diluent was added, thoroughly mixed and taken up in a syringe. This was then injected into a counting chamber (glass *a*). The syringe as well as the flask were rinsed with chloroform and the spheres thus collected were put into a second cup (glass *b*). The radioactivity of the two specimens was determined separately. The sum of the two gave the total activity of the spheres. Conversely, the total activity less the amount retained in the syringe and the flask must be the amount injected. The results of one such series of experiments out of many are given in Table I. They indicate that by estimating the residue the actual amount injected may be obtained with a fluctuation of only $\pm 2\%$ and provides an index of the degree of accuracy obtainable by this method.

Table II

Activity in the lungs of rabbits after the injection of radioactive spheres (P^{32})

Rabbit No.	Activity injected	Activity found in the lungs	Difference which has passed through A-V anastomoses	
			Absolute	% of amount injected
1	14,000	12,265	1735	12,4
2	13,200	11,158	2042	15,5
3	14,300	11,575	2725	19,1
4	12,500	10,425	2075	16,6
5	12,800	11,212	1588	12,4
6	12,400	10,788	1612	13,0
7	17,000	15,130	1870	11,0
8	16,400	13,978	2422	14,8
9	15,600	12,760	2840	18,2
10	14,700	12,658	2042	13,9
Mean				14,7
Standard deviation				$\pm 2,7$

Results. The experiments were conducted on ten specimens of each species. The activity of the spheres injected and the amount retained in the lungs were determined in each case and the activity passing through A-V anastomoses calculated. The results obtained in the rabbit experiments are summarised in Table II.

The results for the other species were obtained in a similar manner. The final evaluation is recorded in Table III.

Table III

The extent of arterio-venous anastomoses in various species of animals

No. of experiments	Species	Percentage of spheres passing through A-V anastomoses
10	Rabbits	$14,7 \pm 2,7$
10	Rats	$23,7 \pm 3,8$
10	Guinea pigs	$25,3 \pm 5,1$
10	Cats	$34,5 \pm 4,6$

Discussion. Analyses of our experimental results indicate the existence of A-V anastomoses in all the species examined, as well as variations in the extent of anastomoses in the different species. It was also observed

that within a small range of variation the extent of anastomoses is comparatively constant for each species.

As the values indicated in Table 3 do not take into account anastomoses of a diameter below 25μ , the amount of blood by-passing the capillary system may be even greater.

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Zusammenfassung

Bei Katzen, Kaninchen, Meerschweinchen und Ratten wurde untersucht, in welchem Ausmass das Pulmonalarterienblut normalerweise durch arterio-venöse Kurzschlüsse fliesst.

Bei allen genannten Tierarten waren Kurzschlüsse nachweisbar. Sie führen bei Katzen etwa 35% des Minutenvolumens des rechten Herzens, bei Meerschweinchen und Ratten etwa 25% und bei Kaninchen 15%.

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Beitrag zur Kenntnis des menschlichen Linseneiweisses

Wenig zahlreich sind Untersuchungen der Eiweissbausteine von Einzellinsen beim Menschen. Erst die Einführung der mehrdimensionalen Papierchromatographie hat eine solche ermöglicht, indem MOORE und STEIN¹ dieselbe auch für die quantitative Bestimmung ausgebaut haben. Auf diese Weise konnten wir erstmalig die getrennten Anteile der Rinde und des Kernes einer normalen menschlichen Linse quantitativ und qualitativ auf ihren Aminosäuregehalt hin untersuchen. Das Linseneiweiss der menschlichen Linse, das 30–40% des Gesamtgewichtes ausmacht, lässt sich nach den Arbeiten von MOERNER, JESS², KRAUSE³, BLOCH und SALIT⁴, SCHAEFFER und MURRAY⁵, MERRIAM und KINSEY⁶ u.a.m. durch Aussalzen und elektrophoretisch in verschiedene Fraktionen aufteilen.

Eine Analyse der einzelnen Bausteine dieser Fraktionen mittels chemischer und mikrobiologischer Methode ist vereinzelt durchgeführt worden. Für unsere eigenen Untersuchungen haben wir eine normale frische menschliche Linse eines 43jährigen Mannes in Kern und Rinde aufgetrennt und die Hydrolysate der beiden Anteile auf ihren Aminosäuregehalt qualitativ und quantitativ analysiert. Für die zweidimensionale Chromatographie der Hydrolysate wurde erst aufsteigend eine Mischung Propanol/Wasser verwendet und anschliessend absteigend wassergesättigtes Phenol. Die erste Stufe der Anfärbung mit Moore- und Stein-Reagens erfolgt gesamthaft auf dem Papier, die zweite

¹ S. MOORE und H.W. STEIN, *Reagents*, aus HINSBERG-LANG, *Medizinische Chemie*, 2. Aufl. (München 1951), S. 427.

² A. JESS, *Arch. Ophth.* 105, 428 (1921).

³ A. C. KRAUSE, *Biochemistry of the Eye* (Baltimore Johns Hopkins Press, 1934).

⁴ R. BLOCH und P. W. SALIT, *Arch. Biochem.* 10, 277 (1945).

⁵ A. I. SCHAEFFER und I. D. MURRAY, *Amer. J. Ophthalm.* 44, 833 (1950).

⁶ F. C. MERRIAM und V. E. KINSEY, *Arch. Ophthalm.* 44, 651 (1950).