

trifugated, and all the supernatant liquid removed except a volume equal to that of the cells. With platinum loop the cells are placed at the center of a cover-glass rimmed with vaselin which is at once inverted on a slide. The lymph-endotheliocytes with the neutral red in the hof of the nucleus number about 4 per cent. By the intraperitoneal injection of 10 cc. or more of whole rabbit's blood on three or four successive days the number is increased to 6 or 7 per cent.

Hem-endotheliocytes have not been demonstrated in the peripheral blood of normal rabbits. On the intravenous injection of 4 or 5 doses of 5 cc. of India ink at about 5-day intervals, there appears in the peripheral blood, within 24 hours after an injection, a cell identical with the hyaline type observed in the peritoneal exudates. This hem-endotheliocyte is characterized by one or more carbon particles in a clear cytoplasm practically devoid of neutral red. A maximum of 4 per cent has been observed. The appearance and disappearance of these leukocytes is subject to much variation. Three cc. of blood is readily obtained by puncture of marginal ear vein (ear other than the one used for ink injections) of the animal placed head downward in a box. Films on slides may be fixed for 2 hours in a solution consisting of 5 parts of commercial formalin and 95 parts of Zenker's fluid without acetic acid. The fixed films are transferred to 10 per cent formalin for 5 minutes, stained with hematoxylin (Harris without acetic) for 2 minutes, washed for a few seconds, and blotted dry at once. The neutral red is preserved in the cells that react to it.

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The effect of surface active substances on the diffusion of water through membranes.

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According to the investigations of Jacques Loeb,¹ the diffusion of water through membranes is a linear function of the solute,

¹Loeb, Jacques, *J. Gen. Physiol.*, 1919, ii, 189.

as would be expected according to the law of van't Hoff, only if the concentration of the salt solution is greater than $M/16$. In lesser concentrations than this the so-called "initial rate" of diffusion is represented by a curve having a maximum corresponding to a concentration varying from $M/512$ to $M/256$. If the concentration becomes greater the rate of diffusion decreases until a concentration of $M/16$ is reached. The experiments of Loeb, and of Bethe and Toropoff² seem to indicate the identity of electro-osmosis and free osmosis in low concentrations, and in this way furnish the explanation of the influence of electrolytes on the rate of diffusion by the difference of effect of the ions on the quantity of charge on the Helmholtz double layer on the capillary walls. Grumbach³ has shown that surface active substances exert an influence on capillary electric forces and consequently on the diffusion potential.

Our own investigations are concerned with the influence of surface active substances on the capillary kinetic phenomena.

Collodion sacs of 50 cc. capacity were attached to upright manometer tubes of 2 mm. diameter. The sacs were made on the inside of a 50 cc. Erlenmeyer flask, coated twice with 4 per cent collodion, and dried for one-half hour after the first coating, and 4 to 5 hours after the second. The sacs were filled with electrolytes of $M/32$ concentration immersed in distilled water.

In the case of a given electrolyte the manometer readings obtained with different sacs were relatively constant. Having established the normal rate of diffusion, the sacs were treated by filling them with solutions of surface active substances. After five to fifteen minutes the sacs were thoroughly rinsed, and again filled with electrolyte solutions. It was noted that this treatment of the membrane resulted in a very different rate of diffusion, as measured by the change of height of water in the manometer.

In the case of sodium chloride, sodium sulphate and sodium citrate and potassium ferrocyanide, the diffusion of water through the membranes into the electrolyte solutions was increased. The surface active substances causing this phenomenon

² Bethe, A., and Toropoff, T. H., *Ztschr. f. Physiol. Chem.*, 1914, lxxxviii. 656; lxxxix, 597.

³ Grumbach, *Ann. d. Chim. et de Phys.*, (8) 24,433, (1911); *Journ. d. phys.*, 2,283,385, (1912). Quoted by Freundlich, *Kapillarchemie*, Leipzig, 1922, 358.

were caproic acid, methylamine, ethylamine, theobromin sodio-salicylate, sodium oleate and sodium glycocholate, the latter two substances producing the greatest alteration in the membrane.

In the case of certain electrolytes with polyvalent cations, the rate of positive osmosis was decreased. In the case of calcium chloride solution in sacs previously treated with sodium glycocholate, actual negative osmosis occurred, *i. e.*, water diffused from the electrolyte solutions into distilled water. This negative osmosis cannot be explained on the basis of valency alone, as aluminium salts fail to exhibit the phenomenon. The negative osmosis increases with the degree of dilution up to a maximum of M/64. At a concentration of M/8 the negative osmosis is completely compensated by the normal osmotic pressure. Repeated and long continued washing of the membranes after treatment with the surface active substance tended to decrease the negative osmotic effect. On the other hand, it was found that the phenomenon was obscured if an appreciable amount of surface active solution was allowed to remain in the sac.

Negative osmosis has been described by other authors in the case of certain acids, but not in the presence of neutral salts.¹ In Table I are given protocols of a typical experiment.

TABLE I.

Electrolyte solutions in sac.	Manometer Readings.					
	Before treatment of membrane.		After treatment with sodium glycocholate (M/40).		After treatment with sodium oleate (M/10).	
	Minutes	mm.	Minutes	mm.	Minutes	mm.
Na ₂ SO ₄ (M/32)	20	+37	—	—	10 20	+60 +82
Na ₂ SO ₄ (M/32)	20	+68	5 10	+62 +124		
Sod. Citrate (M/32)	20	+60	4	+90		
Pot. Ferrocyanide (M/32)	20	+78	5	+172		
Pot. Ferrocyanide (M/32)	20	+75			20	+132
CaCl ₂ (M/32)	20	+7			20	balance
CaCl ₂ (M/32)	20	+4	20	—19		
CaCl ₂ (M/64)	20	+6	20	—39		
CaCl ₂ (M/128)	20	+5	20	—16		