

Guiana, a normal speed of 7.6 centimeters a second is recorded,⁷ and on a later occasion as speed of 15.2 centimeters a second.⁸

4. There appears to be little, if any, real difference in speed towards and away from the nest. Large and small workers have practically the same speed for all temperatures during the summer months; after two months of low temperature, the large workers are conspicuously more active than the small workers (ninth column of table I).

5. This species of ant runs as numerously in the night as during daylight hours; maximum activity falls between noon and midnight, according to the scarcely sufficient observations summarized in table IV. Within the limits of 14° and 38° centigrade, temperature appears to have little effect on the number of ants running in the files.

¹ Wheeler, W. M., *Proc. Amer. Acad. Arts and Sci.*, Boston, **51**, 1915 (255-286), p. 259

² Mayr, G., *Verh. Zool. Bot. Gesell. Wien.*, **5**, 1855 (273-478), p. 319.

³ Wheeler, W. M., *Proc. Amer. Acad. Arts and Sci.*, Boston, **52**, 1917 (457-569).

⁴ Wheeler, W. M., *Ibid.*, p. 521; and *Bull. Amer. Mus. Nat. Hist.*, New York, **21**, 1905 (321-333).

⁵ Woodworth, C. W., *Univ. of Cal. Pub.*, Berkeley, Cal., Agr. Exp. Sta. Bull. **207**, 1910 (53-82).

⁶ Shapley, Harlow, *Bull. Ecol. Soc. Amer.*, Tuscon, Ariz., **3**, 1919, No. 4.

⁷ Beebe, William, *Jungle Peace*, New York, 1919, p. 229.

⁸ Beebe, William, *Atlantic Monthly*, Boston, October 1919 (454-464), p. 458.

THE INFLUENCE OF IONS ON THE OSMOTIC PRESSURE OF SOLUTIONS

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I. The osmotic pressure of solutions is usually measured indirectly, namely, by the determination of the freezing-point. Direct measurements have thus far been rare for the reason that it is difficult to prepare membranes permeable for water but not for the solute. The measurements thus far obtained on non-electrolytes by Morse,¹ and by the Earl of Berkeley² and their collaborators show that the actual values observed are not far from those expected according to van't Hoff's theory.

The difficulties in the preparation of semipermeable membranes are considerably diminished when we substitute colloidal solutions for solutions of crystalloids. By using solutions of proteins, collodion membranes satisfy the demand of semipermeability.

Gelatin is a very convenient protein for the purpose of such experiments. Like proteins in general, it is an amphoteric electrolyte being capable of forming salts with acids as well as with bases. The hydrogen ion concentration at which gelatin is neither in combination with acid nor with base is $10^{-4.7}N$ ($p_H = 4.7$ in Sørensen's logarithmic symbol). This

hydrogen ion concentration is the isoelectric point. When acid is added to a solution of isoelectric gelatin, part of the acid combines with the gelatin to form gelatin-acid salts, and the proportion of isoelectric gelatin transformed into gelatin-acid salt increases with the amount of acid added. There exists always a definite equilibrium between free acid, gelatin-acid salt, and isoelectric gelatin. Since the physical properties of isoelectric gelatin are different from those of the gelatin-acid salt it is necessary to define a gelatin solution not only by the concentration of gelatin present but also by its hydrogen ion concentration. The writer was able to show that gelatin forms monogelatin salts with many dibasic or tribasic acids, e.g., oxalic, tartaric, and phosphoric acids, while it forms digelatin salts with sulfuric acid.³

When we add a base, e.g., NaOH or Ca(OH)₂ to isoelectric gelatin, metal gelatinates are formed, and the relative amount of non-ionogenic gelatin transformed into metal gelatinate also depends upon the hydrogen ion concentration.⁴

II. We can measure the osmotic pressure of gelatin solutions by putting them into collodion bags (cast in the form of Erlenmeyer flasks) closed with a rubber stopper perforated by a glass tube which serves as a manometer. A 1% solution of gelatin to which a certain amount of acid or alkali had been added was put into the collodion bag which was dipped into a beaker containing water of the same hydrogen ion concentration as that inside the collodion bag. One per cent solutions of gelatin-acid salts had the highest osmotic pressure when the p_H was about 3.4 and solutions of metal gelatinates had their highest osmotic pressure at a p_H of about 8.4. At about 24° the permanent osmotic pressure was reached in less than 20 hours.

The influence of ions on the osmotic pressure of gelatin solutions was as follows:

(1) The osmotic pressure of 1% solutions of metal gelatinates of a p_H of about 8.4 is a little over twice (but less than three times) as great when the metal is monovalent than when it is bivalent.

(2) The osmotic pressure of 1% gelatin-acid solutions of p_H 3.4 is a little over twice (but less than three times) as great when the acid anion is monovalent than in the case of SO₄.

(3) The addition of neutral salts or alkalies to 1% metal gelatinate solutions of p_H 8.4 lowers the osmotic pressure of the solution and the depressing effect increases with the valency of the cation of the electrolyte added.

(4) The addition of neutral salts or acids to 1% gelatin-acid solutions of p_H 3.4 lowers the osmotic pressure of the solution and the depressing effect increases with the valency of the anion.⁵

This influence of electrolytes on the osmotic pressure of gelatin solutions has no connection with the fact that gelatin is a colloid.⁶ This

latter fact plays only a technical rôle in our experiments since it makes it easy to find a strictly semipermeable membrane. The writer has been able to show that the influence of ions on the osmotic pressure of gelatin solutions is connected with the existence of an electrical double layer at the boundary of watery phase and collodion membrane, whereby the water diffuses into the solution in the form of electrified particles.

III. When we separate a solution of a non-electrolyte like cane sugar from distilled water by a collodion membrane, water diffuses into the solution with an initial velocity which is (up to gram molecular concentrations) a linear function of the concentration of the solute. When we separate a solution of an electrolyte from distilled water by a collodion membrane, water diffuses also into the solute but with an initial velocity which for lower concentrations of the solution is no longer a linear but a more complicated function of the concentration and the nature of the ions. The facts observed can be expressed in the following rules:

(a) When pure water is separated from a solution of an electrolyte by a collodion membrane, water diffuses into the solution as if its particles were positively charged and as if they were attracted by the anions and repelled by the cations of the solution with a force increasing with the valency and another constitutional quantity of the ion

(b) When the collodion membrane has previously been dipped for a short time into a solution of a protein and if the protein solution is then removed Rule *a* holds also except that in the presence of hydrogen ions and of simple cations with a valency of 3 or above, in sufficient concentration, the particles of water diffuse into the solution as if they were negatively charged and as if they were attracted by the cation and repelled by the anion of the electrolyte with a force increasing with the valency and another property of the ion.

(c) The relative influence of the oppositely charged ions of an electrolyte is not the same for different concentrations of an electrolyte. At lower concentrations the influence of that ion prevails which has the opposite sign of charge as the watery phase, while in higher concentrations (of many, but possibly not of all electrolytes), the effect of the other ion prevails. The turning point lies for a number of electrolytes at a molecular concentration of about $M/256$. Finally a concentration is reached where this ionic influence on diffusion ceases and the gas pressure effect prevails.⁷

These three rules apply also to solutions of gelatin salts. In the case of gelatin-acid salts water is negatively charged and is attracted by the gelatin ion (which is positively charged) and repelled by the acid anion. This repulsion is greater when the anion is bivalent (SO_4) than when it is monovalent, e.g., Cl. Hence the initial rate of diffusion of water into the gelatin-acid solution is greater in the case of gelatin chloride or gelatin nitrate, etc., than in the case of gelatin sulfate.

In the case of metal gelatinate the particles of water diffusing through the membrane are positively charged and attracted by the gelatin anion and repelled by the cation with which the gelatin is combined. This repelling effect is greater in the case of bivalent cations (Mg, Ca, Ba, etc.), than in the case of monovalent cations (e.g., Li, Na, K, NH_4). Hence the initial rate of diffusion of water into solutions of metal gelatinates is greater in the case of sodium gelatinate than in the case of calcium gelatinate. When we add an electrolyte to the solution of a gelatin salt the increase in concentration of electrolyte brings about the depressing effect of that ion which has the same sign of charge as the electrified watery phase, as expressed in Rule *c*.

The permanent osmotic pressure of a solution separated from pure water by a semipermeable membrane is that pressure which has to be applied to the solution side in order to cause equal numbers of particles of water to pass simultaneously through the membrane in opposite directions. It follows from this that the relative influence of electrolytes must be the same on the permanent osmotic pressure as on the initial velocity of diffusion through the membrane from pure water to solution, and this was found to be the case with the solutions of gelatin salts.

IV. The terms "attractive and repulsive effect" of the ion on the charged particles of water served the purpose of simplifying the presentation of the facts observed. In reality the "attractive and repulsive effect" of ions as expressed in Rules *a*, *b*, and *c* are the expression of an additive effect of the oppositely charged ions of an electrolyte on the density of charge of the double layer formed at the boundary of collodion membrane and watery phase. This was proved by experiments on electrical endosmose. When the collodion membrane is bounded on both sides by identical solutions, an equal number of particles of water will diffuse in opposite directions through the collodion membrane. When, however, an external potential difference is produced on the two sides of the membrane, an electrical endosmose will be established to one of the two electrodes according to the sign of the electrification of the watery phase in contact with the collodion membrane. By this method I ascertained the influence of electrolytes on the sign of charge on the watery phase which was expressed in Rules *a*, *b*, and *c*.

Cases of abnormal of osmosis, e.g., the fact that liquid diffuses through parchment membranes from oxalic acid into pure water had been known for more than 50 years and it had been suggested by several authors, Girard, Bernstein, Bartell and Hocker, and Freundlich,⁸ that potential differences on both sides of the membrane caused these anomalies. To prove this view it was necessary to measure these potentials. Direct measurements of such potentials have been made by Girard, by Bartell and Hocker,⁸ by T. Hamburger,⁹ and by the present writer (as yet unpublished), but the results have thus far not been satisfactory. One can,

however, test the idea indirectly by experiments on electrical endosmose. This method of verification seemed to be confronted with a serious difficulty. The influence of ions on electrical endosmose has been investigated by Perrin¹⁰ who found that only one of the oppositely charged ions of an electrolyte, namely, the one which has the same sign of charge as the watery phase, has any influence (namely, a retarding one) on the velocity of endosmose, while the ion with the opposite charge has no influence. In my experiments on common osmose both ions of an electrolyte influenced the rate of diffusion of water simultaneously but in an opposite sense (as stated in Rules *a*, *b*, and *c*). I have investigated the influence of ions on the rate of electrical endosmose and found that Perrin's rule does not hold for collodion membranes, but that the Rules *a*, *b*, and *c* express not only the influence of ions on the transport of water through collodion membranes in common osmosis but also in the case of electrical endosmose.

According to the formula of Helmholtz as modified by Perrin, we know that in the case of electrical endosmose the following relation holds:

$$v = \frac{9.\epsilon.E.D}{4\pi.\eta.l}$$

where v is the quantity of liquid carried electro-osmotically, ϵ is the potential difference between the two strata of the double layer, E the external electromotive force, D the dielectric constant of the medium, η the coefficient of internal friction, and l the distance of the external electrodes.

In our experiments all quantities occurring in the formula except v and ϵ were kept approximately constant. If the theory of Helmholtz is correct, we must, therefore, attribute the influence of ions on the rate of transport of water in electrical endosmose to their influence on ϵ , i.e., the density of charge on the watery phase; and since, moreover, my experiments show that the influence of ions is the same for electrical and for free endosmose through collodion membranes we must conclude that the influence of ions on the initial rate of diffusion and on the osmotic pressure of a solution is due to the influence of ions on the density of charge in the double layer at the boundary of watery phase and membrane.

V. This permits us to define more accurately the influence of ions on osmotic pressure. At the boundary of a membrane and liquid an electrical double layer is formed. In the case of collodion membranes the latter usually assumes a negative charge while the watery phase assumes a positive charge. When the collodion membrane is bounded on one side by pure water on the other by a solution of an electrolyte the anions of the latter increase the negative charge on the membrane and the cations diminish it, both effects increasing with the valency and another property of the ions. At lower concentrations of the electrolyte the influence of the anion increases more rapidly with increasing concentration of the electrolyte than the depressing effect of the cation, while at higher concentra-

tions the reverse occurs. The turning point lies for a number of electrolytes at a molecular concentration of about $M/256$. Positively charged particles of water in the pores or interstices of the membrane will be driven to that side of the membrane which is more negatively charged. Since this is usually the solution side, water will be driven from the side of pure water into the solution. When, however, the membrane has a higher negative charge on the side of pure water, the flow of the positively charged liquid through the membrane will be from the side of the solution to that of pure water (negative osmosis). This occurs in the case of solutions of $(\text{CaOH})_2$ and $\text{Ba}(\text{OH})_2$ when the concentration exceeds $M/256$.

When the collodion membrane has been treated with a protein, it is also generally negatively charged except when the solution contains hydrogen ions or simple trivalent or tetravalent cations beyond a certain concentration (which for H is 10^{-4} N); in this case the membrane is positively and the watery phase is negatively charged. If we add in this case an electrolyte to the water the charge on the membrane is increased by the cations and diminished by the anion of the electrolyte, both influences increasing with the valency of the ion, and another property which is still to be defined. In lower concentrations the influence of the cation increases more rapidly than that of the anion, in higher concentrations of the electrolyte the reverse is true. Whenever the positive charge on the solution side of the membrane is greater than on the opposite side, the negatively charged particles of water will diffuse from the side of pure water to the side of solution. When, however, the positive charge on the solution side of the membrane is less than that of the side of pure water, the negatively charged layer of liquid will flow through the membrane from the side of the solution to that of pure water (negative osmosis). This will happen especially in the case of dibasic or tribasic acids when the concentration exceeds a critical limit. (Since the collodion membrane is permeable for electrolytes we are dealing in reality not with pure water separated by the membrane from solution, but with the condition of weak solution separated from a more concentrated solution.)

It is obvious that with increasing concentrations of the solution the opposite effects of the oppositely charged ions of an electrolyte on the density of charge of the double layer tend to become equal and then the gas pressure effect becomes the main or only driving force for the diffusion of water into solution. For lower concentrations of electrolytes the algebraic sum of both forces, the differences of potential on both sides of the membrane and the gas pressure effect, determine the rate of diffusion of water through a membrane into a solution.

The new experiments on which these conclusions are based will appear in the *Journal of General Physiology*.

¹ Morse, H. N., "The osmotic pressure of aqueous solutions". *Carnegie Inst. Washington, Publ.*, **198**, 1914.

- ² Earl of Berkeley, and Hartley, E. G. J., *London, Proc. R. Soc. (A)*, **92**, 1916 (477).
³ Loeb, J., *J. Gen. Physiol.*, **1**, 1918-19 (559).
⁴ Loeb, J., *Ibid.*, **1**, 1918-19 (483).
⁵ Loeb, J., *Ibid.*, **2**, 1919-20 (87, 273).
⁶ The influence of electrolytes on the osmotic pressure of gelatin is not due to differences in the degree of electrolytic dissociation of the gelatin salts, since, e.g. Na gelatin and Ca gelatin of the same concentration of gelatin and hydrogen ions have practically the same conductivity.⁴
⁷ Loeb, J., *J. Gen. Physiol.*, **2**, 1919-20 (173, 255); these PROCEEDINGS, **5**, 1919 (440).
⁸ Girard, P., *Paris, C. R. Acad. Sci.*, **146**, 1908 (927); **148**, 1909 (1047, 1186); **150**, 1910 (1446); **153**, 1911 (401); La pression osmotique et le mécanisme de l'osmose, *Publications de la Société de Chimie-physique*, Paris, 1912; Bernstein, J., *Electrobiologie*, 1912; Bartell, F. E., *J. Amer. Chem. Soc.*, **36**, 1914 (646); Bartell, F. E., and Hocker, C. D., *Ibid.*, **38**, 1916 (1029, 1036); Freundlich, H., *Kolloid-Zs.*, **18**, 1916 (11).
⁹ Hamburger, T., *Zs. physik. Chem.*, **92**, 1917 (385); (*Ann. Physik, Beiblätter*, **42**, 1918 (77)).
¹⁰ Perrin, J., *J. Chim. Physique*, **2**, 1904 (601); **3**, 1905 (50); *Notice sur les titres et travaux scientifiques de M. Jean Perrin*, Paris, 1918.

EVIDENCE ON THE NATURE OF NUCLEAR ACTIVITY

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Binuclearity hypotheses, founded in part on Richard Hertwig's chromidial hypothesis, have influenced interpretations of extra-nuclear bodies in the cytoplasm profoundly. The obvious dual capacity of the metazoan nucleus, exhibited in kinesis and interkinesis, i.e., in generative (propagatory), and somatic phases, has been made to lend itself to analogy with a true binucleate condition and to an assumption that the nucleus contains two kinds of chromatin. Of these one is supposed to be propagatory (idiochromatin), in evidence at the time of cell division, the other trophic (trophochromatin, somatochromatin), formed by the idiochromatin, but resident in the cytoplasm. This phase of the binuclearity idea is quite independent of that to which support is given by the demonstration of dimorphism in chromosomal groups during oogenesis and spermatogenesis.

The somatic phase of the nucleus covers the period during which it may be assumed that the nuclear enzymes have passed from the nucleus to the cytoplasm, and the cytoplasm has become the seat of synthetic activities. The nucleus at this time is in a "resting" condition; it seems comparatively empty, is acidophile, and basophilic granules may be found in the cytoplasm.

What is the nature of these basophilic bodies? Are they of direct or indirect nuclear origin?

The accounts of the extrusion of chromatin from the nucleus are numerous. Under the influence of the binuclearity hypothesis supposed particles

velopment of seismologic and volcanologic research directly before both national and international bodies qualified to furnish advice and promote a broad coöperation of great value.

Yours respectfully,

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ERRATA

P. 215. For $v = \frac{9. \epsilon. E. D}{4\pi. \eta. l}$ read $v = \frac{q. \epsilon. E. D}{4\pi. \eta. l}$.

P. 268. In theorem II for "but $B_{\nu\lambda} \equiv 0$ " read "but $B_{\nu\lambda}$ not $\equiv 0$ "; and in theorem IV for "or $0 \frac{+}{+}$ " read "or not $\equiv 0$ ".

P. 416. For the denominator of (1) read $2^a p^b$ where $a = \frac{1}{2} p^n - 1 (p-1)^2$, $b = \frac{1}{2} n p^n - 2(p-1)^2 - 1$, and on p. 417 read p^b for the denominator of the second fraction and 2^a for the left side of the next formula.