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Maximum values of osmotic concentration in plant tissue fluids.

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The observations of a number of botanists have shown that extremely high concentrations may characterize plant tissue fluids, especially when the plants¹ occur in a highly saline substratum. To Fitting² belongs the credit of first demonstrating that extremely high osmotic concentrations are found in some desert plants,³ although Drabble and Lake⁴ and Drabble and Drabble⁵ had preceded him in showing the fundamental relationship between environmental conditions and the osmotic concentration of plant tissue fluids.

As early as 1902 Cavara reported cryoscopic determinations on saps of high concentration⁶ and in 1905 gave results in full⁷ for a large series of determinations made at Cagliari. His maximum values were found in the sap of halophytes growing in localities where the concentration of the soil solution progressed with the advance of the season. He reports freezing point depressions of 7.25° in *Obione portulacoides*, 7.48° in *Salicornia fruticosa*, and 7.25° to 8.50° in *Halocnemum strobilaceum*. His determinations

¹ Our present observations apply to the tissue fluids of flowering plants only. No attempt is made here to discuss the concentrations found in such lower organisms as those studied by G. J. Peirce (Pub. Carn. Inst. Wash., 1914, No. 193, p. 47-69) or G. Senn (*Verh. Schw. Naturf. Ges.*, 1911, xciv).

² H. Fitting, *Zeitschr. f. Bot.*, 1911, iii, 209-275.

³ Fitting found a number of species of plants in the North-African deserts, the leaf cells of which were not plasmolyzed by 3 gram molecular KNO₃ solution. Theoretically potassium nitrate of this concentration should be the equivalent of about 100 atmospheres. The technical difficulties of applying the plasmolytic method are such as to lead one to question its value as a means of investigating in a quantitative manner the unusually high concentrations found in desert plants.

⁴ E. Drabble and H. Lake, *New Phytologist*, 1905, viii, 189.

⁵ E. Drabble and H. Drabble, *Biochem. Jour.*, 1907, ii, 117.

⁶ F. Cavara, *Rendic. Congr. Bot. Palermo*, 1902, 66.

⁷ F. Cavara, *Contrib. Biol. Veg.*, 1905, iv, 41-84.

were, however, made on sap extracted without the antecedent treatment necessary to render the tissues permeable as has been shown to be necessary by Dixon and Atkins⁸ and others.⁹ His constants are, therefore, as pointed out by Atkins,¹⁰ probably sub-maximum because of incomplete extraction.

Work on the spring flora of the Arizona deserts¹¹ was probably carried out in a manner to obviate the objections to the preceding studies. In this series the maximum concentrations were found in *Atriplex canescens*, a shrub of the salt spots, in which $\Delta = 5.65$, $P = 67.5$, and in *Mortonia scabrella*, a small shrub of the mesa-like slopes, for which one determination gave $\Delta = 4.78$, $P = 57.2$.

Concentrations of about fifty atmospheres have been demonstrated in the leaf tissue fluids of more or less sclerophyllous trees *Capparis ferruginea* and *Guaiacum officinale* and in those of the succulent-leaved halophytic half shrub *Batis maritima* of the saline coastal flats of Jamaica.¹² Cryoscopic studies on mangrove vegetation¹³ have indicated maximum concentrations of about fifty atmospheres in *Avicennia nitida*, although two questionable determinations indicated seventy atmospheres. Using plasmolytic methods, von Faber¹⁴ reports concentrations ranging from 24 to 72 atmospheres in East Indian species of the mangrove association.

During the summer of 1920, while engaged in field operations in collaboration with the Department of Agriculture in the Great Salt Lake region, we had the opportunity of making several hundred determinations of the osmotic concentration of plant tissue fluids by the cryoscopic method. These measurements were made on sap extracted after freezing of the tissues¹⁵ and with

⁸ H. H. Dixon and W. R. G. Atkins, *Proc. Roy. Dublin Soc.*, 1913, N. S., xiii, 422-433.

⁹ R. A. Gortner, J. V. Lawrence, and J. Arthur Harris, *Biochem. Bull.*, 1916, v, 139-142, pl. 1.

¹⁰ W. R. G. Atkins, "Some Recent Researches in Plant Physiology," London, 1916, 94.

¹¹ J. Arthur Harris, J. V. Lawrence, and R. A. Gortner, *Phys. Res.*, 1916, ii, 1-49.

¹² J. Arthur Harris and J. V. Lawrence, *Bot. Gaz.*, 1917, lxiv, 285-305.

¹³ J. Arthur Harris and J. V. Lawrence, *Biol. Bull.*, 1917, xxxii, 202-211.

¹⁴ F. C. von Faber, *Ber. Deutsch. Bot. Ges.*, 1913, xxxi, 277-281.

¹⁵ R. A. Gortner and J. Arthur Harris, *Pl. World*, 1914, xvii, 49-53.

such care as to render the results reasonably free from criticism. Such a series, based on species which have for ages been subject to the influence of the highly saline substratum afforded by the bed of the ancient Lake Bonneville, should furnish some indication of the maximum concentration¹⁶ to be found in the leaf tissue fluids of flowering plants.

While high concentrations were demonstrated in a number of species, the highest was found in the typical salt desert half-shrub *Atriplex confertifolia*. It alone will be considered.

Two collections made on the rocky cliffs of Stansbury Island, Great Salt Lake, on July 14 gave freezing point depressions of 6.96° and 7.97°. If we may use the formula of Lewis,¹⁷ upon which published tables of osmotic concentration have been based,¹⁸ these depressions indicate osmotic pressures of 82.9 and 94.7 atmospheres respectively.

The highest concentrations were found in plants growing on the low ridges in the salt-flats¹⁹ along the southern shore of Great Salt Lake. A determination on material collected July 16 gave $\Delta = 6.22$, $P = 74.2$.

On July 18 a determination on plants in about the same type of locality gave $\Delta = 10.00$, $P = 118.5$. Finally, on July 27 a determination made in this locality on the leaves of this species indicated a freezing point lowering of 13.6°. The equation used would indicate a concentration of 153.1 atmospheres.²⁰

¹⁶ A difficulty in work on the leaves of desert plants lies in the fact that the maximum concentrations must be expected during the periods of more extreme drought. During such periods the saps may become concentrated by desiccation merely. We know very little concerning the functional activities of such leaves or whether they are retained after the beginning of a period of more adequate moisture. There is, therefore, the possibility that leaves which are too desiccated to be longer functional may be utilized for determinations and indicate concentrations which are really larger than those in which the metabolic processes of the cells may be normally carried on. We believe that except as specifically indicated, the concentrations here recorded were determined on leaves in fairly normal condition.

¹⁷ G. N. Lewis, *Journ. Amer. Chem. Soc.*, 1908, xxx, 668-683.

¹⁸ J. Arthur Harris and R. A. Gortner, *Amer. Jour. Bot.*, 1914, i, 75-78; Harris, *Amer. Jour. Bot.*, 1915, ii, 418-419.

¹⁹ T. H. Kearney, L. J. Briggs, H. L. Shantz, J. W. McLane, and R. L. Piemisel, *Jour. Agr. Res.*, 1914, i, 365-417, pl. 52-58.

²⁰ A sample from *Atriplex nuttallii* showed a freezing point lowering of about 14.4°, indicating a concentration of 169.3 atmospheres. The leaves appeared more dried than those of *Atriplex confertifolia*, and we are inclined to await further measurements before accepting this constant.

These determinations show that concentrations measured by a depression of 13.0° , presumably the equivalent of 153 atmospheres, may be found in the tissue fluids of apparently normal leaves.

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The carbohydrate-fat ratio in relation to the production of ketone bodies in diabetes mellitus.

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Since relatively large amounts of fats are used in the construction of maintenance diets in the treatment of diabetes mellitus it is important to know the limits within which fat may be employed with safety. The normal composition of fat demands that carbohydrates shall be simultaneously oxidized. Zeller¹ has shown in two normal men and two normal dogs that of the total calories, the protein intake being kept low, 10 per cent. must be yielded by carbohydrate if 90 per cent. arises from fat in order to prevent the production of the ketone bodies. In commenting on these experiments Lusk² calls attention to the fact that it is possible that for the proper oxidation of fat, the end product of which is B-oxybutyric acid, the burning of one triose molecule may be necessary for the normal oxidation of one molecule of B-oxybutyric acid. The attempt has been made in this work to establish the proportion of available carbohydrate to fat when ketone bodies appear in the urine.

Diabetic cases are treated as follows: Freed from sugar and acetone body excretion, sugar tolerance ascertained and then the following experiment. The individual is put on a diet having a protein intake that will enable nitrogen equilibrium to be maintained with the fat and carbohydrate given. During different periods the protein intake is kept constant and the amount of carbohydrate and fat are varied isodynamically, the proportion

¹ Zeller, *Archiv. fur Physiologie*, 1914, p. 213.

² Lusk, "Science of Nutrition," third ed., pp. 270-271.