

The present investigation has concerned itself mainly with the water soluble extractive form of sulphur, which besides containing a compound which is either taurin or an immediate precursor of taurin, contains another group of compounds which appear to bear a close resemblance to the group of neutral sulphur compounds found in the urine. In view of the fact that Folin considers the neutral sulphur of the urine as a measure of tissue metabolism, this observation becomes of special significance. The possibility of comparing the metabolic activity of different tissues with one another, and of the same tissue under different conditions, is at once apparent.

No very close resemblance can be demonstrated until we know the chemical structure of these compounds.

The resemblances so far found are as follows: The neutral sulphur compounds of the tissues and of the urine are both soluble in water, soluble in dilute alcohol, not precipitated by phosphotungstic or tannic acids, precipitated by mercuric acetate. They do not precipitate with barium chloride direct or after boiling with hydrochloric acid. They contain lead blackening sulphur.

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The study of autolysis by physico-chemical methods.

By **ROBERT L. BENSON** and **H. GIDEON WELLS**.

Further studies of autolytic changes in animal tissues by means of the depression of the freezing point and rise in conductivity show the great value of these methods of estimating the rate and progress of autolysis. The results obtained in this way give a much more accurate and valuable indication of autolytic changes in any given tissue than the commonly used determination of the percentage of nitrogen in coagulable form. Autolysis comprises the disintegration of the cell components and involves a great many substances, some of which are coagulable proteins and many of which are not. If we determine the proportion of nitrogen that is made non-coagulable by heat, we get a figure which is the same whether the coagulable nitrogen that has been made incoagulable is in the form of proteoses and peptones, or has been carried to the ultimate amino-acids or even further. The several steps that take place in the autolysis of nucleins also have no effect on this figure after the

first splitting out and rendering soluble of the nucleic acid complex. Only the autolytic changes which affect coagulable or insoluble nitrogenous cellular constituents are shown, and the changes in such substances as collagen or the other non-coagulable nitrogenous tissue elements are not brought out. In other words, the ratio of coagulable and non-coagulable nitrogen in autolyzing tissues shows only one of the many changes that are being accomplished by the autolytic processes.

On the other hand the freezing point determination gives an absolute, delicate and reliable measure of the disintegration of the tissue, since practically every step of this disintegration results in an increase in the number of molecules in the solution. A freezing point curve is, therefore, a correct picture of the total disintegrative change that is taking place in the mixture, which a non-coagulable nitrogen curve cannot well be. If we supplement the freezing point curve with a conductivity curve we secure, in addition, information as to qualitative changes, for the conductivity curve indicates only the increase in the number of free ions, which we know are largely supplied by certain of the products of autolysis, while the difference between the two curves gives us a measure of the newly formed non-electrolytes. The information obtained by these two methods is, therefore, much more instructive as to the actual amount and rate of autolytic change than are the results of coagulable nitrogen estimations, and in addition the methods involved are much simpler and easier. A score of freezing point determinations and a hundred conductivity measurements can be obtained with no more expenditure of time and labor than one nitrogen determination, and much smaller quantities of material suffice — a point of great importance in many investigations.

As a general rule the curves of conductivity increase and freezing point depressions for the same tissue parallel each other fairly closely; at first the change in conductivity proceeds slower than the change in freezing point, but later the conductivity continues to rise when the depression of freezing point has come nearly to a standstill. This last phenomenon probably depends upon the liberation of ammonia from the amino-acids and purines by the amidases, and the formation of organic acids. Blood serum, lymph and cerebro-spinal fluid show no evidence of autolysis by

physical methods, but the conductivity of blood slowly falls as the hemoglobin is liberated from the corpuscles. The inhibiting effect of blood serum upon autolysis seems to be less readily destroyed by heat than is usually estimated.

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The influence of adrenalin in phlorhizin diabetes.

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These experiments were performed with the object of ascertaining whether or not the contention of Blum as well as of Eppinger, Falta and Rudinger, that adrenalin stimulates the conversion of fat into dextrose, is well founded. On careful analysis of their data, one may find every reason to believe that the animals used for their adrenalin experiments were not glycogen free, and that the extra sugar eliminated after the administration of adrenalin did not come from the ingested fat, but from glycogen or from the sugar of the blood.

If a phlorhizinized animal be exposed to cold and rendered glycogen free, any intraperitoneal injection of adrenalin ought to be followed by an extra elimination of sugar and a rise in the D:N ratio, provided the theory of the conversion of fat into carbohydrate is true. That this is not the case will be seen from the accompanying protocols.

	DOG No. 5.
March 6, 1909.	Dog fasting.
March 7, 1909.	Dog fasting.
March 8, 1909.	2 gm. of phlorhizin injected at 8 A. M., 3:30 P. M. and 10 P. M.
March 9, 1909.	2 gm. of phlorhizin injected at 8 A. M., 3:30 P. M. and 10 P. M. At 5:15 P. M. the dog was given a bath at a temperature of 8° C. for 30 minutes and while wet was placed in a cold room for 5½ hours.
March 10, 1909, 9:30 A. M.	2 gm. phlorhizin injected subcutaneously.
March 10, 1909, 10:30 A. M.	Catheterized and bladder washed.
	Weight of dog 9.12 kg.