

3. If our impression that the nucleus of the growing cell is actually stained prove correct, the use of stains in the plasma in which tissue is grown should certainly facilitate the study of nuclear growth.

4. Certain observations made last year in this laboratory (too few to serve as more than a suggestion) seem to indicate that another dye (methylene blue) acted as a stimulant to the growth of connective tissue. This lead also should be followed out and the effect of all possible stains studied in the hope of discovering dyes which will have a sharp selective action on growing tissue.

5. The growth of animal cells in a strength of dye much more than sufficient to kill many pathogenic organisms lends encouragement to the efforts now being made in this laboratory to apply the observations on the bactericidal effect of gentian violet and allied stains to the treatment of disease.

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On the hexosamine of chondroitin sulphuric acid.

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In a previous communication¹ the writers reached the conclusion that the nitrogenous component of chondroitin sulphuric acid was glucosamine. The conclusion was based on the analytical data of the hydrochloride of the amino sugar, and on the magnitude of its optical rotation.

However, recently it was discovered that the optical activity of the amino sugar differed considerably from that of glucosamine, if measured under very definite conditions. The conditions required are the following: low temperature of the solution, comparatively high concentration of the sugar solution, and measuring the initial rotation immediately after the solution of the sugar is accomplished. Under such conditions it was found that the specific rotation of the amino sugar of the chondroitin sulphuric acid was about 25 per cent. higher than that of glucosamine. Both

¹ *Jour. Biol. Chem.*, XV, p. 155, 1913.

substances displayed mutirotaion and reached equilibrium simultaneously. Also in its melting point and in its solubility the amino sugar differed from glucosamine. Finally the osazones of the two substances differed in their melting points and in their solubility in alcohol and in water.

On the basis of these data it seems justified to conclude that the hexosamine is not identical with glucosamine, but is isomeric to it.

Work on the configuration of the sugar is in progress.

| Substance. | Crystal-form. | Rotation. | | Solubility. | Melting Point. |
|--------------------------------|-------------------------|--|--------|--|--------------------------------|
| | | Initial. | Final. | | |
| | | [α] _D ⁰ = | | | |
| Glucosamine hydrochloride..... | Short thick prisms. | +101.6 | +73.65 | Difficultly sol. in 80 per cent. EtOH; insol. in abs. EtOH. | Decomposes slowly above 200°. |
| Osazone..... | Needles. | | | Difficultly sol. in abs. EtOH. | Melts with decomposition 206°. |
| Hexosamine hydrochloride..... | Long prismatic needles. | +129.5 | +93.82 | Easily sol. in 80 per cent. EtOH; difficultly sol. in abs. EtOH. | Melts with decomposition 180° |
| Osazone..... | Long needles. | | | Easily sol. in EtOH; sol. in hot H ₂ O. | 175-180° |