

what below the hatching length, and practically all of the newly hatched specimens fell down to about three fourths of their original length. Speaking in terms of reduction in size, it is astonishing to note that some of the largest larvæ have been reduced to about 1/600 of their maximum larval mass.

Another, and even more interesting phenomenon, is the fact that when the starved specimens almost reach the smallest size possible and are then given plenty of food, they will again begin growing in size. A number of the larvæ which were half grown when placed under starvation for the first time, have through alternating periods of "feasting and fasting" attained that size three times and are now on the way to their fourth "childhood"; and even some of the large specimens have started dwindling down to their third "childhood" after having twice attained the practically maximum larval size.

Occasionally these larvæ are found in large numbers in insect, seed and drug collections, and naturally destroyed as soon as discovered. The writer would appreciate any amount of this living material that the reader may happen to find if he has no use for it himself. The larvæ, pupæ or living adults of other dermestids are equally desirable for the purpose of comparative studies. In response to a recent circular letter many men have already sent me some valuable material. The names of the donators will appear in the forthcoming detailed publication of this extensive and of necessity prolonged investigation.

The problem has now attained enormous proportions and involves the use of thousands of specimens. Many normal larvæ of different sizes, as well as many specimens in the different periods of starvation have been sectioned during the past few years, and comparative cytological studies of the various structures of the organisms are being made. Physiological studies with special reference to metabolic water and excretion have also been started.

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SPECIAL ARTICLES

THE RÔLE OF THE NUCLEUS IN OXIDATION¹

IN 1897 Spitzer² reported that nucleoproteins extracted from certain animal tissues have the same oxidizing power as the tissues themselves. The idea that the nucleus is a center of oxidation was advocated by Loeb,³ who pointed out that this would explain why cells deprived of nuclei live but a short time and are unable to regenerate missing parts. R. Lillie⁴ sought to obtain direct experimental evidence by applying reagents which become colored on oxidation. He found the greatest amount of color in the neighborhood of the nucleus, indicating that it is a center of oxidation. Subsequent workers,⁵ using stains which change color on oxidation, failed to agree as to the results.

Mathews⁶ has stated that the nucleus is directly concerned in oxidation.

Warburg⁷ found that NaOH increased oxidation in the sea urchin egg, but did not penetrate sufficiently to cause a change of color in the interior of eggs stained with neutral red. This is regarded by some as indicating that oxidation is largely confined to the surface of the cell.⁸ R. Lillie⁹ has recently found that the formation of indophenol in leuco-

¹ Preliminary communication.

² *Pflüger's Archiv*, 67: 615, 1897.

³ *Archiv für Entwicklungsmechanik der Organismen*, 8: 689, 1899.

⁴ *Am. Jour. Physiol.*, 7: 412, 1902.

⁵ Cf. Wherry, E. T., *SCIENCE*, N. S., 37: 908, 1913; Schultze, W. H., *Verh. deutsch path. Ges.*, 16: 161, 1913; Reed, G. B., *Jour. Biol. Chemistry*, 22: 99, 1915. Unna, P. G. und Godoletz, L., *Oppenheimer's Handb. d. Biochem. Ergänzungsband*, 8: 327, 1913.

⁶ Mathews, A. P., "Physiological Chemistry," 1915, p. 180.

⁷ Warburg, O., *Zeit. f. physiol. Chemie*, 66: 305, 1910; *Biochem. Zeit.*, 29: 414, 1910.

⁸ This conclusion does not seem to be necessary. Cf. Loeb and Wasteneys, *Jour. of Biochemistry*, 14: 459, 1913; also Osterhout; *Ibid.*, 19: 335, 1914. Owing to the buffer action of the protoplasm and to the presence of pigment the penetration of a small amount of alkali is not easily detected.

⁹ *Jour. of Biol. Chemistry*, 15: 237, 1913.

cytes indicates that there is rapid oxidation at the surface of the cell as well as at the surface of the nucleus.

The objection might be made to the use of indophenol reaction that the result may depend somewhat on the manner in which the reagent penetrates. If the oxidizing substances of the cell are largely concentrated in the nucleus, those which are diffused throughout the cytoplasm will first meet the reagent at the cell surface and produce at that point a deposit of granules of indophenol. In the same manner the oxidizing substances which are retained within the nucleus will first meet the reagent at the surface of the nucleus and produce a deposit in that region. It would therefore appear that the reaction might be depended on if it showed the nucleus to have the greatest oxidative activity, since its error would lie in the opposite direction. But any conclusions drawn from it regarding oxidation at the surface of the cytoplasm would be of doubtful value.

It would seem that more reliable evidence can be obtained by investigating cases where it is not necessary that the reagent should penetrate from without owing to the fact that the cell itself produces the reagent.

The writer has studied a case of this kind in the Indian Pipe (*Monotropa uniflora*), which is extremely well suited to such investigations, because the cells contain a colorless chromogen which oxidizes and darkens very rapidly upon injury. An additional advantage is that the leaves are so thin and transparent that they may be placed under a microscope and the details of the cell structure studied with care before the cells are injured or treated with reagents.

In a typical leaf cell the cytoplasm is transparent and nearly colorless, with a few granules, while the nucleus is only slightly less transparent, is finely granular and has a nucleolus. When a leaf is mounted in a drop of water under a cover glass the cells remain for hours unchanged in appearance.

If an intact portion of the leaf is cut or crushed the cells in the neighborhood soon change. In the course of five or ten minutes

the nuclei of the cells nearest the injury assume a more coarsely granular (or vacuolated) appearance and soon begin to darken. The darkening does not begin at the surface, but appears to take place almost simultaneously throughout the whole mass of the nucleus. Not until the nucleus has become very dark (so as to stand out very conspicuously when the preparation is viewed under the low power of the microscope) does the cytoplasm begin to darken perceptibly. It may be several hours after the nucleus has darkened perceptibly before a change of color can be perceived in the cytoplasm. The darkening of the cytoplasm does not seem to be more rapid at the surface than elsewhere.

That the darkening is due to oxidation is shown by the fact that it is retarded by the partial exclusion of air¹⁰ and is inhibited by the usual means employed to prevent the action of oxidases. When young leaves (free from discolorations) are torn¹¹ and placed in water the torn edges become dark. This does not occur in 0.1 *M* HCl, 0.1 *M* KCN,¹² 0.1 *M* NaOH, or in boiling water. If the colorless chromogen is extracted by 0.1 *M* NaOH and kept in a tightly stoppered bottle so as to exclude oxygen it remains pale yellow for months, but if oxygen be admitted it soon turns deep red.

That the darkening of the nucleus is due to oxidation taking place in the nucleus itself and not to the taking up by the nucleus of a stain produced in the cytoplasm or vacuoles is shown by the following experiment: Plants were ground in a mortar and allowed to stand until they became black. The juice was squeezed out and centrifuged, giving an inky fluid. In this were placed pieces of leaves

¹⁰ That the oxidation is not completely inhibited by exclusion of air is doubtless due to the fact that a considerable supply of combined oxygen is present in the cell which can be used for oxidation of the chromogen.

¹¹ Cutting with a knife was avoided on account of the action of the metal.

¹² In 0.1 *M* NaOH and 0.1 *M* KCN the whole leaf becomes pale yellow and then colorless. The yellow color is doubtless due to the fact that the KCN solution is alkaline.

which had been treated with 0.1 KCN and then with water. The solution was allowed to stand until it became concentrated by evaporation: it then appeared black. It was found that where the nuclei had been squeezed out of the cut cells by the knife they had taken up some stain but not more than the cytoplasm. In cells which were merely cut open there was little or no staining.

We must therefore conclude that oxidation occurs more rapidly in the nucleus than elsewhere in the cell. The only way to escape this conclusion would be by assuming that at the moment of injury there is a sudden migration into the nucleus of some or all of the substances necessary for the oxidation. This is not only very improbable from a theoretical standpoint, but observation shows that it can not be the case, for in this migration the substances would mingle and produce the pigment either outside the nucleus, or at its surface, before any pigment appeared in the interior of the nucleus. Observation of the nucleus shows that the pigment appears as soon within the nucleus as at its surface.

We may therefore conclude that the substances necessary for oxidation do not suddenly migrate into the nucleus at the moment of injury but that they must exist there before the cell is injured.

We may ask why the nucleus does not become darkened in the normal condition of the cell. The investigation of several workers have made it probable that the pigments produced by oxidation under normal conditions are at once reduced, giving up their oxygen to other substances in the cell. When injury occurs the reduction is checked more than the oxidation, with the result that the pigment accumulates.

It is also probable that in many cases the injury brings the cells into contact with more oxygen than under normal conditions.

In order to compare these results with those produced by the indophenol reagent, leaves were placed in a mixture of equal parts of alpha naphthol (saturated aqueous solution) and para phenylene diamine (1 per cent. aqueous solution). If the reagents are freshly

made up there is little action, but if they have stood long enough to take up oxygen or if H_2O_2 is added a purple color develops in the cells, which eventually becomes deeper in the nucleus. The result depends greatly on the condition of the reagent and the rate at which it penetrates the tissue.

The general conclusion is that while the indophenol reaction indicates that the nucleus is the center of oxidation it does not give as definite information on this point as does the formation of natural pigments within the cell resulting from the oxidation of substances normally present.

SUMMARY

Injury produces in the leaf-cells of the Indian Pipe (*Monotropa uniflora*) a darkening which is due to oxidation. The oxidation is much more rapid in the nucleus than in the cytoplasm and the facts indicate that this is also the case with the oxidation of the uninjured cell.

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SOCIETIES AND ACADEMIES

AMERICAN MATHEMATICAL SOCIETY

At the invitation of Adelbert College and the Case School of Applied Science, Cleveland, Ohio, the twenty-fourth summer meeting of the American Mathematical Society was held at these institutions on Tuesday, Wednesday and Thursday, September 4-6, 1917. This was the society's second visit to Cleveland, the annual meeting having been held there in the winter of 1912-1913. On the present occasion the interest was reinforced by the meeting of the Mathematical Association of America, immediately following on September 6-7. The arrangements, which were in charge of a committee representing both organizations, included a joint session on Thursday morning, at which Professor L. P. Eisenhart presented an address on "Darboux's contribution to geometry," and a joint dinner on Wednesday evening, attended by seventy-six members and friends, to whom President Thwing, of Western Reserve University, spoke a word of welcome, which was followed by a number of informal responses to the calls of the toastmaster, Professor E. V. Huntington. The program on Wednesday afternoon included an inspection of the harmonic analysis apparatus of