## Letters to the Editor

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### Action of the Alkaloids and Carbon Monoxide on the Enzymatic Activity of Plants

IN a series of investigations which I began during 1929 [1] I was able to demonstrate, with the help of my collaborators, that the debatable question of the function of alkaloids in plants could be profitably examined by investigating the various enzymatic activities, beginning with those of germinating seeds.

The first case that I examined was that of barley in which exists, as is well known, an increasing activity of amylase with increase of germination. Beside, I noticed a net augmentation of activity due to the addition of dilute solutions of strychnine nitrate. In turn [2] the activities of lipase, amylase, protease, and oxidase were measured in castor oil seeds germinating in pure water and in strychnine nitrate solutions of different concentrations. In every case the addition of strychnine at the concentration optimum of 1 per thousand causes an increase of activity. It should be noted that this increase is not due to action of the alkaloid on the enzymes already formed, because the effect is not observed with the enzymatic extracts of seeds.

The amount of increase of the activity is different, the most sensible effects being in lipase and oxidase. The effect of alkaloids being consequently selective for different enzymatic activities explains, in my opinion, why they do not always help the growth of plants; in effect, they can determine a want of equilibrium in the various biochemical activities, which for full growth of the plant we suppose must be properly proportioned. Investigations showed that the *Ricinus* seedlings, that were followed for a month during their germination, showed constant improvement, by the addition of solutions of strychnine nitrate. The alkaloid contained in the bark of the *Ricinus* seeds, ricinine, has shown itself necessary for the germination, but strychnine can be substituted for it. Caffeine has also been found to be advantageous up to an advanced age; but this is an isolated fact, because this alkaloid is toxic for barley, flax, and other plants. From the observed facts, the opinion that alkaloids must be considered as hormones of the plants is strengthened.

My researches show that carbon monoxide itself alters in a sensible way the enzymatic activities of plants. This gas, also in small quantities, stops the assimilatory activity of green plants in light, but not the respiration (oxidase): so it was supposed that it also had selective action on the enzymatic secretions.[3] It was effectively shown that carbon monoxide depresses amylase activity of barley and of *Ricinus* powerfully; while in the same *Ricinus* it increases slightly the lipase activity. In consequence, the behaviour of the gas during the germination of seeds was to be expected to be irregular; thus it was found that while wheat is able to germinate in an atmosphere rich in carbon monoxide, the field cabbage and hemp only germinate with great difficulty, while flax and tomato do not germinate at all. Leguminous plants, such as lupin and pea, showed special behaviour: they are able to germinate in an atmosphere rich in carbon monoxide, and also if the operation is accomplished under a cover with seeds made sterile: a considerable absorption of nitrogen which causes much depression is observed.[4]

The determination of the contained combined nitrogen denoted an increase from 20 to 40 per cent in a few days. This fact combined with the selective action of carbon monoxide and of alkaloids on the enzymatic activity of germinating seeds makes one believe that the nitrogen assimilation in leguminous plants, as other authors have supposed, is due to the stimulating action of nitrogen bacteria, but not exclusively to these bacteria. The nitrogen assimilation is related to enzymatic activities which are latent in leguminous plants. In further confirmation of this are the new investigations that we are making now; from which it appears that the alkaloids also are able to promote nitrogen assimilation by lupins and peas.

The other effects, observed up to now in different seeds, in relation to different enzymatic activities, are summarised in the accompanying table:

| Seed. | Exciting Substance. | Percentage Variation of the Enzymatic Activity. | | | |
|---|---|---|---|---|---|
| | | Lipase. | Per-oxidase. | Amylase. | Protease. |
| *Ricinus* | Carbon monoxide (50 per cent air) | +5 | .. | −70 | .. |
| ,, | Strych. 1 °/₀₀ pH = 4·6 | +20 | +150 | +4 | +10 |
| ,, | Strych. 1 °/₀₀ pH = 6·8 | from +50 to +250 | .. | .. | .. |
| ,, | Ricinine 1 °/₀₀ | +30 | .. | .. | .. |
| Barley | Caffeine 0·5 °/₀₀ | .. | .. | −20 | .. |
| ,, | Strych. 0·5 °/₀₀ | .. | .. | +80 | .. |
| ,, | Carbon monoxide | .. | .. | −70 | .. |

Investigations on these lines are being continued in this Institute, and will be extended to other kinds of plants and alkaloids.

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R. Scuola Superiore di Chimica Industriale, Bologna, March 12.

[1] M. Padoa, *Giorn. Chim. Ind. Appl.*, **11**, 504; 1929: **12**, 247, 495; 1930.
[2] M. Padoa e A. Spada, *Giorn. Biologia Appl. all' Ind. Chim.*, **1**, Nos. 1, 3, 5, 6; 1931.
[3] M. Padoa e Nerina Vita, *Ann. Chim. Appl.*, **19**, No. 4; 1929. *Biochem. Z.*, **244**, Nos. 4-6; 1932.
[4] Nerina Vita, *Biochem. Z.*, **245**, Nos. 1-3; 1932.

### Colour Response in a Leech

COLOUR change in response to environmental conditions has been described in the following groups: vertebrates, crustaceans, and certain molluscs. There are two types of mechanism by which rapid redistribution of the pigment in the skin is brought about. In molluscs, the pigment is contained in minute elastic bags, which can be expanded by means of a specially arranged musculature, and contract because of their own elasticity. In crustaceans and vertebrates, the pigment is present as granules inside stellate cells, and can migrate through the protoplasm, being either localised in the centre of the cell or distributed more or less evenly through its processes.

The common fresh-water leech *Glossosiphonia complanata* has conspicuous stellate cells in its skin, containing brown or black pigment. I have observed that the pigment in these cells undergoes changes in configuration, contracting together if the animal is kept in total darkness, and spreading out into the stellate shape if it is kept in light. They are therefore chromatophores of the vertebrate-crustacean type. Curiously enough, they are so few and so widely spaced that their activity has little or no effect on the general coloration of the leech.