

From these observations, it appears that *O*-substitution in the 2 position of L-fucose enhances the H(0) activity of the parent compound whereas simultaneous ether formation in positions 3 and 4 sharply decreases the serological potency. The above findings make it likely that 2-*O*-methyl-L-fucose in a specific linkage [c.f. ⁵, ⁶] is the major factor responsible for the blood group H(0) activity of *Taxus cuspidata*, but fructose and arabinose may contribute to the activity of the macromolecule since WATKINS and MORGAN⁵) found these sugars slightly inhibiting in the H(0) anti-H eel system. To the best of our knowledge the 2-methyl-ether of L-fucose has not yet been found in nature.

A systematic study on fucose ethers and their immunological properties will be published elsewhere.

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- 1) SPRINGER, G.F.: *Naturwiss.* 42, 37 (1955).
- 2) SPRINGER, G.F.: *J. Immunology* (in press).
- 3) SPRINGER, G.F.: *Federat. Proc.* 1956, No. 2003. — SPRINGER, G.F.: *Naturwiss.* 43, 93 (1956).
- 4) MORGAN, W.T.J.: In Josiah Macy, Jr. Foundation, Transactions: Polysaccharides in Biology I, Princeton, April 1955, ed. by G.F. Springer.
- 5) WATKINS, W.M., and W.T.J. MORGAN: *Nature* [London] 169, 825 (1952).
- 6) KUHN, R., and H.G. OSMAN: *Z. physiol. Chem.* 303, 1 (1956).
- 7) HIRST, E.L., L. HOUGH and J.K.N. JONES: *J. Chem. Soc. [London]* 1949, 928.
- 8) CONCHIE, J., and E.G.V. PERCIVAL: *J. Chem. Soc. [London]* 1950, 827.
- 9) PARTRIDGE, S.M.: *Biochemic. J.* 42, 238 (1948).
- 10) WALDRON, D.M.: *Nature* [London] 170, 461 (1952). — EDWARD, J.T., and D.M. WALDRON: *J. Chem. Soc. [London]* 1952, 3631.

Undefined Adrenocortical Factor and Extraadrenal Participation in the Acute Inhibition of Testicular Extract Induced Edema in Rats

We published some observations on the long-term suppression of a permeability response of the rats' hind-paw, injecting at the plantar side 0.3 to 0.8 mg. of bull-testis extract dissolved in 0.11 ml. saline¹). Results of several acute experiments, investigating edema formation in 85 adult rats of 165 to 215 g., 7 to 10 hours after the control edema was induced and the different injections were given, are the subject of the present communication. Details of reading of the developing edema are given in ²). The inhibition of edema is signed as "good", if no remarkable edema develops around the tibia.

A. We demonstrate that excessive doses (20 to 35 I.U.) of intravenous corticotrophine (RMC-Roskilde), given in two parts, frequently evoke a good inhibition, i.e. in 8 out of 12 rats. A smaller per cent inhibition could be produced by administering the depot-preparation Cortrophine-Z (Organon-Oss), in doses of 30 to 40 I.U., i.e. in 7 out of 14 rats. Cortrophine-Z was somewhat effective also in the adrenal demedullated rat, operated 3 to 4 months earlier, and having the adrenocortical weight of more than 12 mg. per 100 g., i.e. in 5 out of 10 cases. At present we do not have explanation for the divergencies in this group. Both preparations, however, fail to influence edema formation in the adrenalectomized (6 rats)⁴). In conclusion: excessive adrenocortical stimulation could be able to produce an acute inhibition of this type of edema. In earlier papers we reported on the seemingly similar action of high salicylate (or adrenaline) doses²,³). It is important to note, however, that if we accept that the excessive corticotrophine doses used, represent maximal adrenocortical stimulus, another action of salicylate-injury must be suggested too: because sufficiently high salicylate (or adrenaline) doses regularly inhibit edema. The nature of this extraadrenal factor, and its interaction with the adrenocortical participant, however, remains obscure.

B. Cortisone, or hydrocortisone (20 to 50 mg. s.c.) do not inhibit edema within 10 hours either in the normal, or in the

adrenalectomized, i.e. 23 out of 25 rats remain uninfluenced⁵). Also 5 to 10 mg. of free hydrocortisone, given intraperitoneally, remained ineffective in 4 further cases. It seems that the adrenocortical material being responsible for an acute corticotrophine inhibition of edema will be other than cortisone-hydrocortisone. According to the literature, the known adrenocortical hormones of the rat, corticosterone and aldosterone, hardly inhibit acute edema. No decisive experiments were, however, hitherto reported.

C. An other control experiment was carried out, in which Cortrophine-Z remained ineffective against edema formation in the adrenalectomized animal, also in the presence of the above amounts of cortisone-hydrocortisone, i.e. in 12 out of 14 cases. An extraadrenal effect of corticotrophine, as regards edema formation, could not be, therefore, demonstrated — even in the presence of great amounts of cortisone-hydrocortisone. The extraadrenal factor of salicylate injury, therefore, seems to be other than corticotrophine participation. In fact: although amounts of some adrenocortical factor are indispensable for its effect, the edema inhibiting action of a massive subcutaneous dose of salicylate (525 to 650 mg. pro kg.) is somewhat different.

We succeeded in inhibiting edema as soon as 3 to 4 hours after salicylate, by administering heavy salicylate doses (= ineffective against edema in the adrenalectomized) to adrenalectomized rats, if the above (similarly ineffective) amounts of cortisone-hydrocortisone were given 6 to 7 hours before. This inhibition, however, although requiring higher cortisone supply, remarkably surpassing the so-called "permissive" amounts, seemed to be the function of the heavy salicylate dose²).

In conclusion: corticotrophine amounts do not inhibit testicular extract induced edema in the adrenalectomized rat, even if cortisone is administered simultaneously and in a greater dose, whilst salicylate amounts, given to the cortisone treated under similar conditions, do so⁶). The experiments reported here indirectly suggest the existence of some hitherto undefined, indispensable adrenocortical factor, participating in the acute inhibition of the testicular extract induced edema of the rat.

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- 1) KELEMEN, E., B. TANOS, L. HAJDU and P. FORGÁCS: *Nature* [London] 175, 122 (1955).
- 2) KELEMEN, E.: *Acta med. hung.* 9, 125 (1956).
- 3) KELEMEN, E., M. MAJOROS, J. IVÁNYI and K. KOVÁCS: *Experientia* [Basel] 6, 435 (1950).
- 4) The number of these trials is greater, if we take into account trials of group C.
- 5) A cortisone-induced inhibition begins to appear 24 to 72 hours later¹).
- 6) Recent experiments demonstrate that although adrenal medulla and the pituitary, both participate in the salicylate inhibition of the intact animal, this participation — in contrary with the adrenocortical factor — does not seem indispensable.

Stem Elongation in a Rosette Plant, Induced by Gibberellic Acid¹)

Gibberellin *A* and gibberellic acid are two compounds which are produced by the mold *Gibberella fujikuroi* (Saw.) Wr. [the sexual stage of *Fusarium moniliforme* (Sheld.) Snyder & Hansen emend.]²), and which have been found to increase the degree of stem elongation in numerous higher plants [rice, barley, buckwheat, cucumber, *Ipomoea*, etc.³), wheat⁴), peas⁵), maize⁶)]. The purpose of the present note is to show that these compounds are also capable of causing stem growth in a plant under conditions which normally permit only the formation of a leaf rosette with an extremely shortened axis.

The plant used was a biennial strain of *Hyoscyamus niger* L., the henbane. This plant requires, first, an exposure to low temperature, and subsequently, long days in order to flower as well as to elongate. Otherwise, it remains vegetative and acaulous. The experimental plants were grown in a greenhouse in which the temperature was not permitted to drop below 20° C, the maximal temperature which can induce flowering and stem elongation being 17° C⁷). Part of the treatments was done under long-day, part under short-day conditions (18—20 hrs. and 9 hrs. of light daily, respectively). The crystalline gibberellin preparation used in this study consisted of a mixture of gibberellic acid and gibberellin *A*

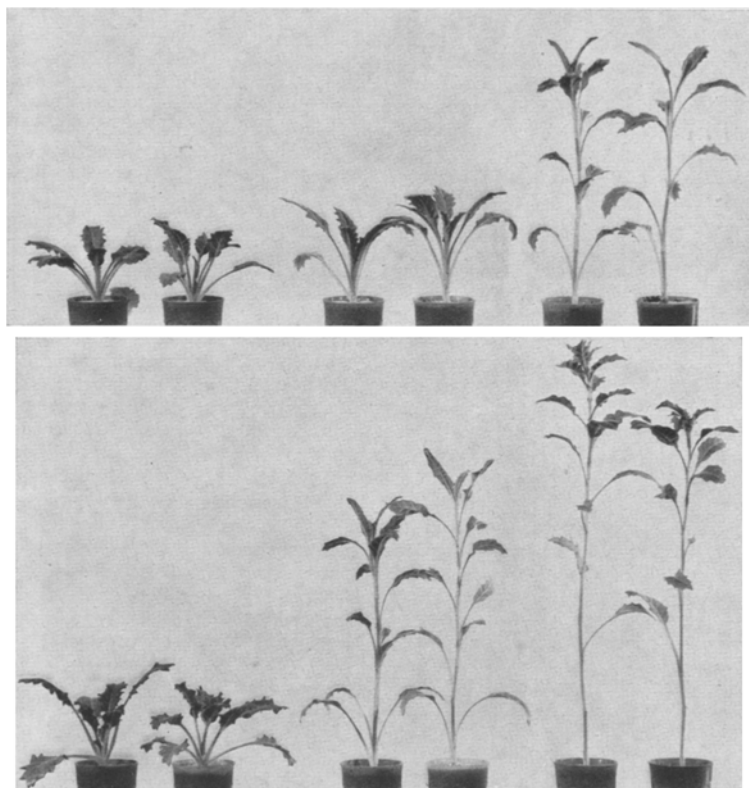


Fig. 1. Effect of Ga on stem elongation on biennial *Hyoscyamus*. Above: treatment in short days, below: in long days. From left to right, two plants each: controls (plants treated with Polyglycol 31-containing water only); plants treated with 10 mg./l. of Ga; plants treated with 50 mg./l. of Ga

($[\alpha]_D^{25} = 62^\circ$). For the sake of brevity, this preparation will henceforth be called Ga. It was used in aqueous solution, to which a wetting agent (Polyglycol 31, Dow Company, Seal Beach, Cal., USA.) had been added. The solution was applied with an eye dropper into the center of the rosette. The treatment consisted of a daily application of 0.2 cm³ during a period of four weeks. The measurements of stem elongation were taken one week after the last application.

Table 1. The effect of Ga on stem elongation in biennial *Hyoscyamus*

| Ga | | Treatment in long day | | Treatment in short day | |
|------------------------|----------------------------|-------------------------------|---------------------------|-------------------------------|---------------------------|
| Concentration (mg./l.) | Total amount ^{a)} | Stem elongation ^{b)} | Stem length ^{c)} | Stem elongation ^{b)} | Stem length ^{c)} |
| 0 (control) | 0 | — | — | — | — |
| 0.01—1.0 | 0.06—6.0 | — | — | — | — |
| 10 | 60 | 15 | 27 | 27 | 6 |
| 50 | 300 | 8 | 36 | 13 | 27 |

a) Total amount per plant ($\mu\text{g.}$); b) Beginning of stem elongation (days); c) Stem length after 5 weeks (cm.).

Table 1 summarizes the results. It is evident that, under conditions which normally maintain the rosette habit of growth in biennial *Hyoscyamus*, treatment with 10 mg./l. and 50 mg./l. of Ga has caused striking stem elongation (see Fig. 1). In these two treatments, the total amounts of Ga which were administered to each plant, were 60 $\mu\text{g.}$ and 300 $\mu\text{g.}$, respectively, and the first response became visible when a total of 30 $\mu\text{g.}$ and 80 $\mu\text{g.}$ had been administered. A total of 6 $\mu\text{g.}$ per plant (1 mg./l.) was without effect. The minimal dosage which is necessary for a response lies thus between 6 $\mu\text{g.}$ and 30 $\mu\text{g.}$ per plant. This is considerably more than the amounts which are needed to enhance stem elongation in non-rosette plants. Thus, in certain pea varieties⁵⁾ and in certain dwarf mutants of maize⁶⁾, as little as 0.01 $\mu\text{g.}$ gibberellic acid per plant result in a measurable response, although much more is required to make these varieties and mutants grow like normal strains. A reason for the difference in the minimum levels of Ga which are necessary for response in *Hyoscyamus*

as compared to non-rosette plants lies possibly in the circumstance that the action of gibberellic acid (and/or gibberellin A) seems to be qualitatively different in these two plant types. The effect of the substance in non-rosette plants seems to be predominantly, if not exclusively, upon cell elongation [see ⁸⁾]. Since in a *Hyoscyamus* rosette, stem internodes are practically nonexistent, the application of Ga in this plant must be causing additional cell division before cell elongation can take place. It should also be noted that the comparatively high amounts of Ga which were applied did not produce any abnormal features in the plants. With respect to stem elongation, leaf size and shape, and plant color the Ga treated *Hyoscyamus* plants resembled plants which had received a cold and a long-day treatment. In particular, none of the effects encountered in treatments with high auxin dosages were observed. When comparable amounts of indole-3-acetic (up to 500 $\mu\text{g.}$ per plant) were applied in the same manner as Ga and during the same period of time, a reduction in the size of the new leaves occurred, and their petioles showed some curvature. However, the auxin failed to induce any stem elongation.

Fig. 1 and Table 1 also show that the effect of Ga on biennial *Hyoscyamus* is considerably more pronounced when the treatment is given under long-day conditions. 50 mg./l. of Ga were needed in short days for the effect which in long days was produced by 10 mg./l. This suggests that, at least as far as stem elongation is concerned, Ga acts in that developmental step which is normally completed under the specific influence of low temperature. Ga seems to act only to a lesser extent in the subsequent step which normally requires long days for its completion.

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²⁾ YABUTA, T *et al.*: J. Agric. Chem. Soc. Japan **15**, 257 (1939); **17**, 721 (1941). — CURTIS, P.E., and B.E. CROSS: Chem. and Ind. **1954**, 1066.

³⁾ YABUTA, T., and T. HAYASI: J. Agric. Chem. Soc. Japan **15**, 403 (1939).

⁴⁾ KATO, J.: Mem. Coll. Sci., Kyoto Imp. Univ. [B] **20**, Nr. 3, 189 (1953).

⁵⁾ BRIAN, P.W., and H.G. HEMMING: Physiol. Plantarum **8**, 669 (1955).

⁶⁾ PHINNEY, B.O.: Proc. Nat. Acad. Sci. USA. **1956**.

⁷⁾ LANG, A.: Züchter **21**, 241 (1951).

⁸⁾ SUMIKI, Y.: J. Agric. Chem. Soc. Japan **26**, 393 (1952). — BRIAN, P.W. *et al.*: Physiol. Plantarum **8**, 899 (1955). — PHINNEY, B.O.: Personal communication.

Increase of the Non-Protein Amino Nitrogen in Sea-Urchin Eggs upon Fertilization

Investigations from this Laboratory¹⁾ have led to the suggestion that as an immediate result of the activation of the sea-urchin egg a process of re-arrangement of the egg proteins is initiated. An increased activity of cytoplasmic protease(s) has also been demonstrated²⁾ thus suggesting a faster renewal of the egg proteins. Some time earlier, ÖRSTRÖM³⁾ had found an increase of the non-protein N, mostly to be accounted for as an increase of histidine, in the eggs of *Paracentrotus* during the first ten minutes following fertilization. On the other hand, according to KAVANAU⁴⁾, an increase or a decrease of the free amino acids content takes place at fertilization depending on whether the eggs come from freshly collected animals or from ones which had been kept for some time under conditions which prevent spawning.

Hence it appeared important to check whether ÖRSTRÖM's results could be confirmed on two of the most common sea-