
ATTI ACCADEMIA NAZIONALE DEI LINCEI
CLASSE SCIENZE FISICHE MATEMATICHE NATURALI
RENDICONTI

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A comparison between the effects of fusicoccin and some of its derivatives on seed germination and on cell enlargement

Atti della Accademia Nazionale dei Lincei. Classe di Scienze Fisiche, Matematiche e Naturali. Rendiconti, Serie 8, Vol. 55 (1973), n.5, p. 555–558.
Accademia Nazionale dei Lincei

<http://www.bdim.eu/item?id=RLINA_1973_8_55_5_555_0>

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SEZIONE III

(Botanica, zoologia, fisiologia e patologia)

Fisiologia vegetale. — *A comparison between the effects of fusicoccin and some of its derivatives on seed germination and on cell enlargement* (*). Nota di M. IDA DE MICHELIS, presentata (**) dal Corrisp. E. MARRÈ.

RIASSUNTO. — La fusicoccina, che stimola come o più che le auxine la crescita per distensione nei tessuti dei fusti, è stata pure recentemente dimostrata capace di un'azione di stimolo pari o maggiore di quella delle gibberelline e delle citochinine sulla germinazione di semi quiescenti. Ciò apre il problema se il recettore primario del composto sia lo stesso nei due processi. In un tentativo preliminare di risolvere questo problema si è confrontata l'azione sulla distensione (test internodio di pisello) e rispettivamente sulla germinazione (test semi di lattuga var. Grand Rapids) della fusicoccina e di tre suoi derivati. La fusicoccina e il suo dideacetil derivato sono risultati parimenti efficaci sia sulla crescita per distensione che sulla germinazione, mentre l'epimero in 9 del dideacetildederivato e il corrispondente 8-chetoderivato sono apparsi del tutto inattivi.

Questi risultati sono in accordo con l'ipotesi che l'azione della fusicoccina investa lo stesso recettore primario in entrambi i processi.

INTRODUCTION

Fusicoccin (FC), a diterpene glucoside produced by *Fusicoccum amygdali* [1], has been shown to stimulate cell enlargement strongly in a number of plant tissues [2, 3]. Recent work in this laboratory shows that the same compound is also able to promote seed germination and to counteract the inhibitory effects of agents such as abscisic acid or irradiation with infra red [4]. The two processes of cell enlargement and seed germination have in common the characteristic of being both at a large extent controlled by hormonal stimulation.

The response to the various hormones is, however, quite different in the two cases, auxins being generally much more active on cell elongation, while gibberellins and cytokinins are more active on germination.

It seems therefore important to understand whether the strong promoting action of FC on the two processes is mediated by a single or by a dual mechanism of action. In the present paper a preliminary attempt to answer this question has been carried out by comparing the effects of FC and 3 of its derivatives, which were kindly supplied to us by Prof. A. Ballio, on cell enlargement with those on seed germination.

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(**) Nella seduta del 26 novembre 1973.

ABBREVIATIONS: FC, fusicoccin; DAF, dideacetyl-fusicoccin; EDAF, 9-epi-dideacetyl-fusicoccin; DAK, 8-oxo-9-epi-dideacetyl-fusicoccin.

The derivatives of FC used in this work are: dideacetyl-fusicoccin (DAF), its epimer in 9, 9-epi-dideacetyl-fusicoccin (EDAF) and the corresponding 8 keto derivative 8-oxo-epi-dideacetyl-fusicoccin (DAK). DAF had already been reported by Ballio *et al.* [5] as just as active as FC on cell enlargement. No data are available on the physiological activity of EDAF and DAK.

MATERIALS AND METHODS

The effects on cell enlargement were investigated by using the pea internode segments test, according to the procedure described by Lado *et al.* [2]. The experiments were run on samples of 12 sections in 4 ml of different solutions.

The effects on germination were evaluated by determining the rate of germination of lettuce seeds (variety Grand Rapids) incubated in Petri dishes on filter paper moistened with different solutions, in the dark at 25°C, under the experimental conditions described by Lado *et al.* [4]. FC, DAF, EDAF and DAK were dissolved in ethanol and made up to volume with distilled water to obtain stock solutions 6×10^{-4} M containing 10^{-1} M ethanol.

RESULTS

A. - *Cell enlargement* (pea internode test). The data in fig. 1 confirm that DAF, at 10^{-5} M concentration is just as active as FC (1.5×10^{-5} M) in promoting cell enlargement in this material.

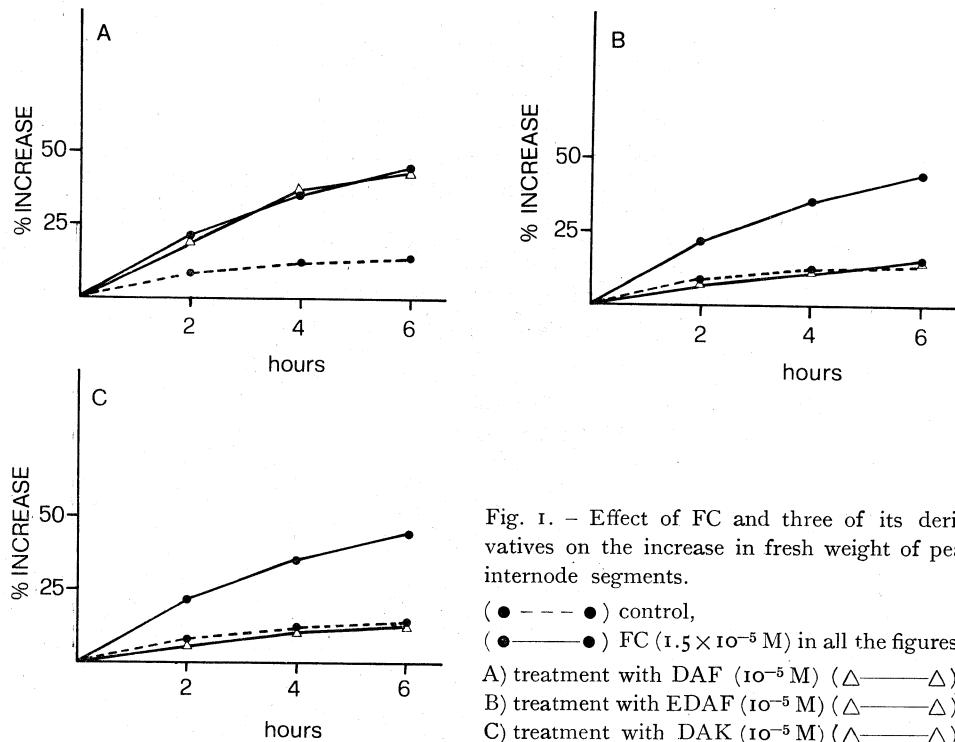


Fig. 1. - Effect of FC and three of its derivatives on the increase in fresh weight of pea internode segments.

- (● - - - ●) control,
- (● — ●) FC (1.5×10^{-5} M) in all the figures.
- A) treatment with DAF (10^{-5} M) (Δ — Δ);
- B) treatment with EDAF (10^{-5} M) (Δ — Δ);
- C) treatment with DAK (10^{-5} M) (Δ — Δ).

EDAF and DAK appear completely inactive not only at the concentration indicated in figg. 1-B and 1-C (10^{-5} M), but also in the concentration range between 3×10^{-5} M to 3×10^{-6} M.

B. - *Germination of lettuce seeds.* As shown in fig. 2-A, FC appears very active in increasing the rate of germination of lettuce seeds in the dark. The main effect seems that of a consistent acceleration of the process, the optimal FC concentration in the germination medium being around 1.5×10^{-5} M.

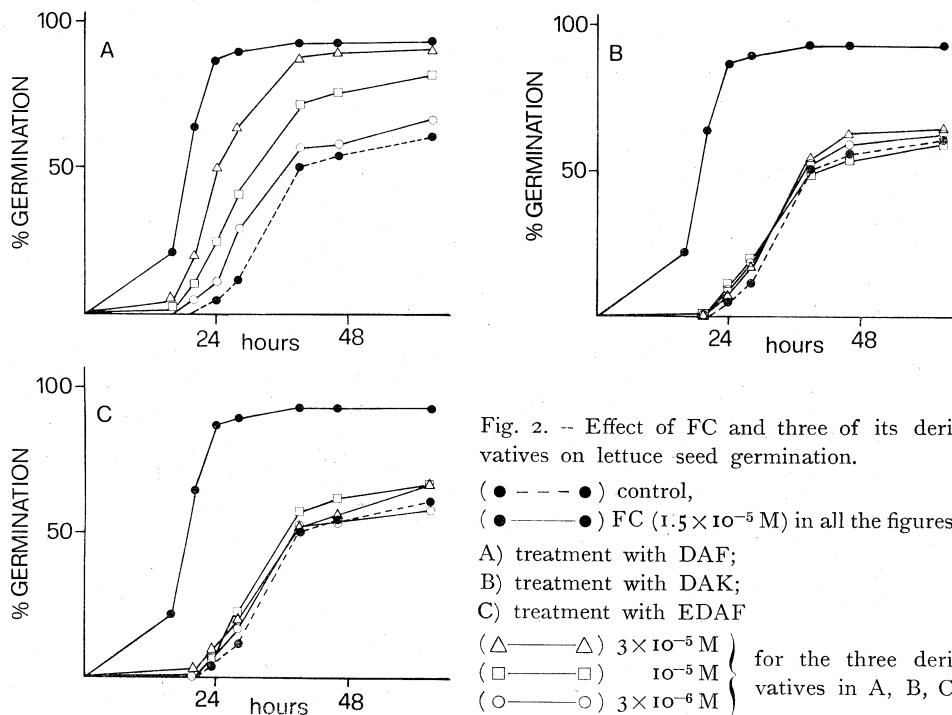


Fig. 2. -- Effect of FC and three of its derivatives on lettuce seed germination.
 (●---●) control,
 (●—●) FC (1.5×10^{-5} M) in all the figures.

A) treatment with DAF;
 B) treatment with DAK;
 C) treatment with EDAF
 (Δ—Δ) 3×10^{-5} M } for the three derivatives in A, B, C.
 (□—□) 10^{-5} M }
 (○—○) 3×10^{-6} M }

At higher concentrations toxic effects of FC become apparent, such as irregular bending of epicotyl and roots, abnormalities of root and shoot morphology, etc. These results are in full agreement with what had been observed in this laboratory by Lado *et al.*

The data of fig. 2 also show that the comparative efficiencies of the 3 derivatives on this germination test are exactly the same as those observed for the pea internode cell enlargement test: namely, DAF is practically just as active as FC, while EDAF and DAK are completely ineffective, in the whole concentration range investigated.

DISCUSSION AND CONCLUSIONS

The results reported above can be summarized as follows:

- 1) It is confirmed that the acetyl groups in 3' and 19 are not essential for the activity of FC on cell enlargement in the pea internode test [5].

In contrast, when the stereochemistry in 9 is reversed, as in EDAF and DAK, the growth promoting action of FC is completely suppressed.

2) In agreement with previous findings in this laboratory [4] FC is found to markedly accelerate the rate of germination of lettuce seeds.

In this germination test, just as in the above mentioned cell enlargement test, DAF is just as active as FC, while EDAF and DAK appear completely inactive.

The differences in structure between FC and the derivatives tested [6] in this investigation do not appear such as to markedly change their capacity to penetrate into cells (presumably by passive diffusion). It seems therefore probable that the observed differences of activity between FC and DAF, on one hand, and EDAF and DAK, on the other, are due to the fact that the chirality of carbon atom 9 is critical for the interaction between FC (or its derivatives) and its (presumably proteic) receptor in the cell. If it is so, the present data should be interpreted as a preliminary indication that the interaction with the same receptor is mediating the effects of FC both on seed germination and on cell enlargement.

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