recently, an inhibiting effect of OPH on the calcium-calmodulin system has been shown. The effect of OPH during the early phases of radish (*Raphanus sativus* L.) seed germination has been investigated. Seed germination is characterized by the reactivation of membrane functions: proton extrusion, evolution of potassium transport (leakage and following reabsorption), and negative transmembrane electric potential.

OPH inhibited the increase in fresh weight, which takes place in early germination, at least up to the first 40 h. This inhibition was accompanied by a delay in the reactivation of proton extrusion and potassium uptake; the leakage of this latter cation was not affected by the toxin. The leakage of calcium, which takes place during early germination, was enhanced by OPH. OPH inhibited the incorporation of labeled precursors into both proteins and RNA. The presence of calcium in the incubation medium did not affect H^+ extrusion, K^+ uptake, and fresh weight increase. The administration of EGTA or calmidazolium (a powerful inhibitor of calmodulin) enhanced the inhibitory effect of OPH. OPH inhibited the *in vitro* activation of the calmodulin-dependent brain phosphodiesterase, with a kinetics similar to the one found in maize. Since the inhibition was relevant only at high OPH concentration and after a long preincubation period, the effect might not be physiologically significant.

These results show that in germinating radish seeds the effect of OPH is accompanied by enhanced calcium permeability, in contrast with what previously found in maize roots. These results might suggest that OPH acts at membrane level through the calcium-calmodulin system, even though the *in vitro* effect of OPH on calmodulin does not appear sufficient to support the hypothesis that the *in vivo* effect of the toxin depends on an inhibition of the calcium-calmodulin system.

STRATEGIES FOR SELECTING *ARABIDOPSIS THALIANA* MUTANTS RESISTANT TO FUSICOCCIN

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We are interested in developing selective systems for the isolation of *Arabidopsis* mutants altered in their sensitivity to fusicoccin (FC). Preliminary experiments demonstrated that FC induces in *Arabidopsis* (like the majority of higher plants) a wide variety of physiological responses. Among them, FC promotes germination, overcomes ABA inhibition on germination, induces stomata opening and wilting. At the biochemical level, FC stimulates the activity of a K^+-dependent, erythrosin B and vanadate-sensitive H^+-ATPase located in the plasmamembrane. The receptor site for FC in *Arabidopsis* seems to be a 34 Kda polypeptide with properties similar to the high affinity FC binding sites of other higher plants.

Arabidopsis mutants resistant or hypersensitive to FC can be isolated when a selectable phenotype, directly related to the action of the toxin, is identified: this

implies the development of selective systems easily and clearly differentiating treated individuals from controls.

Selection procedures based on the action of FC on germination (stimulation of germination, or counterinhibition of ABA effect) proved to be unsuitable because the toxin at all concentration tested (from 10^{-7} M to 10^{-3} M) does not allow a clear discrimination between treated and control seeds. The same is true when the loss of turgor elicited by FC on plants exposed to water stress is evaluated.

More promising seem procedures based: a) on the indirect evaluation of the increased transpiration rate induced by FC, and b) on the synergistic effect of the toxin with the herbicide Paraquat.

Stomatal opening induced by FC (10^{-5} M) on well watered plants incubated at 25 °C in the dark for 12h entertaines a transpiration rate three times higher than that of control plants. Since evapo-transpiration is the major component in the energy balance of leaf, stomatal opening results in cooling of the leaf surface (about 1 °C of difference between treated and control plants) which can be easily visualized with infrared thermography.

The second approach takes the advantage of the synergistic effect of FC with Paraquat. The herbicide, a lipophilic cation, by competing with ferredoxin for electrons from PS 1 and subsequent reoxidation by molecular oxygen, generates superoxide radicals which induce leaf bleaching. We found that in *Arabidopsis*, Paraquat at low doses (5×10^{-7} M) does not induce leaf bleaching after light exposure unless FC is present in the incubation medium. This might depend on a higher Paraquat uptake by the tissue treated with FC or on some metabolic change induced by the toxin. By this selection procedure, 11 seedlings among about 1.200 M2 plants have been selected for resistance to leaf bleaching induced by FC and Paraquat. M3 progenies of these seedling are actually under analysis for the confirmation of the phenotype and for the determination of the physiological basis of resistance.

EFFECT OF SEIRIDINS AND CYCLOPALDIC ACID ON TOBACCO PROTOPLASTS

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Several phytotoxins are produced *in vitro* by the species of *Seiridium* associated with canker diseases of cypress trees in the Mediterranean area. Seiridin and *iso*-seiridin are produced by *Seiridium cardinale*, a strain of *S. cupressi* and *S. unicorne*. In addition, the second species produces a more potent toxin, cyclopaldic acid. The present work was aimed to assay the effect of the three fungal metabolites on plant protoplasts.

Protoplast preparations were made from sterile shoot cultures of *Nicotiana plumbaginifolia*. The protoplasts were resuspended in a buffered mannitol solution