that these three physiological activities of pectic enzymes may be regulated by factors such as endo-polygalacturonase inhibiting proteins (PGIP) and pH.

Homogeneous endo-polygalacturonase (PG), purified from *Aspergillus niger*, was applied to potato medullary tissue disks. After 4 hrs of incubation at various pH, maceration of the tissue was measured and viability of cells was assessed by the Evans Blue staining procedure. The optimum pH for the macerating activity was between 5.0 and 5.5, in accordance with the optimum pH at which *A. niger* PG depolymerized polygalacturonic acid (PGA) *in vitro*. The *A. niger* PG also caused cell death of potato cells at pH 5.0. However, in the presence of an excess of PGIP purified from *Phaseolus vulgaris*, the macerating activity of PG was completely inhibited, while the killing activity was maintained. The experiments were repeated using PG purified from *Fusarium moniliforme* and similar results were obtained. In the presence of an excess of PGIP, *F. moniliforme* PG lost its macerating activity and maintained its killing activity.

Experiments performed with a homogeneous endo-pectate lyase (PL) purified from *Erwinia carotovora* gave similar results. PL depolymerized PGA *in vitro* at an optimum pH of 9.0 and had very little activity at pH 5.5. Macerating activity of PL was exhibited at pH 9.0, whereas the enzyme did not discernibly macerate the potato tissue at pH lower than 7.0. Killing activity, on the contrary, was displayed by PL at all the pH values tested, that is from 9.0 through 5.5.

In order to ascertain the role of PGIP in regulating the macerating and killing activity of fungal endopolygalacturonase, we have amplified by polymerase chain reaction (PCR) and cloned a 0.7 kb fragment of *Phaseolus vulgaris* genomic DNA. This fragment corresponded to the N-terminal coding region of the gene for PGIP. We will now screen both a genomic and a cDNA library of *P. vulgaris* in search of full lenght PGIP clones and use these clones to transform tomato plants *via Agrobacterium* Ti plasmid-derived vectors. Transformed plants will be analyzed for susceptibility to the toxic effects of fungal polygalacturonases.

ACTIVATION OF THE PLASMA MEMBRANE H^+-ATPase BY FUSICOCCIN

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The effect of fusicoccin (FC) on the plasma membrane (PM) H^+-ATPase activity has been characterized in a purified PM fraction obtained from radish seedlings by phase partitioning.

FC-induced stimulation of the PM H^+-ATPase is strongly pH dependent: the absolute increase in activity is maximal around pH 7 (thus shifting the pH optimum of the ATPase activity of about 0.3 pH units) and percent stimulation increases with the increase of pH up to 100-130% at pH 7.5.

Incubation of the membranes at 25 to 33 °C prior to FC treatment leads to a dramatic decrease of both PM H^+-ATPase stimulation and FC binding to the membrane, thus confirming the involvement of the receptor in FC-induced stimulation of the PM H^+-ATPase.

In the presence of 10 μM FC, stimulation of the PM H^+-ATPase is maximal within 3 minutes, indicating that the transduction of the signal from the FC-receptor complex to the enzyme is very rapid.

The functional molecular weight of the FC-stimulated ATPase (determined by the radiation-inactivation technique) is about 350,000, while that of basal activity is about 170,000 and that of the receptor is less than 50,000. The simplest interpretation of these results is that at least another protein, besides the FC receptor, is involved in FC-induced stimulation of the PM H^+-ATPase.

CHARACTERIZATION OF THREE PHYTOTOXIC COMPOUNDS FROM *PSEUDOMONAS SYRINGAE* PV. *PAPULANS*

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Blister spots of apple and pear, a serious disease present in several areas of North America and Europe, is caused by *Pseudomonas syringae* pv. *papulans*. The disease is characterized by dark brown blisters on fruits and tiny cankers on branches. Occasionally midvein necrosis and distorsions of leaves occur. Recent studies have shown that the organic extracts of *P. s.* pv. *papulans* culture filtrates had phytotoxic activity when assayed on apple and bean leaves. A chemical investigation of the crude residue left from the organic extracts made it possible to isolate three metabolites, which proved to be weakly toxic on bean and apple leaves.

The trivial names of papuline, *o*-hydroxynitropapuline and papulinone were assigned to the three toxic substances.

Spectroscopic studies (UV, IR, ^1H and ^{13}C-NMR and HR-MS) have shown that papuline is the methyl ester of β-phenyllactic acid also when compared with the synthetic methyl ester of (*S*)-2-hydroxy-3-phenylpropanoic acid. The above spectroscopic techniques integrated with ^1H-nOe experiments allowed to assign to the *o*-hydroxynitropapuline the structure of *meta*-hydroxy-*para*-nitro disubstituted derivative of papuline, and to papulinone the structure of 4-(1-hydroxy-2-phenylethyl)-4-carbomethoxyoxyetan-2-one, a new β-lactone.

The synthesis of papuline, *o*-hydroxynitropapuline and papulinone is constitutive in *P. s.* pv. *papulans*; however the presence of *L*-phenylalanine, a β-phenyllactic acid precursor, in the medium apparently increases their accumulation in culture.

The three substances are all chemically correlated with β-phenyllactic acid, a substance with plant growth activity. Papuline assayed at a physiological concentration (1-0.01 mM) also modulates tomato seedlings growth.