Phosphorylation-dependent Interaction between Plant Plasma Membrane \( \text{H}^+ \)-ATPase and 14-3-3 Proteins*

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The \( \text{H}^+ \)-ATPase is a key enzyme for the establishment and maintenance of plasma membrane potential and energization of secondary active transport in the plant cell. The phytotoxin fusicoccin induces \( \text{H}^+ \)-ATPase activation by promoting the association of 14-3-3 proteins. It is still unclear whether 14-3-3 proteins can represent natural regulators of the proton pump, and factors regulating 14-3-3 binding to the \( \text{H}^+ \)-ATPase under physiological conditions are unknown as well. In the present study \textit{in vivo} and \textit{in vitro} evidence is provided that 14-3-3 proteins can associate with the \( \text{H}^+ \)-ATPase from maize roots also in a fusicoccin-independent manner and that the interaction depends on the phosphorylation status of the proton pump. Furthermore, results indicate that phosphorylation of \( \text{H}^+ \)-ATPase influences also the fusicoccin-dependent interaction of 14-3-3 proteins. Finally, a protein phosphatase 2A able to impair the interaction between \( \text{H}^+ \)-ATPase and 14-3-3 proteins was identified and partially purified from maize root.

The plant plasma membrane \( \text{H}^+ \)-ATPase (1, 2) generates an electrochemical gradient across plant cell plasma membrane that provides the driving force for secondary active transport and for cell turgor maintenance. A number of central physiological processes, such as stomata opening, plasmolysis loading, or root ion uptake, depend on regulation of \( \text{H}^+ \)-ATPase activity. Despite a large body of evidence indicating that different stimuli, like hormones or light and stresses regulate these processes by affecting \( \text{H}^+ \)-ATPase activity, up to now very little is known about molecular mechanisms controlling \( \text{H}^+ \)-ATPase activity.

It was demonstrated that the C-terminal region of the enzyme is an autoinhibitory domain. In fact, proteolytic removal of this region (3) or heterologous expression of a C-terminal truncated enzyme results in an increase of \( \text{H}^+ \)-ATPase activity (4). It was suggested, on the basis of similarity of biochemical parameters of activation, that \( \text{H}^+ \)-ATPase stimulators such as lysophosphatidylcholine or the toxin from \textit{Fusarium oxysporum} formyldali, fusicoccin (FC), act through C-terminal domain displacement (5–7), which therefore could represent a common mechanism for different proton pump effectors. It was also originally proposed that FC did not bind directly to the \( \text{H}^+ \)-ATPase but rather to receptors located at the plasma membrane. A search for FC receptors led to their identification as members of the 14-3-3 eukaryotic protein family (8–10). This discovery shed some light on the FC mechanism of \( \text{H}^+ \)-ATPase activation. In fact it was clearly shown by different authors that FC is able to promote the association of 14-3-3 proteins with the C-terminal domain of the \( \text{H}^+ \)-ATPase (11–13), thereby activating the enzyme (14). These lines of evidence indicate that the FC receptor is made up of a complex between 14-3-3 and \( \text{H}^+ \)-ATPase and suggest that association of 14-3-3 proteins with the proton pump could represent a mechanism regulating the proton pump under physiological conditions.

It is a common property of 14-3-3 proteins that they have the capability to associate with target proteins through binding to consensus motifs containing phosphorylated residues (15, 16). Recently it was shown that a similar mechanism holds true also for plant 14-3-3 association with plant enzymes such as nitrate reductase (17, 18) and sucrose phosphate synthase (19, 20). Because it is known that \( \text{H}^+ \)-ATPase is a phosphorylated enzyme \textit{in vivo} (21–23), it is conceivable that a phosphorylation/dephosphorylation mechanism, by endogenous protein kinases and phosphatases, may regulate the association of 14-3-3 with the \( \text{H}^+ \)-ATPase.

In the present study \textit{in vivo} and \textit{in vitro} data are reported demonstrating that 14-3-3 proteins are able to associate with the \( \text{H}^+ \)-ATPase in a FC-independent manner and that the interaction depends on the phosphorylation status of the proton pump. We also demonstrate that a protein phosphatase 2A (PP2A) from maize roots affects the association of 14-3-3 proteins with the \( \text{H}^+ \)-ATPase.

**EXPERIMENTAL PROCEDURES**

**Chemicals**—FC was prepared according to Ballio et al. (24). Okadaic acid, microcystin-LR, and anti-PP2A antibodies were purchased from Calbiochem, (La Jolla, CA). \( \gamma^\text{32P} \text{ATP} \) (specific activity, 110 TBq/mmol) and thrombin were from Amersham Pharmacia Biotech. Alkaline phosphatase, from calf intestine, was from Roche Molecular Biochemicals. Protein kinase A, catalytic subunit, and myelin basic protein (from bovine brain) were from Sigma. Chemicals for gel electrophoresis were from Bio-Rad. All other reagents were of analytical grade.

**Plant Material**—Maize seeds (Ze a mays L. cv. Paolo) from Dekalb (Mestre, Italy) were germinated and grown in the dark for 6 days, as already described (25).

**In Vivo Incubation of Maize Roots with Okadaic Acid and FC**—Maize roots (8 g) were cut into small pieces (approximately 5 mm) and incubated in 20 ml of 20 mM Tris-Mes (pH 7.0), containing 300 mM sucrose, for 1 h at room temperature. When indicated, 1 \( \mu \text{M} \) okadaic acid (OA) and 10 \( \mu \text{M} \) FC were added.

**Purification of \( \text{H}^+ \)-ATPase from Maize Roots**—Two-phase partitioned plasma membranes were obtained from 200 g of maize roots as described previously (26). When indicated, plasma membranes were treated with 0.5% Triton X-100 (26). For the purification of \( \text{H}^+ \)-ATPase, plasma membrane proteins were solubilized with dodecyl-\( \beta \)-maltoside as described by Johansson et al. (27) and fractionated by anion exchange HPLC (26).
overnight at 4 °C. After incubation, the membrane was washed three times, dried, and subjected to autoradiography at 80 °C. A, lane C, H'-ATPase purified from control tissue; lane FC, H'-ATPase purified from maize roots incubated with 10 μM FC. B, lanes C and FC, as in A, but incubation with 32P-labeled 14-3-3 was performed in the presence of 10 μM FC. The experiment was repeated three times, and similar results were obtained.

Purification of Protein Phosphatase from Maize Roots—The cytosolic fraction obtained from the purification of H'-ATPase (supernatant from the 50,000 × g centrifugation) was used as starting material. Cytosol was dialyzed overnight at 4 °C against 20 mM Tris-Mes (pH 7.0) and loaded onto a DEAE-biogel (Bio-Rad) column (100 × 20 mm) equilibrated with 20 mM Tris-Mes (pH 6.5) containing 1 mM DTT and 1 mM PMSF (buffer A). Elution was performed in 40 min by a linear gradient from 0 to 0.5 mM NaCl in buffer A, at a flow rate of 2.0 ml/min.

Fractions of 2 ml were collected and tested for the ability to abolish the interaction between HPLC-purified H'-ATPase and 14-3-3 in an in vitro overlay assay (13). Active fractions were pooled, dialyzed, and concentrated to 3 ml in a Sartorius collodion bag (cut-off, 12 kDa) and concentrated to 3 ml in a Sartorius collodion bag (cut-off, 12 kDa) and loaded onto a Bio-sil TSK-250 (Bio-Rad) HPLC gel filtration column (7.5 × 300 mm), equilibrated in 20 mM Tris-Mes buffer (pH 7.0), containing 150 mM NaCl and 0.5 mM DTT, at 1.0 ml/min flow rate. Active fractions were collected and used for the biochemical characterization of protein phosphatase activity.

Identification of H'-ATPase-Dephosphorylating Protein Phosphatase—To identify protein phosphatase activities able to act on H'-ATPase, a functional assay was set up by testing the ability of chromatographic fractions to inhibit H'-ATPase/14-3-3 association. 2 μg of HPLC-purified plasma membrane H'-ATPase were incubated with 1 μg of each fraction in 50 mM Tris-Mes (pH 7.0), containing 10 mM MgCl2, 1.2 mM CaCl2, 1 mM EGTA (free calcium about 100 μM), 1 mM DTT, 1 mM PMSF, 10 μM leupeptin, 5 μM chymostatin, and 5 μM pepstatin, for 30 min at 37 °C. Treated H'-ATPase was then used in the overlay assay (13).

Protein Phosphatase Assay—Protein phosphatase activity was estimated using 32P-labeled myelin basic protein (MBP) as substrate. 32P-MBP was prepared by incubation of 1 mg of MBP in 20 mM Tris-HCl (pH 7.5), 100 mM NaCl, 12 mM MgCl2 with 10 units of protein kinase A (catalytic subunit) and 80 μCi of [γ-32P]ATP, 200 μM ATP in a final volume of 250 μl. After 30 min at 30 °C, 250 μl of 20% trichloroacetic acid was added, and the mixture was centrifuged for 2 min in a Ole-Dich microcentrifuge at 5000 × g. The supernatant was discarded, and the pellet was solubilized in 250 μl of H2O and trichloroacetic acid precipitation was repeated eight times. Finally the pellet was dried under reduced pressure and solubilized in 250 μl of 50 mM Tris-HCl (pH 7.0). The specific activity of the 32P-labeled MBP was 1 MBq/mg.

The phosphatase assay mixture (50 μl) contained 50 mM Tris-Mes (pH 7.0), 1 mM DTT, 1 mM PMSF, 10 μM leupeptin, 5 μM chymostatin, 5 μM pepstatin, 5 μM of 32P-labeled MBP, and 1 μg of purified protein phosphatase. When indicated, 10 mM MgCl2, 1.2 mM CaCl2, and 1 mM EGTA (100 μM free Ca2+) or various concentrations of OA and MC were added.

Western Blotting—SDS-PAGE was performed as described by Laemmli (28), in a Mini Protean apparatus (Bio-Rad). Proteins were transferred onto nitrocellulose membranes using a semidy LKB apparatus.
Association of 14-3-3 Proteins with the H\textsuperscript{+}-ATPase

Results

The H\textsuperscript{+}-ATPase Purified from Maize Roots Interacts in Vitro with 14-3-3 Proteins in the Absence of FC—The H\textsuperscript{+}-ATPase was solubilized and partially purified by anion exchange HPLC from maize roots previously incubated in the presence or in the absence of 10 \( \mu \)M FC (26). The partially purified H\textsuperscript{+}-ATPase (2 \( \mu \)g) from FC-treated and control tissue were run on 10% SDS-PAGE, blotted onto nitrocellulose membrane, and incubated with \( ^{32}\text{P} \)-labeled 14-3-3, both in the absence and in the presence of 10 \( \mu \)M FC in the incubation mixture.

Results shown in Fig. 1A demonstrate that 14-3-3 proteins associate with H\textsuperscript{+}-ATPase in the absence of FC and that in vitro preincubation of the tissue with FC increases the interaction with the proton pump; the extent of the increase was 40%, as estimated by densitometric analysis. In Fig. 1B, the effect of in vitro incubation of FC on the interaction between 14-3-3 proteins and H\textsuperscript{+}-ATPase from FC-treated and from untreated tissue is shown. It is worth noting that FC in vitro administration strongly enhances the interaction not only between 14-3-3 and H\textsuperscript{+}-ATPase purified from control tissue but remarkably (42%, by densitometric analysis) also with the H\textsuperscript{+}-ATPase purified from FC-treated tissue.

These results demonstrate that 14-3-3 proteins are able to interact with the H\textsuperscript{+}-ATPase also in a FC-independent manner and indicate that in vitro FC treatment affects the capability of the proton pump to associate in vitro with 14-3-3 proteins, suggesting that the phosphorylation status of the purified H\textsuperscript{+}-ATPase may be involved.

In Vivo Administration of OA Stimulates H\textsuperscript{+}-ATPase Activity and Increases Plasma Membrane-bound Levels of 14-3-3—OA, a strong protein phosphatase inhibitor, was administered in vivo to maize roots to ascertain whether the phosphorylation status of H\textsuperscript{+}-ATPase influences the association with 14-3-3 proteins and consequently H\textsuperscript{+}-ATPase phosphohydrolytic activity.

Maize roots segments were incubated with 1 \( \mu \)M OA or 10 \( \mu \)M OA, and 100 \( \mu \)M OA or 1 \( \mu \)M OA; and subjected to SDS-PAGE, and tested in the overlay assay. A, lane C, control H\textsuperscript{+}-ATPase; lane AP, H\textsuperscript{+}-ATPase incubated with alkaline phosphatase. B, lanes C and AP, as in A, but incubation with the \( ^{32}\text{P} \)-labeled 14-3-3 was performed in the presence of 10 \( \mu \)M FC. The experiment was repeated three times, and similar results were obtained.

(2 h, 0.8 mA cm\textsuperscript{-2}). Immunodetection of 14-3-3 and H\textsuperscript{+}-ATPase was performed according to Marra et al. (26), using the ECL detection system from Amersham Pharmacia Biotech, following the manufacturer’s instructions. Immunodetection of protein phosphatase was carried out using antibodies recognizing the catalytic subunit of mammalian PP2A (Calbiochem) and anti-rabbit secondary antibodies conjugated to horseradish peroxidase (Bio-Rad).

Analytical Methods—Protein concentration was determined by the method of Bradford (29) using bovine serum albumin as the standard. H\textsuperscript{+}-ATPase activity was assayed according to Serrano (30).

FIG. 3. Effect of alkaline phosphatase on the interaction between 14-3-3 proteins and H\textsuperscript{+}-ATPase. The overlay assay was run under the same conditions described in the legend to Fig. 1. HPLC-purified H\textsuperscript{+}-ATPase (2 \( \mu \)g) was incubated with 4 units of alkaline phosphatase from calf intestine for 30 min at 37 °C, subjected to SDS-PAGE, and tested in the overlay assay. A, lane C, control H\textsuperscript{+}-ATPase; lane AP, H\textsuperscript{+}-ATPase incubated with alkaline phosphatase. B, lanes C and AP, as in A, but incubation with the \( ^{32}\text{P} \)-labeled 14-3-3 was performed in the presence of 10 \( \mu \)M FC. The experiment was repeated three times, and similar results were obtained.

FIG. 4. Purification of protein phosphatase from maize roots. A, elution profiles. Upper panel, anionic exchange on DEAE-Biogel. Middle panel, HPLC anionic exchange on DEAE TSK-5-PW. Lower panel, gel filtration on HPLC Bio-sil TSK 250. Continuous lines represent absorbance at 280 nm, dotted lines indicate the [NaCl] elution gradient, and shaded bars indicate the inhibition of 14-3-3/H\textsuperscript{+}-ATPase interaction. The relative amount of bound 14-3-3 proteins was estimated from overlay assays using Scion Image software. B, effect of the protein phosphatase tested by overlay assay. 2 \( \mu \)g of HPLC-purified H\textsuperscript{+}-ATPase were incubated with 1 \( \mu \)g of the three-step purified fraction of protein phosphatase. After 30 min of incubation at room temperature, the sample was subjected to SDS-PAGE, blotted onto nitrocellulose, and analyzed in the overlay assay. Panel a, lane C, control H\textsuperscript{+}-ATPase; lane PP2A, H\textsuperscript{+}-ATPase incubated with protein phosphatase; lane PP2A+OA, as in PP2A, but in the presence of 1 \( \mu \)M OA. Panel b, as in panel a, but incubation with the \( ^{32}\text{P} \)-labeled 14-3-3 was performed in the presence of 10 \( \mu \)M FC.
Association of 14-3-3 Proteins with the H\textsuperscript{+}-ATPase

| Fraction | Protein | Total activity | Specific activity | Purification |
|----------|---------|----------------|------------------|--------------|
|          | mg      | pmol dephosphorylated MBP \cdot min \cdot mg\textsuperscript{-1} | pmol dephosphorylated MBP \cdot min \cdot mg\textsuperscript{-1} | fold          |
| Cytosol  | 320.0   | 3040.0         | 9.5              | 1            |
| DEAE-biogel | 13.2   | 310.2          | 23.5             | 2.5          |
| HPLC DEAE TSK-5-PW | 0.4  | 237.8         | 594.4            | 62.5         |
| HPLC Bio-sil TSK 250 | 0.025 | 32.2          | 1288.8           | 135.6        |

Fig. 5. Biochemical characterization of the protein phosphatase. A, effect of OA and MC on the MBP-dephosphorylating activity of the partially purified protein phosphatase. 5 μg of \({}^{32}\)P-labeled MBP was incubated with 1 μM of partially purified protein phosphatase in the presence of different concentrations of OA (\(\square\)) and microcystin-LR (\(\square\)). The illustrated data represent activity means ± S.E. for three independent experiments run in duplicate. B, immunoblotting of the partially purified protein phosphatase. 20 μg of the three step-purified protein phosphatase were subjected to SDS-PAGE and blotted onto nitrocellulose. Immunodetection was performed using antibodies recognizing the catalytic subunit of mammalian PP2A. Lane 1, Coomassie staining; lane 2, immunoblotting.

Identification of a Protein Phosphatase from Maize Roots That Negatively Regulates the Interaction between H\textsuperscript{+}-ATPase and 14-3-3 Proteins—Results obtained with alkaline phosphatase prompted us to investigate whether protein phosphatase activity able to modulate binding of 14-3-3 to H\textsuperscript{+}-ATPase were present in maize roots. To this purpose, cytosol from maize roots was fractionated by anion exchange DEAE-biogel chromatography followed by DEAE and gel filtration HPLC purification (Fig. 4A). Fractions were directly tested for their ability to inhibit the association between H\textsuperscript{+}-ATPase and 14-3-3 proteins in the overlay assay. Specificity of the protein phosphatase activity was evaluated by assaying fractions also in the presence of 1 μM OA and 1 μM MC. Relative amounts of bound 14-3-3 were estimated by densitometric analysis. In Fig. 4B the effect of the HPLC size exclusion-purified fraction on the interaction between H\textsuperscript{+}-ATPase and 14-3-3 proteins in the overlay assay is shown: protein phosphatase treatment of the H\textsuperscript{+}-ATPase nearly completely inhibited binding of 14-3-3 proteins. OA addition (1 μM) in the incubation mixture restored the interaction. Importantly, protein phosphatase treatment drastically reduced also the FC-dependent interaction between H\textsuperscript{+}-ATPase and 14-3-3 proteins.

Biochemical Characterization of the Partially Purified Protein Phosphatase—Biochemical and immunological analysis allowed us to ascertain that a protein phosphatase activity with properties typical of the PP2A family was present in fractions able to inhibit the H\textsuperscript{+}-ATPase/14-3-3 interaction. The characterization of the partially purified protein phosphatase (size exclusion fraction) was carried out using \({}^{32}\)P-MBP as substrate. Specific activity and yield of each chromatographic step are shown in Table I.

Protein phosphatase activity was completely independent of Ca\textsuperscript{2+} and Mg\textsuperscript{2+} ions in concentrations ranging from 1 μM to 1 mM (data not shown), whereas it was drastically affected (Fig. 5A) by OA (IC\textsubscript{50} = 0.9 nM) and MC (IC\textsubscript{50} = 0.6 nM), in accordance with data reported for PP2A (31). A further confirmation was provided by Western blotting analysis using antibodies raised against a conserved sequence of the catalytic domain of mammalian PP2A. As shown in Fig. 5B, the antibodies recognized a single polypeptide of approximately 37 kDa, the same molecular mass reported for the catalytic subunit of plant PP2A (31).
Association of 14-3-3 Proteins with the H¹-ATPase

DISCUSSION

Results from recent research have contributed much progress to the understanding of the mechanism of H¹-ATPase stimulation by the phytotoxin FC. In fact it was clearly demonstrated by different authors that FC promotes the irreversible association of stimulatory 14-3-3 proteins with the H¹-ATPase. Because an ever-increasing body of evidence points to 14-3-3 proteins as general regulators of key enzymes in the plant cell (32), results from FC research also suggest that 14-3-3 proteins may be involved in the regulation of H¹-ATPase under physiological conditions. In fact, different pieces of evidence (11, 13) suggest that 14-3-3 proteins can also interact with the proton pump in a FC-independent manner. Although the molecular basis of the FC-independent interaction between H¹-ATPase and 14-3-3 proteins is still unknown, phosphorylation/dephosphorylation events on the H¹-ATPase are likely, because a common feature of 14-3-3 association with target proteins involves the interaction with consensus sequences containing a phosphorylated amino acid (15, 16).

This work was aimed at ascertaining whether a similar mechanism can account for the interaction between 14-3-3 proteins and H¹-ATPase. To this purpose, the ability of H¹-ATPase to associate in vitro with 14-3-3 proteins after in vivo treatments able to alter its phosphorylation status was investigated. Data reported here show that in vivo incubation of maize roots with OA provoked a significant increase of the plasma membrane-bound 14-3-3 proteins, as well as a strong stimulation of H¹-ATPase activity. These results strongly suggest that phosphorylation of H¹-ATPase is essential for its binding to 14-3-3 proteins and concomitant stimulation. We also demonstrated that FC in vivo incubation of maize roots, a treatment resulting in an increase of membrane-bound 14-3-3 and stimulation of H¹-ATPase activity, was able to increase the in vitro interaction of H¹-ATPase with 14-3-3 proteins. Also this finding is suggestive of the occurrence of a post-translational modification on the proton pump, influencing its capability to interact with 14-3-3 proteins. Interestingly, Olsson et al. (23) demonstrated that in vivo FC treatment protects from dephosphorylation a phosphothreonine residue (Thr-948 in the Arabidopsis isoform AHA1) located in the very end of the C-terminal domain of the H¹-ATPase. It was hypothesized that the effect is due to direct binding of 14-3-3 to the phosphorylated threonine or alternatively to a 14-3-3-induced conformational change protecting the amino acid from dephosphorylation. Our results indicate that the phosphothreonine residue may be physically involved in the binding of 14-3-3 proteins, being part, as proposed by Olsson at al. (23), of a novel consensuss sequence regulating the physiological association of 14-3-3 proteins with the H¹-ATPase.

Furthermore, in vivo data indicate that the phosphorylation status of the H¹-ATPase is relevant also for the FC-dependent binding of 14-3-3 protein; in fact, the H¹-ATPase purified from in vivo FC or OA-treated roots bound 14-3-3 protein more efficiently also in the presence of FC. This latter result is also worth noting, because results obtained up to now point instead to different, independent mechanisms for phosphorylation and FC-mediated binding of 14-3-3 proteins to the H¹-ATPase (13, 14).

A further demonstration of the relevance of phosphorylation as a regulatory mechanism was provided by in vitro dephosphorylation treatments of the proton pump; in fact, incubation of the H¹-ATPase with commercially available alkaline phosphatase and more remarkably with a PP2A partially purified from maize roots, completely abolished the interaction with 14-3-3 proteins. Notably, this treatment was also able to impair the FC-dependent association of 14-3-3 proteins. In fact, FC in vitro addition only partially restored the association of 14-3-3 proteins with the proton pump.

In conclusion, our data, taken together, unequivocally demonstrate that the interaction between 14-3-3 proteins and H¹-ATPase is dependent on the phosphorylation status of the enzyme; moreover, they also indicate that phosphorylation/dephosphorylation events, mediated by a still unidentified protein kinase and possibly a PP2A, very likely represent the basis for the physiological 14-3-3-mediated regulation of H¹-ATPase. In addition, our data raise the question as to how FC promotes the association of 14-3-3 proteins with the H¹-ATPase. In fact, whereas it was proposed that FC can act by a mechanism able to replace the phosphorylation requirements of the H¹-ATPase (13, 14), our data indicate that the phosphorylation status of the H¹-ATPase can influence also the FC-dependent 14-3-3 association with the proton pump and consequently that FC does not merely mimic the phosphorylation effect but rather acts through a more complex mechanism.

REFERENCES

1. Palmgren, M. G. (1998) Adv. Bot. Res. 28, 1–70
2. Michelet, B., and Boutry, M. (1995) Plant Physiol. 106, 1–6
3. Palmgren, M. G., Sommarin, M., Serrano, R., and Larsson, C. (1991) J. Biol. Chem. 266, 20470–20475
4. Regenberg, B., Villalba, J. M., Lanfermeijer, F. C., and Palmgren, M. G. (1995) Plant Cell 7, 1655–1666
5. Rasi-Caldogno, S., Pugliarello, M. C., Olivari, C., and De Michielis, M. I. (1993) Plant Physiol. 103, 391–396
6. Johansson, F., Sommarin, M., and Larsson, C. (1999) Plant Cell 5, 321–327
7. Lanfermeijer, F. C., and Prins, H. B. A. (1994) Plant Physiol. 104, 1277–1285
8. Marra, M., Fullone, M. R., Fogliano, V., Masi, S., Matteli, M., Pen, J., and Aducu, P. (1994) Plant Physiol. 106, 1497–1501
9. Korthout, H. A. A. J., and De Boer, A. H. (1994) Plant Cell 6, 1681–1692
10. Oecking, C., Eckerskorn, C., and Weiler, E. W. (1994) FEBS Lett. 352, 163–166
11. Jahn, T., Fuglsang, A. T., Olsson, A., Bruntrup, I. M., Collinge, D. B., Volkmann, D., Sommarin, M., Palmgren, M. G., and Larsson, C. (1997) Plant Cell 9, 1805–1814
12. Oecking, C., Pietrowski, M., Hagemeyer, J., and Hagemann, K. (1997) Plant J. 12, 441–453
13. Fullone, M. R., Visconti, S., Marra, M. Fogliano, V., and Aducu, P. (1999) J. Biol. Chem. 273, 7688–7702
14. Bauningaard, L., Fuglsang, A. T., Jahn, T., Korthout, H. A. A. J., de Boer, A. H., and Palmgren, M. G. (1999) Plant J. 13, 661–671
15. Muslin, A. J., Tanner, J. W., Allen, P. M., and Shaw, A. S. (1996) Cell 84, 889–897
16. Yaffe, M. B., Rittinger, K., Volinia, S., Caron, P. P., Aitken, A., Leffers, H., Gambin, S. J., Snerdson, S. J., and Canley, L. C. (1997) Cell 91, 961–971
17. Bachmann, M., Huber, J. L., Athwal, G. S., Wu, K., Ferl, R. J., and Huber, S. C. (1996) FEBS Lett. 38, 26–30
18. Moorhead, G., Douglas, P., Morrice, N., Scarabel, M., Aitken, A., and Maxkintosh, C. (1996) Current Biology 6, 1104–1113
19. Torosser, D., Athwal, G. S., and Huber, S. C. (1998) FEBS Lett. 435, 110–114
20. Moorhead, G., Douglas, P., Cotelle, V., Harthill, J., Morrice, N., Meek, S., Deiting, U., Stitz, M., Scarabel, M., Aitken, A., and Maxkintosh, C. (1999) Plant J. 18, 1–12
21. Schaller, G. E., and Sussman, M. R. (1998) Planta 173, 509–518
22. Xing, T., Higgins, V. J., and Blumwald, E. (1996) Plant Cell 8, 555–564
23. Olsson, A., Svennelid, F., Ek, B., Sommarin, M., and Larsson, C. (1998) Plant Physiol. 118, 551–555
24. Ballin, A., Carilli, A., Santurbanto, B., and Tuttobello, L. (1968) Ann. Ist. Super. Sanita 4, 317–332
25. Marra, M., Ballio, A., Fullone, M. R., and Aducu, P. (1992) Physiol Plant. 98, 1029–1034
26. Marra, M., Fogliano, V., Zambardi, A., Fullone, M. R., Nasta, D., and Aducu, P. (1996) FEBS Lett. 382, 293–296
27. Johansson, F., Sommarin, M., and Larsson, C. (1994) Physiol Plant. 92, 389–396
28. Laemmli, U. K. (1970) Nature 268, 680–685
29. Bradford, M. M. (1976) Anal. Biochem. 72, 248–254
30. Serrano, R. (1989) Methods Enzymol. 157, 533–544
31. Smith, R. D., and Walker, J., C. (1996) Annu. Rev. Plant Physiol. Plant Mol. Biol. 47, 101–125
32. Chung, H-J., Sehne, P. C., and Ferl, R. J. (1999) Trends Plant Sci. 4, 367–371
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