

groups and reduction or exchange of disulfide bonds. As a result, we found that approximately half of the alfa' and alfa subunit of beta-conglycinin were disulfide-linked, together or with P34, prior to N-terminal propeptide processing. Sedimentation velocity experiments, size exclusion chromatography, and two-dimensional polyacrylamide gel electrophoresis (PAGE) analysis, with Blue native-PAGE followed by sodium dodecyl sulfate-PAGE, indicated that the beta-conglycinin complexes containing the disulfide-linked alfa'/alfa subunits were complexes of dodecamer. The alfa' or alfa subunits, when disulfide-linked with P34, were mostly present in hexamer. Our results suggest that disulfide bonds are formed between alfa'/alfa subunits residing in different beta-conglycinin hexamers, but the binding of P34 to alfa' and alfa subunits reduces the linkage between beta-conglycinin hexamers. Finally, we found a subset of other major storage protein, glycinin existed as noncovalently associated complexes with beta-conglycinin by two-dimensional PAGE analysis. Our data suggest that the associations of pro alfa' or pro alfa subunits with P34 or glycinin may play an important role in the transport of P34 and glycinin to the PSV via Golgi bodies.

P03-12

Expression profiling of glutathione transferase (*Gst1*) gene in maize seedlings infested by the bird cherry-oat aphid (*Rhopalosiphum padi* L.)

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The purpose of performed analyses was to evaluate the impact of bird cherry-oat aphid (*Rhopalosiphum padi* L.) feeding on the transcriptional activity of glutathione transferase (*Gst1*) gene within maize seedlings. Furthermore, the selected entomological parameters (length of prereproductive period and daily fecundity) were recorded in order to assess the average time of generation development (T) and the intrinsic rate of natural increase (r_m) of the aphid population on *Zea mays* plants.

The bioassays were conducted in a growth chamber at four levels of aphid infestation (5, 10, 20 and 40 individuals per seedling). Transcriptional responses of *Gst1* gene in aphid-infested maize seedlings were investigated at 1, 2, 4, 8, 24 and 48 hour post infestation (hpi). The qRT-PCR technique was used to quantify the relative expression of the targeted gene in maize tissues (GAPDH, glyceraldehyde 3-phosphate dehydrogenase gene was used as a reference control).

Expression profiles of the *Gst1* gene within *R. padi*-stressed maize seedlings showed differential transcript accumulation in dependence on the number of aphid individuals and duration of exposure to the examined biotic stressor. The expression levels of analysed gene in maize tissues were gradually elevated during the early phases of infestation (1, 2 and 4 hpi) when compared to the aphid-free control. Importantly, the most substantial up-regulation of *Gst1* gene in stressed *Z. mays* seedlings was reached at 8 hour of aphid infestation. The presented results evidence the involvement of cytosolic GSTI enzyme in overcoming detrimental imbalance in the redox status as a result of the aphid-triggered oxidative burst in maize seedlings.

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P03-13

The *N. tabacum* *RIP1* gene encodes for a novel pollen tube protein involved in Rac5 GTPase dependent control of polarized cell growth

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Polarization of eukaryotic cells through signaling of Rho-GTPases is essential for morphogenesis, division, and motility of unicellular organisms and plays a key role in differentiation and development of various cell types in metazoa. Polarized cell growth in pollen tubes of *Nicotiana tabacum* is controlled by the Nt-Rac5 GTPase that accumulates in the tube tip and belongs to a plant specific subfamily of Rho-GTPases. In a yeast two hybrid screen for unknown effectors of Nt-Rac5 we identified the unspecified protein Nt-Rip1. Here we functionally characterize the Rip1 protein that is exclusively expressed in growing pollen tubes. Rip1 is a 600aa protein that comprises an N-terminal hydrophobic region predicted as trans membrane domain, as well as a C-terminal domain of unknown function (DUF593). Transient over expression of Rip1 inhibits tobacco pollen tube growth and interferes with membrane trafficking and secretion. Localization studies with YFP-Rip1 constructs in *N. tabacum* pollen tubes reveal that Rip1 resides at the apex of the growing tip, sub-apical of the vesicle-accumulation zone. Our pull down data identify Nt-Rip1 to be a physical interactor of Nt-Rac5. We performed a deletion analysis of the *RIP1* gene and show that interaction between Rac5 and Rip1 is restricted to the N-terminus of Rip1. Interestingly we could provide evidence that Rip1 co-localizes with known trans golgi network (TGN) markers sub-apical in the pollen tube tip near the actin ring. Subsequent binding studies could provide evidence that Rip1 associates with Actin. Analysis of several Rip1 truncations shows, that DUF593 is responsible for the specific localization pattern of Rip1 sub-apical of the vesicle accumulation zone. We conclude that Rip1 associates with post Golgi-organelles and the actin cytoskeleton, hence linking membrane trafficking to polarized cell growth regulated through the Rac5-GTPase.

P03-14

Cyclin D-CDKs during maize seed germination: activity and regulation

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The cell cycle has a determinant role in growing and development of multicellular organisms, including plants. During germination cells restart cell division and activate the cell cycle machinery to generate a new plant. Some of the basic mechanisms that regulate cell cycle is the formation of cyclin-CDK complexes, which determine cell cycle progression. Although mammals and plants share similarities in the regulatory machinery, plants contain specific types of CDKs and cyclins that have no orthologs in mammals. The aim of this work is to study regulatory events of some G1 phase cyclin-CDK complexes during maize germination. We have