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Poster Abstract - F.02

REDOX CONTROL OF GLUTENIN SUBUNIT ASSEMBLY IN A HETEROLOGOUS EXPRESSION SYSTEM

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Gliadins and glutenins are the main storage proteins that accumulate in wheat endosperm during seed development. While gliadins are monomeric, glutenins form large polymers composed of high-molecular-weight and of low-molecular-weight subunits held together by interchain disulphide bonds. The high-molecular weight glutenin subunits have been extensively studied and found to be major determinants of gluten elasticity. Less research attention has been devoted to the low-molecular-weight glutenin subunits (LMW-GS) that still constitute a large fraction of wheat storage proteins and have also been proposed to influence the end-use quality of wheat flours. LMW-GS are generally characterised by the presence of three intrachain disulfide bonds and of two additional cysteine residues that are believed to mediate polymeric assembly. The large number of different LMW-GS and the complexity of the glutenin polymers somehow hamper the direct study of the assembly process as it occurs in wheat endosperm cells. To overcome these problems, we have studied the folding and assembly of a model LMW-GS expressed in tobacco leaf protoplasts. Expression of both wild-type and epitope-tagged forms of a LMW-GS in this system resulted in the formation of dimers and higher-order oligomers. Analysis of mutants in which different cysteines were substituted with serines allowed to assess the role of both intrachain and interchain disulfides in the folding and assembly processes. Kinetic analysis of the polymerization process revealed that newly synthesized monomers are promptly incorporated into oligomeric and polymeric forms. The polymerization pattern established soon after synthesis did not significantly change when analyzed over a several hour period. We also found that the assembly status could be influenced by treatments able to affect the redox of the endoplasmic reticulum. Treatment with a reducing agent was sufficient to reduce glutenin polymers *in vivo*, but polymerization promptly resumed when oxidizing conditions were restored. Conversely, treatment with an oxidizing agent caused an increase in the ratio between polymeric and monomeric forms. A model for the redox-regulated polymerization of LMW-GS is proposed.