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A soybean binding protein (BiP) homolog is temporally regulated in soybean seeds and associates detectably with normal storage proteins *in vitro*

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ABSTRACT

The endoplasmic reticulum (ER) luminal binding protein (BiP) is thought to be a key mediator of folding and assembly of *de novo* synthesized secretory proteins. We have used a maize (*Zea mays* L.) BiP antibody to identify its homolog in soybeans (*Glycine max* (L.) Merril). The accumulation of BiP in developing soybean seeds seems to be coordinated with the onset of active storage protein synthesis. We used a co-immunoprecipitation assay to detect soybean BiP: β-conglycinin interactions. Either a maize BiP antibody or a β-conglycinin antibody co-immunoprecipitated the reciprocal protein from whole seed protein extract enzymatically depleted of adenosine 5'-triphosphate (ATP), while an unrelated antibody failed to immunoprecipitate either one. The association of BiP:β-conglycinin complexes was completely reversed by addition of ATP, a diagnostic feature of molecular chaperone-mediated interaction. However, only a very small fraction of β-conglycinin was found to be associated with BiP in whole cell protein extracts from immature seeds. These results are consistent with a transient association between BiP and β-conglycinin subunits, and suggests its involvement in the biosynthetic transport pathway of storage proteins to protein bodies.

INTRODUCTION

The heat shock proteins (HSP70) are a ubiquitous family of stress-induced proteins, which have been described as polypeptide binding proteins and molecular chaperones (revised in Hendrick and Hartl, 1993). In eukaryote, the HSP70-related proteins belong to a multigene family whose members are targeted to different sub-cellular compartments (for review, see Ellis, 1991). Among members of the HSP70 family, structural differences, related to targeting signals, have been used to assign the conserved members of this family to its sub-cellular localization. The endoplasmic reticulum (ER)-resident HSP70 protein, known as BiP (binding protein), has a functional peptide signal at the amino terminus which directs it synthesis to the ER and a tetrapeptide carboxyl-terminal, K/R/HDEL, which constitutes a general reticuloplasmin retention signal (Munro and Pelham, 1987; Pelham, 1989; Denecke *et al.*, 1992). The retrieval of BiP in the ER is mediated by the interaction with the HDEL/KDEL receptor protein (Pelham, 1988; Semenza *et al.*, 1990; Lee *et al.*, 1993).

As a member of the HSP70 family, the molecular chaperone activity of BiP has been characterized in several systems (for review, see Vitale *et al.*, 1993). BiP

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