Title:	Protein targeting to starch 1, a functional protein of starch biosynthesis in wheat (Triticum aestivum L.)
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Abstract:	In cereal endosperm, native starch comprising amylose and amylopectin is synthesized by the coordinated activities of several pathway enzymes. Amylose in starch influences its physio-chemical properties resulting in several human health benefits. The Granule-Bound Starch Synthase I (GBSSI) is the most abundant starch-associated protein. GBSSI lacks dedicated Carbohydrate-binding module (CBM). Previously, Protein Targeting To Starch 1 (PTST1) was identified as a crucial protein for the localization of GBSSI to the starch granules in Arabidopsis. The function of its homologous protein in the wheat endosperm is not known. In this study, TaPTST1, an AtPTST1 homolog, containing a CBM and a coiled-coil domain was identified in wheat. Protein-coding nucleotide sequence of TaPTST1 from Indian wheat variety ‘C 306’ was cloned and characterized. Homology modelling and molecular docking suggested the potential interaction of TaPTST1 with glucans and GBSSI. The TaPTST1 expression was higher in wheat grain than the other tissues, suggesting a grain-specific function. In vitro binding assays demonstrated different binding affinities of TaPTST1 for native starch, amylose, and amylopectin. Furthermore, the immunoaffinity pull-down assay revealed that TaPTST1 directly interacts with GBSSI, and the interaction is mediated by a coiled-coil domain. The direct protein–protein interaction was further confirmed by bimolecular fluorescence complementation assay (BiFC) in planta. Based on our findings we postulate a functional role for TaPTST1 in starch metabolism by targeting GBSSI to starch granules in wheat endosperm.
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