

Figure S3. SDS-PAGE analysis of ENGase-type activity in *T. atroviride* using (A) culture filtrate or (B) cytosolic fraction. *T. atroviride* WT and Δ *Eng18B* mutants were grown in dextrose broth for 48h at 25°C. Forty μl culture filtrate or cytosolic fraction was mixed with 100 μg of RNase B and incubated at room temperature for 24h for deglycosylation. Twenty μl of the reactions were mixed with 5 μl of loading dye and heat denatured at 100°C for 10 min before loading. L, protein ladder; 1, RNAse B incubated with dextrose broth; 2, fresh RNAse B; 3, WT culture filtrate or cytosolic fraction incubated with RNAse B; 4 and 5, Δ *Eng18B* mutants culture filtrate or cytosolic fraction without RNAse B; 7 and 8, Δ *Eng18B* mutants culture filtrate or cytosolic fractions without RNAse B.