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Phosphomimetically mutated and thus constitutively active kinase of ribosomal protein S6 from *Arabidopsis thaliana* (AtRPS6K2) does phosphorylate TaRPS6 in wheat (*Triticum aestivum*) 40S ribosomal subunit

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Coordination of growth and division in eukaryotic cells depending on availability of nutrients, energy, and in response to internal and external stimuli, is adjusted by multilevel cascade of serine-threonine protein kinases that transmit various signals to the protein-synthesizing apparatus causing its activation or inhibition. Phosphorylation of RPS6 by RPS6-kinase stimulates production of new ribosomes via preferential translation of 5′TOP-mRNAs that encode most proteins of translational apparatus (ribosomal proteins, elongation factors and many of initiation factors, poly(A)-binding proteins, etc.) and proteins of proliferation control. The mechanism of preferential translation of 5′TOP-mRNAs is unknown. Most studies of RPS6-kinase regulation in plants performed on *A. thaliana* that contains AtRPS6K2, which phosphorylates AtRPS6 in 40S ribosomal subunit (40S RS). For full activation, AtRPS6K2 requires phosphorylation by upper-level kinases: pPDK1 (at Ser296) and pTOR (Thr455, Ser437). To investigate the role of RPS6-phosphorylation in preferential translation of some viral and cellular 5′TOP-mRNAs it is important to obtain constitutively active AtRPS6K2. For this purpose we cloned *AtRPS6K2* cDNA-gene and carried out *in vitro*-mutagenesis, replacing codons of Ser(S)296, S437 and Thr(T)455 by triplets that encode phosphomimetic amino acid Glu(E). After expression in *E. coli*, two recombinant proteins were isolated: original AtRPS6K2 and phosphomimetic AtRPS6K2(S296E;S437E;T455E). These kinases were tested *in vitro* for their ability to phosphorylate either purified recombinant AtRPS6 (~30-kDa) or its homolog TaRPS6 in composition of 40S RS isolated from wheat germ (*T. aestivum*). Neither original nor phosphomimetic kinases were able to phosphorylate purified recombinant AtRPS6. Phosphomimetic kinase did phosphorylate TaRPS6 in composition of isolated 40S RS as was evident from SDS-PAG-electrophoresis and subsequent radioautography by incorporation of radioactivity from [γ-^{33}P]ATP into 30-kDa polypeptide. Besides *in vitro* studies, such an approach can find biotechnological applications.