A C-term truncated EIF2Bγ protein encoded by an intronically polyadenylated isoform introduces unfavorable EIF2Bγ–EIF2γ interactions

Eukaryotic translation initiates upon EIF2-GTP·Met-tRNAi ternary complex (TC) recruitment to the ribosomes. EIF2 (α-γ subunits) is a GTPase. Within the EIF2 complex, EIF2γ carries the GTP. Following TC formation, EIF2-GDP is released from the ribosome. EIF2B (α-ε subunits) acts as a guanine exchange factor (GEF) and regulates the GDP to GTP exchange. In the EIF2B complex, EIF2Bε has a catalytic GEF domain. During stress-induced conditions, phosphorylation of EIF2α turns EIF2 into an inhibitor of EIF2B forming an inactive EIF2:EIF2B complex. Within the inactive complex, EIF2γ interacts with EIF2Bγ rather than EIF2Bε. In turn, inactive complex lacks GEF activity, therefore, decreases TC formation and triggers the internal stress response (ISR), which determines the cell fate. Deregulated ISR has been linked to neurodegenerative disorders and cancer. In this work, our experimental collaborators characterized an upregulation of an intronically polyadenylated transcript of EIF2Bγ in breast cancer cells. Based on this finding, we examined the structural impact of this isoform on EIF2-EIF2B interaction. To that end, we generated structural models of EIF2Bγ isoforms bound to their interacting partner EIF2γ. We also performed residue-based interaction analyses in the presence of full-length and truncated EIF2Bγ. Moreover, we dissected the electrostatic potential maps of EIF2γ:EIF2Bγ complexes. As a result, truncated isoform leads to unfavorable electrostatic interactions across the EIF2γ:EIF2Bγ interface (where EIF2γ-K400 and EIF2Bγ-R412 face each other) (Fig 1A). This observation is further confirmed by our electrostatics maps analyses (Fig 1B-C). Based on this, we predict that the truncated EIF2Bγ isoform leads to a potential decrease in the stability of the inactive EIF2:EIF2B complex, which could explain its upregulation in the breast cancer cells. Our work got recently published in Proteins (Circir et al., Proteins, 2021) together with our models located at https://github.com/CSB-KaracaLab/eif2g-eif2bg-ints.

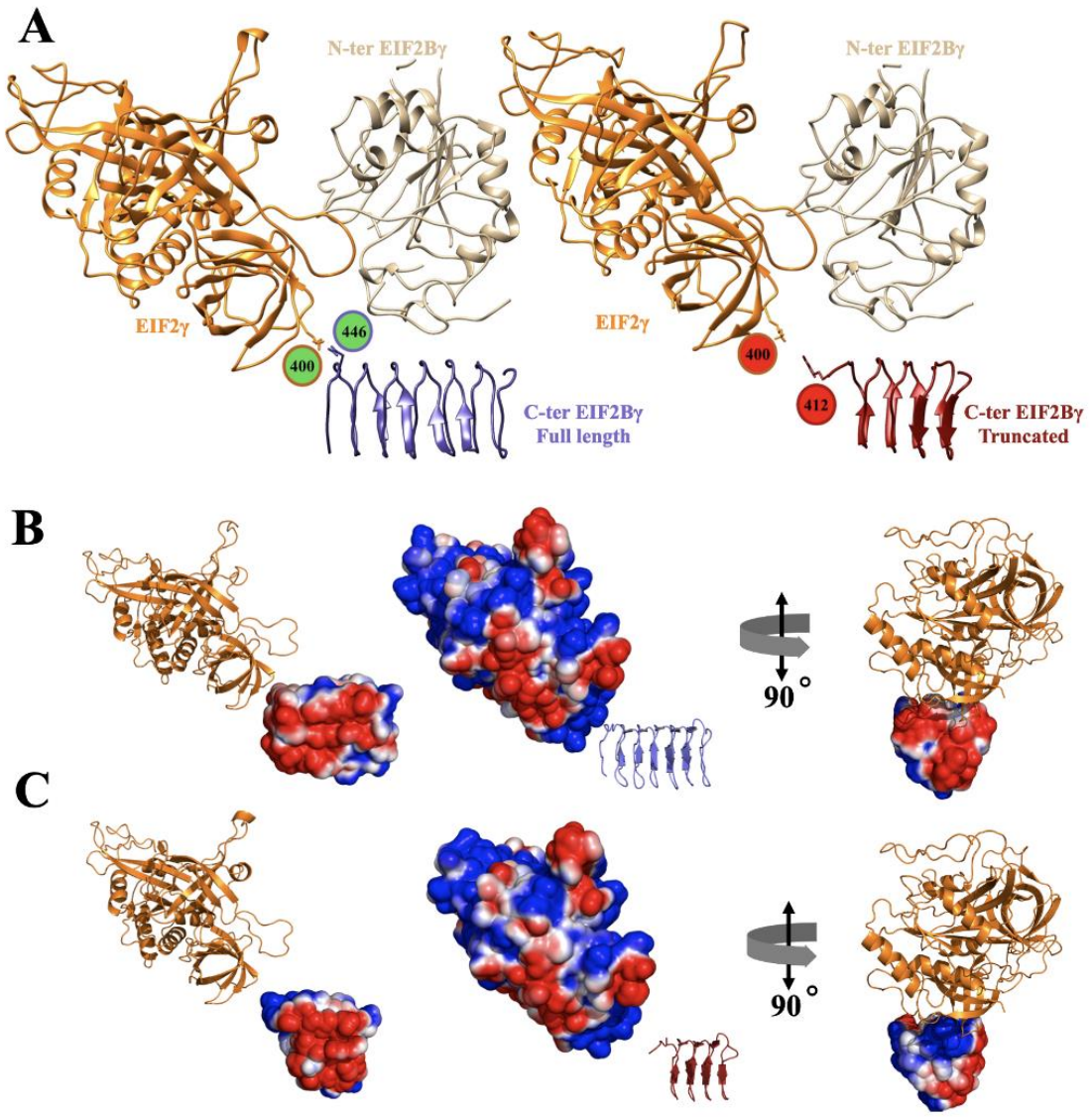

Figure 1 Representation of difference in between isoforms by energetically contributing residues and electrostatic potential maps **A.** Representation of differentially contributing residues of the C-terminus of full length (purple), truncated EIF2Bγ isoform (dark red) and EIF2γ (orange). N terminal of EIF2Bγ in the 6K72 structure is colored with wheat. Residue numbers in circles are emphasized with the corresponding chain colors. Residues that show energetically favorable and unfavorable interactions between EIF2γ:EIF2Bγ complex are colored in green and red, respectively. **B.** Electrostatic potential map of full length EIF2Bγ within the EIF2γ:EIF2Bγ complex is shown, **C.** Electrostatic potential map of truncated EIF2Bγ isoform within the EIF2γ:EIF2Bγ complex is shown. Red, blue and white colors indicate negative, positive and neutral electrostatic potentials with a range of -1 and 1. Poisson-Boltzman electrostatic approach was used and surface charge of proteins were visualized with PyMOL using the APBS 2.1 plugin