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Abstract Submission Form

Poster Title: **Molecular mechanism of EphA2 receptor assembly and the effects on downstream signaling**

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With 14 members, Eph kinases constitute the largest subfamily of receptor tyrosine kinase superfamily. Eph receptor tyrosine kinases and their ephrin ligands regulate cell migration during normal physiology and oncogenesis. Previous research from the Wang lab and others demonstrates that the expression of EphA2 is upregulated in many human cancers, including breast, lung, kidney, and prostate. Interestingly, EphA2 can either function as a tumor suppressor and an oncogenic protein depending on the ligand binding status. The tumor suppressor function is induced by ligand-dependent conical signaling characterized by EphA2 tyrosine phosphorylation and inhibition of Ras/ERK and PI3K/Akt pathways. On the other hand, the oncogenic function of EphA2 is mediated by noncanonical signaling caused by ligand-independent phosphorylation on serine 897 (pS897), which leads to promotion of tumor cell migration and invasion and accentuated cancer stem cell properties. Several cytoplasmic Serine/Threonine kinases including Akt, p90RSK and PKA are responsible for phosphorylating S897. Mechanistically, the Wang lab recently discovered that the unliganded EphA2 receptors are assembled into multimers driven by the ectodomain (*Science*, in press). Three major interfaces between EphA2 dimers were found, two homotypic Head-Head interfaces (LBD-LBD, sushi-sushi) and one Head-Tail heterotypic interface (LBD-FN2). More importantly, Head-Head and Head-Tail interfaces differentially regulate canonical and noncanonical signaling. In this study, we engineered a mouse glioma cell line with double CRISPR knockout of EphA2 and the closely related EphA1, and re-expressed WT and one of the three mutant EphA2 that disrupts all three interfaces (A2LSF-GFP), abolishes head-tail interface alone (A2FN2-GFP), or causes constitutive activation of canonical signaling (A2Q-GFP). Cellular and biochemical assays were performed to evaluate the impact of the mutations. The data reveal how molecular interfaces mediate the formation of the EphA2 multimeric signaling clusters and the potential role of EphA2 assembly in oncogenesis. This discovery may lead to new strategies for drug discovery in cancer treatment.