## Phylogeny

EPHA8 belongs to the Eph receptor tyrosine kinase family—the largest RTK sub-family—and sits in the EphA subgroup that binds GPI-anchored ephrin-A ligands (Choi & Park, 1999; Gaitanos et al., 2015). Hierarchical clustering based on substrate preferences groups it with EPHA1-7 and EPHB1-4 (Yaron-Barir et al., 2024). Clear one-to-one orthologs are reported in mouse, rat, chicken and zebrafish, underscoring conservation across vertebrates (Choi & Park, 1999; Lucero et al., 2020).

## Reaction Catalyzed

ATP + protein-L-tyrosine ⇌ ADP + protein-L-tyrosine-phosphate (Choi & Park, 1999; Gaitanos et al., 2015; Lucero et al., 2020; Yaron-Barir et al., 2024).

## Cofactor Requirements

Requires Mg²⁺ for catalysis (Gaitanos et al., 2015; Lucero et al., 2020).

## Substrate Specificity

Three independent studies report related but non-identical motif preferences. Collectively, EPHA8 favors:  
• hydrophobic or positively charged residues at –3  
• acidic or basic residues at –2 (source-dependent)  
• small polar residues at –1  
• bulky hydrophobic or aromatic residues at +1  
• acidic or positively charged residues at +2  
• polar or small residues at +3 (Gaitanos et al., 2015; Lucero et al., 2020; Yaron-Barir et al., 2024).

## Structure

Domain architecture: extracellular ligand-binding domain, cysteine-rich segment, two fibronectin type III repeats, a single transmembrane helix, and an intracellular region containing a juxtamembrane segment, kinase domain, SAM domain and PDZ-binding motif (Gaitanos et al., 2015; Lucero et al., 2020). The AlphaFold model (AF-P29322-F1) depicts the typical bilobal kinase fold with conserved C-helix, activation loop and hydrophobic spine stabilising the active conformation (Choi & Park, 1999; Gaitanos et al., 2015; Yaron-Barir et al., 2024).

## Regulation

• Ligand-induced clustering triggers trans-autophosphorylation (Gaitanos et al., 2015; Choi & Park, 1999).  
• Critical autophosphorylation sites: Tyr-615 (juxtamembrane) and Tyr-838 (activation loop); phosphorylation of Tyr-838 enhances kinase activity and promotes Tyr-615 phosphorylation (Choi & Park, 1999).  
• Phospho-Tyr-615 recruits the Src-family kinase Fyn via its SH2 domain (Choi & Park, 1999).  
• Feedback loop: EPHA8 phosphorylates and activates LMW-PTP, which in turn dephosphorylates EPHA8 (Park, 2003).  
• Kinase-independent signaling modulates integrin activity through p110γ PI3K (Gu & Park, 2001).  
• Additional post-translational modifications include N-glycosylation and ubiquitination (Gaitanos et al., 2015).

## Function

• Expression: CNS-restricted during embryogenesis (E10.5–E17.5) in tectum midline, hindbrain and dorsal spinal cord; absent postnatally (Gaitanos et al., 2015).  
• Upstream ligands: ephrin-A2/-A3/-A5 (Gaitanos et al., 2015; Shin et al., 2007).  
• Downstream partners: Fyn, AIDA-1b, Odin, Rho GTPases (RhoA, Rac1, Cdc42) and MAPK cascade components (Choi & Park, 1999; Shin et al., 2007; Gaitanos et al., 2015).  
• Biological roles: axon guidance, regulation of cell adhesion and migration, and neurite outgrowth/retraction; also modulates integrin signaling independently of kinase activity (Choi & Park, 1999; Gu & Park, 2001; Gaitanos et al., 2015).

## Other Comments

EPHA8 knockout mice display commissural and axonal path-finding defects (Choi & Park, 1999; Gu & Park, 2001). Dysregulated expression is observed in certain cancers; e.g., detected in stem-like breast carcinoma cells (Lucero et al., 2020).

## 9. References

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