## Phylogeny

MAPK10 (c-Jun N-terminal kinase 3; JNK3) is a stress-activated mitogen-activated protein kinase that descended from the ancestral serine/threonine kinase repertoire present in the last eukaryotic common ancestor. Subsequent gene duplications produced the three mammalian JNK paralogues; while JNK1 and JNK2 are ubiquitously expressed, JNK3 expression is largely confined to neurons and, to a lesser extent, heart and testes (Kyriakis & Avruch, 2012; Ansideri, 2018). Sequence conservation is high within the catalytic core, whereas peripheral regions have diverged to mediate isoform-specific protein interactions.

## Reaction Catalyzed

ATP + protein-L-Ser/Thr ⇌ ADP + protein-L-Ser/Thr-phosphate + H⁺  
(Bardwell, 2006)

## Cofactor Requirements

Mg²⁺ is essential for ATP coordination and phosphotransfer (Bardwell, 2006).

## Substrate Specificity

JNK3 phosphorylates serine/threonine residues followed by proline (S/T-P) but achieves high selectivity through short basic-hydrophobic “D-site” docking motifs on its substrates (Whisenant et al., 2010; Gordon et al., 2013). Canonical neuronal targets include the transcription factors c-Jun and ATF2 as well as STMN2, amyloid-β precursor protein (APP), and additional proteins enriched in the central nervous system (Ansideri, 2018; Kyriakis & Avruch, 2012).

## Structure

The kinase adopts the conserved bilobed MAPK fold: a β-sheet-rich N-lobe with a glycine-rich loop and αC-helix abuts an α-helical C-lobe. Activation depends on dual phosphorylation of Thr183 and Tyr185 within the TxY motif of the activation loop by MAP2K4/7 (Lu et al., 2023; Messoussi et al., 2016). High-resolution crystal structures and AlphaFold models reveal substrate-binding grooves on the ordered catalytic core, whereas the C-terminal tail contains intrinsically disordered regions that provide flexible interfaces for scaffolds and substrates (Giới, 2022; Whisenant et al., 2010).

## Regulation

• Activation: dual phosphorylation of the TxY motif by MAP2K4 and MAP2K7.  
• Docking control: substrate D-sites increase local concentration and catalytic efficiency (Gordon et al., 2013).  
• Scaffolds: neuronal JIP1 and β-arrestin2 organize JNK3 signalling complexes and direct localisation (Musi et al., 2022).  
• Inhibition: protein–protein interaction with MEN1 can reduce phosphorylation of specific substrates such as JUND (Kyriakis & Avruch, 2012).

## Function

Predominantly neuronal, JNK3 links extracellular stress (e.g., pro-inflammatory cytokines, physical insults) to transcriptional and structural responses. Phosphorylation of c-Jun, ATF2 and other AP-1 components modulates gene programmes governing neuronal differentiation, migration and apoptosis (Kyriakis & Avruch, 2012; Musi et al., 2022). Additional substrates extend its influence to neurite outgrowth (STMN2), APP-mediated signalling during differentiation, and circadian regulation via CLOCK–BMAL1 (Ansideri, 2018; Lu et al., 2023).

## Inhibitors

Structure-guided campaigns have produced ATP-competitive and covalent inhibitors that preferentially inhibit JNK2/3 over JNK1 by exploiting subtle active-site differences (Lu et al., 2023; Messoussi et al., 2016). Virtual-screening studies propose additional chemotypes targeting promiscuous binding pockets (Sailapathi et al., 2020). Disruption of scaffold interactions (e.g., JIP1 or β-arrestin2 binding surfaces) is also being explored as an alternative inhibitory strategy (Ansideri, 2018).

## Other Comments

JNK3 is under active investigation as a therapeutic target in neurodegenerative disorders, where aberrant stress-induced apoptosis contributes to neuronal loss. Computational tools such as the D-finder algorithm continue to expand the catalogue of JNK3 substrates and docking motifs, enriching our understanding of its signalling network (Whisenant et al., 2010; Gordon et al., 2013).

## 9. References

Ansideri, F. (2018). Multiple strategies targeting c-Jun N-terminal kinases: synthesis of novel inhibitors and development of a new binding assay methodology. [Journal unknown].

Bardwell, L. (2006). Mechanisms of MAPK signalling specificity. Biochemical Society Transactions, 34, 837–841. https://doi.org/10.1042/BST0340837

Giới, L. (2022). Intrinsic disorder in MAPK10/JNK3 and its role in protein interactions. [Journal unknown].

Gordon, E. A., Whisenant, T. C., Zeller, M., Kaake, R. M., Gordon, W. M., Krotee, P., et al. (2013). Combining docking site and phosphosite predictions to find new substrates: Identification of Smoothelin-like-2 as a c-Jun N-terminal kinase substrate. Cellular Signalling, 25, 2518–2529. https://doi.org/10.1016/j.cellsig.2013.08.004

Kyriakis, J. M., & Avruch, J. (2012). Mammalian MAPK signal transduction pathways activated by stress and inflammation: A 10-year update. Physiological Reviews, 92, 689–737. https://doi.org/10.1152/physrev.00028.2011

Lu, W., Liu, Y., Gao, Y., Geng, Q., Gurbani, D., Li, L., et al. (2023). Development of a covalent inhibitor of c-Jun N-terminal protein kinase 2/3 with selectivity over JNK1. Journal of Medicinal Chemistry, 66, 3356–3371. https://doi.org/10.1021/acs.jmedchem.2c01834

Messoussi, A., Chevé, G., Bougrin, K., & Yasri, A. (2016). Insight into the selective inhibition of JNK family members through structure-based drug design. MedChemComm, 7, 686–692. https://doi.org/10.1039/C5MD00562K

Musi, C. A., Marchini, G., Giani, A., Tomaselli, G., Priori, E. C., Colnaghi, L., & Borsello, T. (2022). Colocalization and interaction study of neuronal JNK3, JIP1, and β-arrestin2 together with PSD95. International Journal of Molecular Sciences, 23, 4113. https://doi.org/10.3390/ijms23084113

Sailapathi, A., Murugan, G., Somarathinam, K., Gunalan, S., Jagadeesan, R., Yoosuf, N., et al. (2020). Proposing the promiscuous protein structures in JNK1 and JNK3 for virtual screening in pursuit of potential leads. ACS Omega, 5, 3969–3978. https://doi.org/10.1021/acsomega.9b03458

Whisenant, T. C., Ho, D. T., Benz, R. W., Rogers, J. S., Kaake, R. M., Gordon, E. A., et al. (2010). Computational prediction and experimental verification of new MAP kinase docking sites and substrates including Gli transcription factors. PLoS Computational Biology, 6, e1000908. https://doi.org/10.1371/journal.pcbi.1000908