## Phylogeny

MAP3K8 (human) clusters within the STE group, MAP3K family, MAP3K8/Tpl2 sub-family (Manning et al., 2002). Clear one-to-one orthologues exist in mouse (Map3k8), rat (Tpl2) and invertebrate Drosophila (MAP3K “slpr”); zebrafish carries several paralogues (Guan et al., 2023; Manning et al., 2002). A unique evolutionary hallmark is the substitution of Pro145 for the otherwise invariant Gly in the Gly-rich loop—an alteration not seen in any other human kinase (Gantke et al., 2011).

## Reaction Catalyzed

ATP + [MAP2K]-Ser/Thr-OH ⇌ ADP + [MAP2K]-Ser/Thr-O-PO₃²⁻ (Gantke et al., 2011).

## Cofactor Requirements

Catalysis requires divalent cations. In vitro activity is supported by Mg²⁺ (5–10 mM) and Mn²⁺ (≈2 mM) (Chan & Reed, 2005; Parikh et al., 2009).

## Substrate Specificity

Confirmed protein substrates include MEK1/2, MKK4, MEK5 and MKK6, whereas MKK7 is not phosphorylated (Chiu et al., 2024; Gantke et al., 2011). Peptide-array profiling revealed a narrow preference: phenylalanine at positions −3/−2/−1 and +1, and lysine residues flanking the phospho-acceptor site; most peptides remain unmodified (Parikh et al., 2009). A universally accepted linear consensus motif is therefore still undefined (Parikh et al., 2009).

## Structure

The protein comprises an N-terminal regulatory segment containing a nuclear-export signal, a central bilobed catalytic domain and a C-terminal inhibitory degron (residues 435–457) (Gantke et al., 2011; Collins et al., 2018). A 2.6 Å crystal structure of the catalytic core (PDB: COT/Tpl2) shows an atypical fold with a flexible P-loop insert (Gutmann et al., 2015). Canonical motifs (Lys-Glu ion pair, HRD catalytic triad, DFG Mg²⁺-binding motif) are present, but the Gly-rich loop uniquely harbours Pro145 (Bayliss et al., 2015; Gantke et al., 2011). Thr290 within the activation segment undergoes autophosphorylation; Ser400 phosphorylation generates a 14-3-3 docking site (Gantke et al., 2011). Hydrophobic regulatory and catalytic spines align in the active state, while the C-terminal tail can fold back to suppress activity until proteolytic removal of the degron (Bayliss et al., 2015). Proper folding in vivo depends on a stoichiometric complex with NF-κB1 p105 and ABIN-2 (Webb et al., 2019).

## Regulation

• Basal inhibition: MAP3K8 is sequestered in a p105–ABIN-2 ternary complex (Gantke et al., 2011).  
• Activation: IKKβ-mediated phosphorylation of p105 triggers its K48-linked ubiquitination and proteasomal degradation, releasing MAP3K8 (Collins et al., 2018).  
• Autophosphorylation/trans-phosphorylation: Thr290 (auto) and Ser400 (auto or trans) are required for full catalytic output and 14-3-3 binding (Gantke et al., 2011).  
• Nuclear turnover: Following stimulus-induced nuclear entry, MAP3K8 is poly-ubiquitinated and degraded; BCL-3 accelerates this process (Collins et al., 2018).  
• Metabolic modulation: Extracellular L-arginine enhances Thr290/Ser400 phosphorylation and signalling amplitude (Gantke et al., 2011).  
• C-terminal degron: residues 435–457 target the kinase for proteasomal destruction; truncations yield constitutive activity (Gantke et al., 2011).

## Function

Expression is high in macrophages and other myeloid cells and can be induced in airway epithelium and adipocytes (Chiu et al., 2024; Gantke et al., 2011). Upstream activators include TLR2/4/9 ligands, IL-1β, TNF, CD40 and TRAF6 via the MyD88–IKK axis (Chiu et al., 2024; Gantke et al., 2011). The dominant downstream pathway is MEK1/2 → ERK1/2, with context-dependent signalling to JNK (via MKK4) and p38 (via MKK3/6) (Chiu et al., 2024; Gantke et al., 2011). MAP3K8 is indispensable for LPS-induced TNF and modulates multiple cytokines/chemokines (IL-1β, IL-6, IL-8, IL-10, IL-12, CCL2, CXCL8) (Gantke et al., 2011; Chiu et al., 2024). Additional roles encompass ERK-dependent lipolysis in adipocytes (Gantke et al., 2011), regulation of macrophage M2 lipid metabolism and restraint of Schistosoma-induced Th2 fibrosis (Kannan et al., 2016), and antiviral defence; Foot-and-mouth disease virus VP1 blocks Thr290 phosphorylation to dampen IFN-β induction (Guan et al., 2023).

## Inhibitors

• C34 – ATP-competitive probe that suppresses MAP3K8-dependent cytokine production (Kannan et al., 2016).  
• Quinoline-3-carbonitrile series; analogue IIIa is a potent biochemical inhibitor (Hu, 2007).  
• 8-Substituted-4-anilino-6-aminoquinoline-3-carbonitriles show selective Tpl2 inhibition with in-vivo anti-inflammatory efficacy (Combination Therapies Targeting ERK1/2 and HDAC6, 2023).  
(IC₅₀ values not reported in cited excerpts.)

## Other Comments

C-terminal truncations that delete the degron stabilize the protein and confer oncogenic potential in lymphoid models (Gantke et al., 2011). Hyper-active MAP3K8 has been linked to rheumatoid arthritis, Crohn’s disease, colitis-associated cancer, melanoma, breast cancer and other inflammatory bowel diseases (Gantke et al., 2011; Webb et al., 2019).

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