## Phylogeny

• Single-copy orthologs occur from bacteria (Escherichia coli YihX) through plants (Arabidopsis thaliana AtFN3K) to lower eukaryotes such as Danio rerio and Mus musculus, underscoring deep conservation of the FN3K family (shrestha2020aredoxactiveswitch pages 1-1).  
• Two independent gene-duplication events in tetrapods produced the human paralogs FN3K and FN3K-related protein (FN3KRP); fish and urochordates retain only an FN3KRP-like locus (delplanque2004tissuedistributionand pages 1-1).  
• Human FN3K and FN3KRP share ≈65 % sequence identity and are clustered on chromosome 17q25.3 (avemaria2015possibleroleof pages 1-2, collard2003amammalianprotein pages 2-4).  
• Sequence/structural comparisons place FN3K within the protein-kinase-like (PKL) clade of the human kinome, most closely related to small-molecule aminoglycoside phosphotransferases rather than canonical eukaryotic Ser/Thr or Tyr kinases (unknownauthors2023elucidatingtheunderstudied pages 61-65, payne2008mappingofthe pages 1-2).

## Reaction Catalyzed

ATP + protein-bound Nε-fructosyl-L-lysine ⇌ ADP + protein-bound Nε-fructosyl-L-lysine-3-phosphate (delpierre2003fructosamine3kinasean pages 1-2).

## Cofactor Requirements

• Catalytic activity requires Mg²⁺; Mg²⁺ chelation abolishes phosphate transfer (delplanque2004tissuedistributionand pages 3-4).  
• In vitro assays confirm ATP-Mg²⁺ as the obligatory nucleotide/cofactor pair (payne2008mappingofthe pages 1-2).

## Substrate Specificity

• Highest turnover is observed on protein-bound fructosamine adducts (fructoselysine); unmodified proteins are not phosphorylated (szwergold2001humanfructosamine3kinasepurification pages 2-4).  
• FN3K also accepts psicosamine- and ribulosamine-modified proteins, albeit with lower efficiency (collard2003amammalianprotein pages 2-4, delplanque2004tissuedistributionand pages 1-1).  
• The small-molecule mimic 1-deoxy-1-morpholino-D-fructose (DMF) is an efficient competitive substrate (delpierre2003fructosamine3kinasean pages 1-2).  
• Substrate recognition is governed by the ketosamine moiety; no linear peptide consensus motif has been defined (delpierre2003fructosamine3kinasean pages 1-2).

## Structure

• Crystal structures of plant FN3K (PDB 6O0V/6O0W) reveal a canonical bilobal PKL kinase fold with conserved VAIK, HGD and DFG catalytic motifs (unknownauthors2023elucidatingtheunderstudied pages 27-32).  
• A conserved P-loop cysteine (Cys24 in human FN3K) forms an inter-subunit disulfide, creating a strand-exchange dimer that functions as a redox switch (shrestha2020aredoxactiveswitch pages 1-1).  
• Human FN3K crystallizes as a domain-swapped dimer; key catalytic residues include Lys41, Glu55, Asp217 and Asp234 positioned for ATP and sugar coordination (garg2025themolecularbasis pages 2-3, garg2025themolecularbasis pages 8-10, payne2008mappingofthe pages 2-3).  
• FN3K lacks the extended activation segment typical of eukaryotic protein kinases, consistent with its specialization for small-molecule substrates (unknownauthors2023elucidatingtheunderstudied pages 27-32).

## Regulation

• Redox control: oxidation of Cys24 triggers disulfide-linked dimerization; reduction reverses the process, modulating activity (shrestha2020aredoxactiveswitch pages 1-1, unknownauthors2023elucidatingtheunderstudied pages 45-51).  
• Dimerization enhances catalytic efficiency (~60 % increase compared with monomer) (garg2025themolecularbasis pages 1-2).  
• NADH binds the ATP pocket and stabilizes the protein thermally while inhibiting kinase activity in a concentration-dependent manner (kannan2024illuminatingthefunctions pages 9-11).  
• No experimentally verified phosphorylation, ubiquitination or other covalent PTMs have been reported; PhosphoSitePlus currently lists sites of unknown functional relevance (kannan2024illuminatingthefunctions pages 28-34).

## Function

• Catalyzes intracellular protein deglycation, thereby repairing early glycation damage and preventing accumulation of advanced glycation end products (delpierre2003fructosamine3kinasean pages 1-2).  
• Highly expressed in erythrocytes, brain, heart, kidney, skeletal muscle and lens; lower activity in lung, spleen and thymus (delplanque2004tissuedistributionand pages 3-4).  
• Localizes to cytoplasm, mitochondria and nucleus, consistent with a broad protein-repair role (kannan2024illuminatingthefunctions pages 9-11).  
• Deglycates the transcription factor NRF2, preserving oxidative-stress responses (beeraka2021thetamingof pages 12-14).  
• Interactome analyses identify partners in glycolysis (LDHA) and lipid metabolism (FASN), linking FN3K to metabolic pathway regulation (kannan2024illuminatingthefunctions pages 6-9).

## Inhibitors

• 1-Deoxy-1-morpholino-D-fructose (DMF) competes with protein substrates and reduces activity in vitro (delpierre2003fructosamine3kinasean pages 1-2).  
• NADH acts as a micromolar-range inhibitor via ATP-site binding (kannan2024illuminatingthefunctions pages 9-11).  
• Dimethyl fumarate has been reported to suppress FN3K activity in cell-free assays (beeraka2021thetamingof pages 14-15).

## Other Comments

• Reduced FN3K activity or loss-of-function polymorphisms elevate protein glycation and associate with diabetic complications, including retinopathy and neuropathy (avemaria2015possibleroleof pages 1-2, shrestha2020aredoxactiveswitch pages 1-1).  
• FN3K expression is altered in hepatocellular and colorectal carcinomas, implicating the enzyme in cancer metabolism via the NRF2 axis (beeraka2021thetamingof pages 12-14).

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