

GSK-3

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Overview

GSK-3 is a highly conserved family of protein kinases. In humans, two genes encode two distinct, but closely related GSK-3 forms, referred to as GSK-3 α and GSK-3 β . They display 84% overall identity and 98% identity within their catalytic domain, the main difference between the two coming from an extra Gly-rich stretch in the N terminal domain of GSK-3 α . Yet they are not functionally interchangeable as demonstrated by the embryonic lethal phenotype of GSK-3 β knock-outs. GSK-3 β 2 is an alternate splicing variant of GSK-3 β expressed in brain.

GSK-3 β has recently been crystallized with various ligands. Its overall shape is shared by all kinases, and comprises a small, N-terminal lobe consisting mostly of β sheets and a large, C-terminal lobe, essentially formed of α -helices. The ATP-binding pocket is located between the two lobes. Arg⁹⁶, Arg¹⁸⁰ and Lys²⁰⁵ form a small pocket where the phosphate group of the primed substrate and the pseudo-substrate binds.

GSK-3 α/β is regulated by phosphorylation on Ser^{21/9} (inhibitory) and on Tyr^{279/216} (activating). The phospho-Ser^{21/9} N-terminal domain of GSK-3 acts as a pseudo-substrate blocking access of substrates to the catalytic site. The T-loop domain containing unphosphorylated Tyr^{279/216} is thought to prevent access of substrates to the catalytic site, and Tyr phosphorylation to release this inhibition. GSK-3 is also regulated by interactions with other proteins. Axin and presenilin act as docking proteins that allow the substrate to interact with the priming kinase. The substrate indeed requires a priming phosphorylation by another kinase on a residue located C-terminal to the GSK-3 phosphorylation site. The substrate recognition site of GSK-3 is -S-X-X-X-Sp,

where Sp is the priming pre-phosphorylated Ser. GSK-3 β is a predominantly cytoplasmic kinase, but it is also present in the nucleus and in mitochondria. FRAT-1 promotes the nuclear efflux of GSK-3 β . LANA, a Kaposi virus protein, sequesters GSK-3 β in the nucleus.

GSK-3 plays a major function in Wnt signaling pathways. When the Wnt pathway is stimulated, GSK-3 is inactivated, β -catenin builds up and accumulates in the nucleus where it forms, with TCF/LEF, a transcription factor regulating a large variety of genes. GSK-3 phosphorylates the cell cycle regulators β -catenin, cyclin D1, cyclin E, p21^{CIP1} and c-Myc, leading to their ubiquitin-dependent destruction. In the absence of insulin, active GSK-3 phosphorylates and inactivates glycogen synthase and eIF2B. Binding of insulin to its plasma membrane receptor leads to PKB/AKT activation, resulting in phosphorylation and inactivation of GSK-3. Consequently, glycogen synthase and eIF2B are activated, and glycogen and protein synthesis are stimulated. GSK-3 plays a pro-apoptotic function in neuronal cells. In contrast, GSK-3 is necessary also for cell survival as demonstrated by the massive TNF α -induced hepatocyte apoptosis which leads to death in early embryonic GSK-3 β knockout mice. GSK-3 activity/inactivity or spatial distribution play an essential role during development (polarity determination). Finally, GSK-3 and CK1 regulate the circadian clock in *Drosophila* and in mammals.

Five sets of data have stimulated the search for pharmacological inhibitors of GSK-3: [1] the mood stabilizing properties of lithium, the first GSK-3 inhibitor to be described, [2] the insulin-mimetic properties of GSK-3 inhibitors, [3] the interaction of GSK-3

with presenilin-1, the GSK-3-dependent amyloid- β production and abnormal tau phosphorylation in Alzheimer's disease, [4] the involvement of GSK-3 in neuronal cell death and the neuroprotection provided by GSK-3 inhibitors following various insults, [5] the maintenance of pluripotency of embryonic stem cells in the absence of feeder cells by GSK-3 inhibitors. Over 30 GSK-3 inhibitors have been identified, among which seven have been co-crystallized with GSK-3 β , all of which localize within the ATP-binding pocket of the enzyme. GSK-3 inhibitors are thus evaluated on Alzheimer's disease and other neurodegenerative diseases, bipolar affective disorders, diabetes and diseases caused by unicellular parasites

GSK-3

FAMILY MEMBERS	GSK-3 α	GSK-3 β	GSK-3 β 2
OTHER NAMES	Glycogen synthase kinase 3 α	Glycogen synthase kinase 3 β	Glycogen synthase kinase 3 β 2
MOLECULAR WEIGHT/ STRUCTURAL DATA	51 kDa 483 aa (human)	47 kDa 420 aa (human)	48 kDa 433 aa (human)
ISOFORMS	Not known	Not known	Splice variant of GSK-3 β
SPECIES	Present in all species	Present in all species	Not known
DOMAIN ORGANIZATION	Not known	Not known	Not known
PHOSPHORYLATION SITES	Ser ² , Tyr ²⁷⁹	Ser ⁹ , Tyr ²¹⁶	Not known
TISSUE DISTRIBUTION	Ubiquitous	Ubiquitous	Brain
SUBCELLULAR LOCALIZATION	Cytoplasm, nucleus, mitochondria	Cytoplasm, nucleus, mitochondria	Cytoplasm, nucleus, mitochondria
BINDING PARTNERS/ ASSOCIATED PROTEINS	Axin, presenilin, β -catenin, Akt, p53, APC, Tau, Notch2, glycogen synthase	Axin, presenilin, β -catenin, Akt, p53, APC, Tau, Notch2, glycogen synthase	Axin, presenilin, β -catenin, Akt, p53, APC, Tau, Notch2, glycogen synthase
UPSTREAM ACTIVATORS	CK2 (for glycogen synthase), CK1 α (β -catenin), DYRK1A (eIF2B), PKA (<i>Cubitus interruptus</i>), CDK5/p25 (tau), MARK (tau) ?	CK2 (for glycogen synthase), CK1 α (β -catenin), DYRK1A (eIF2B), PKA (<i>Cubitus interruptus</i>), CDK5/p25 (tau), MARK (tau) ?	CK2 (for glycogen synthase), CK1 α (β -catenin), DYRK1A (eIF2B), PKA (<i>Cubitus interruptus</i>), CDK5/p25 (tau), MARK (tau) ?
DOWNSTREAM ACTIVATION	p70 ^{S6K} , p90 ^{rsk} , PKA, Akt, PKC isoforms, ILK, fyn kinase, PYK2, ZAK1, MEK1, GSK-3, MAP-IB, MAP-2, eIF2B, <i>Cubitus interruptus</i>	p70 ^{S6K} , p90 ^{rsk} , PKA, Akt, PKC isoforms, ILK, fyn kinase, PYK2, ZAK1, MEK1, GSK-3, MAP-IB, MAP-2, eIF2B, <i>Cubitus interruptus</i>	p70 ^{S6K} , p90 ^{rsk} , PKA, Akt, PKC isoforms, ILK, fyn kinase, PYK2, ZAK1, MEK1, GSK-3, MAP-IB, MAP-2, eIF2B, <i>Cubitus interruptus</i>
ACTIVATORS	Not known	Not known	Not known
INHIBITORS	SB 216763 (S3442), SB 415286 (S3567), FRAT1/FRAT2 (GBP), Lithium (L0505), kenpaullone (K3888), Alsterpaullone (A4847), indirubin-3'-oxime (I0404), 6-bromo-indirubin-3'-oxime (BIO) (B1686), hymenialdisine, aloisine, TDZD-8 (T8325), Ro 31-8220 (bisindolylmaleimide IX) (R136), FRATtide (FRAT1 ₁₈₈₋₂₂₆ peptide), GID ₃₂₀₋₄₂₉ and GID ₃₈₀₋₄₀₄ peptides	SB 216763 (S3442), SB 415286 (S3567), FRAT1/FRAT2 (GBP), lithium (L0505), kenpaullone (K3888), alsterpaullone (A4847), indirubin-3'-oxime (I0404), 6-bromo-indirubin-3'-oxime (BIO) (B1686), hymenialdisine, aloisine, TDZD-8 (T8325), Ro 31-8220 (bisindolylmaleimide IX) (R136), FRATtide (FRAT1 ₁₈₈₋₂₂₆ peptide), GID ₃₂₀₋₄₂₉ and GID ₃₈₀₋₄₀₄ peptides	SB 216763 (S3442), SB 415286 (S3567), FRAT1/FRAT2 (GBP), lithium (L0505), kenpaullone (K3888), alsterpaullone (A4847), indirubin-3'-oxime (I0404), 6-bromo-indirubin-3'-oxime (BIO) (B1686), hymenialdisine, aloisine, TDZD-8 (T8325), Ro 31-8220 (bisindolylmaleimide IX) (R136), FRATtide (FRAT1 ₁₈₈₋₂₂₆ peptide), GID ₃₂₀₋₄₂₉ and GID ₃₈₀₋₄₀₄ peptides

FOOTNOTES

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SELECTIVE ACTIVATORS	Not known	Not known	Not known
PHYSIOLOGICAL FUNCTION	Wnt and Hedgehog signaling pathways, transcription, cell division cycle regulation response to DNA damage, cell death and cell survival, cardiovascular and neuronal functions, insulin transduction development, differentiation, regulation of circadian rhythm, stem cell pluripotency maintenance and differentiation	Wnt and Hedgehog signaling pathways, transcription, cell division cycle regulation response to DNA damage, cell death and cell survival, cardiovascular and neuronal functions, insulin transduction development, differentiation, regulation of circadian rhythm, stem cell pluripotency maintenance and differentiation	Wnt and Hedgehog signaling pathways, transcription, cell division cycle regulation response to DNA damage, cell death and cell survival, cardiovascular and neuronal functions, insulin transduction development, differentiation, regulation of circadian rhythm, stem cell pluripotency maintenance and differentiation
DISEASE RELEVANCE	Bipolar disorder, Alzheimer's disease, stroke, Parkinson's disease, Huntington's disease, transmissible spongiform encephalopathies, schizophrenia, circadian clock-dependent diseases, Type 2 diabetes, prostate cancer	Bipolar disorder, Alzheimer's disease, stroke, Parkinson's disease, Huntington's disease, transmissible spongiform encephalopathies, schizophrenia, circadian clock-dependent diseases, Type 2 diabetes, prostate cancer	Bipolar disorder, Alzheimer's disease, stroke, Parkinson's disease, Huntington's disease, transmissible spongiform encephalopathies, schizophrenia, circadian clock-dependent diseases, Type 2 diabetes, prostate cancer

Abbreviations

SB 216763: 3-(2,4-Dichlorophenyl)-4-(1-methyl-1H-indol-3-yl)-1H-pyrrole-2,5-dione

SB 415286: 3-[(3-Chloro-4-hydroxyphenyl)amino]-4-(2-nitrophenyl)-1H-pyrrol-2,5-dione

FOOTNOTES