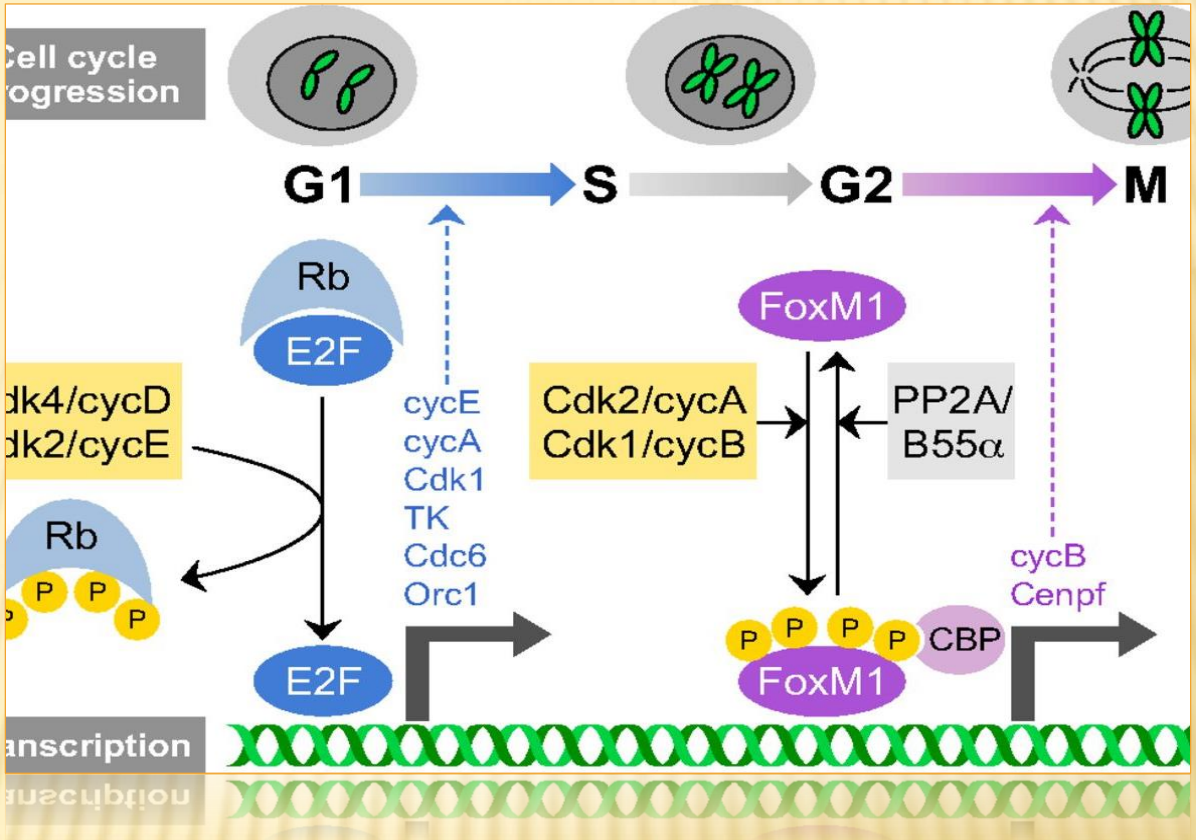
c-SRC KINASE

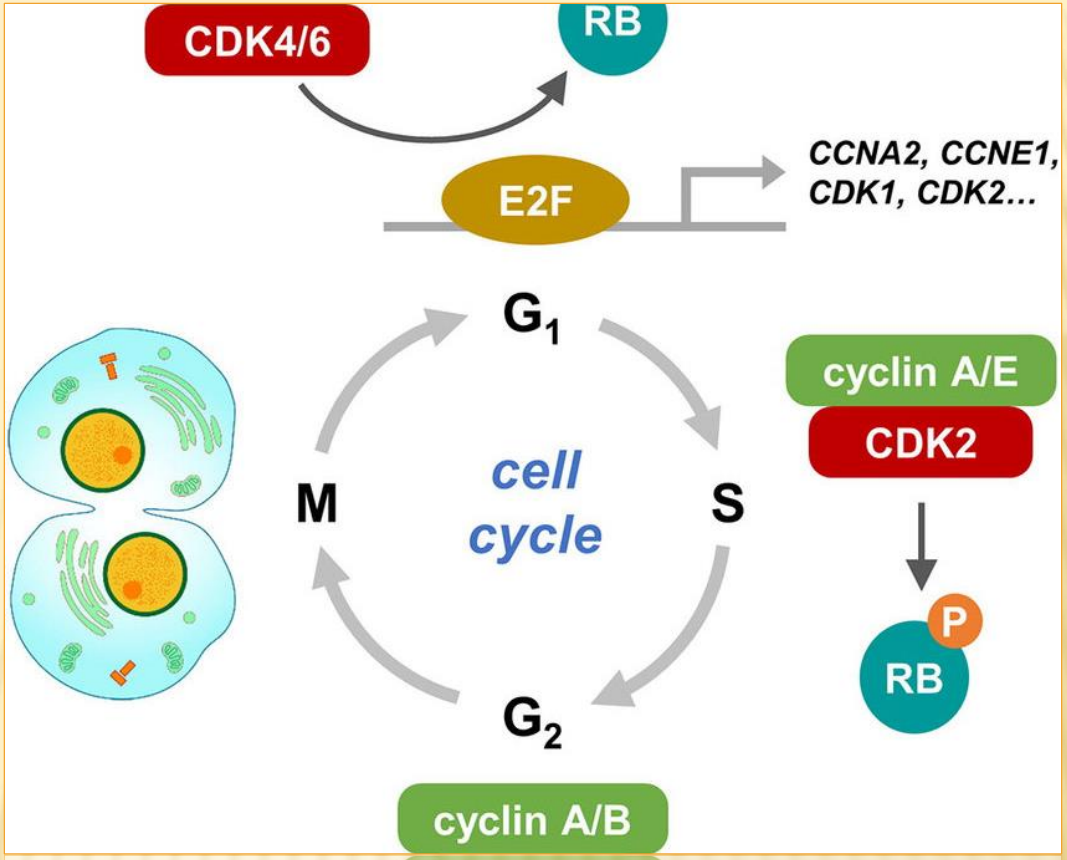
PRESENTATION FOR ENZYME CLASS

FERESHTEH NOROOZI

PK IN CELL CYCLE



PK IN CELL CYCLE

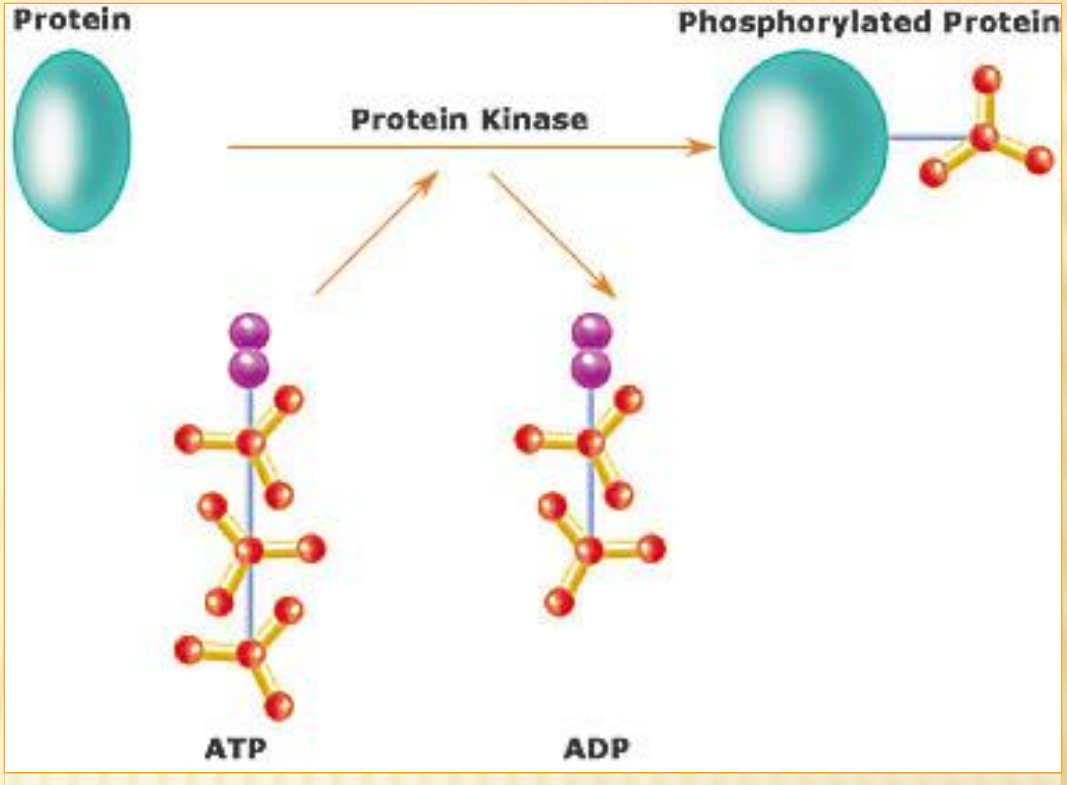


PROTEIN KINASE

- A **protein kinase** is a kinase enzyme that modifies other molecules, mostly proteins, by chemically adding phosphate groups to them (phosphorylation). Phosphorylation usually results in a functional change of the target protein (substrate) by changing enzyme activity, cellular location, or association with other proteins. The human genome contains about 518 protein kinase genes and they constitute about 2% of all human genes.

CHEMICAL ACTIVITY

- The chemical activity of a kinase involves transferring a phosphate group from a nucleoside triphosphate (usually ATP) and covalently attaching it to specific amino acids with a free hydroxyl group. Most kinases act on both serine and threonine (serine/threonine kinases), others act on tyrosine (tyrosine kinases), and a number act on all three (dual-specificity kinases). There are also protein kinases that phosphorylate other amino acids, including histidine kinases that phosphorylate histidine residues to create acid and heat-labile phosphoramidate bonds. Recent evidence preprinted at BioRxiv suggests widespread protein phosphorylation on multiple non-canonical amino acids, including motifs containing phosphorylated histidine, aspartate, glutamate, arginine and lysine in human HeLa cell extracts. Due to the chemical lability of these phosphorylated residues, special procedures and separation techniques are required for their preservation alongside classical Ser, Thr and Tyrphosphorylation



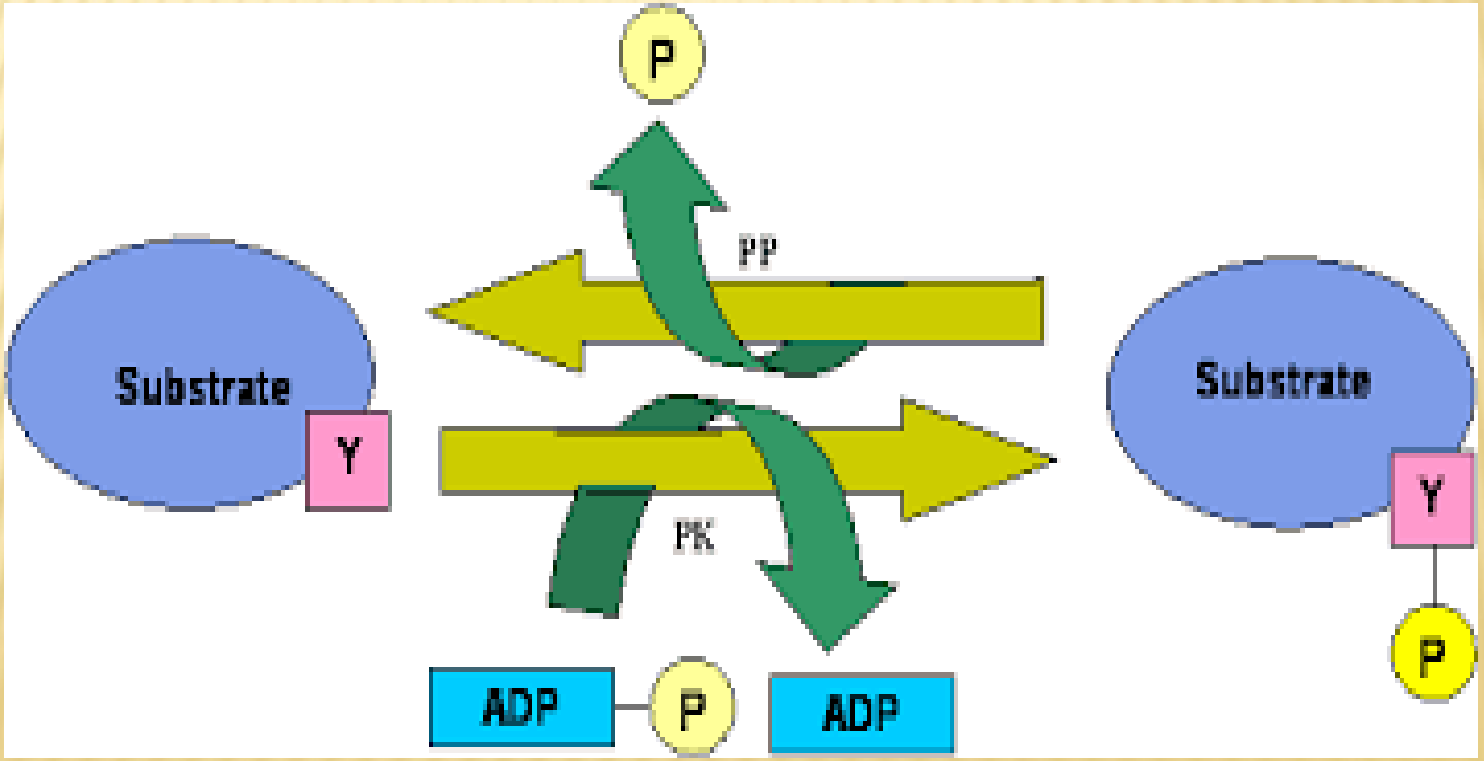
PROTEIN KINASE

PROTEIN KINASE GROUPS

- AGC kinases - containing PKA, PKC and PKG.
- CaM kinases - containing the calcium/calmodulin-dependent protein kinases.
- CK1 - containing the casein kinase 1 group.
- CMGC - containing CDK, MAPK, GSK3 and CLK kinases.
- STE - containing the homologs of yeast Sterile 7, Sterile 11, and Sterile 20 kinases.
- **TK - containing the tyrosine kinases.**
- TKL - containing the tyrosine-kinase like group of kinases.

TYROSINE KINASE

- A **tyrosine kinase** is an enzyme that can transfer a phosphate group from ATP to a protein in a cell. It functions as an "on" or "off" switch in many cellular functions. Tyrosine kinases are a subclass of protein kinase.
- The phosphate group is attached to the amino acid tyrosine on the protein.



FAMILIES

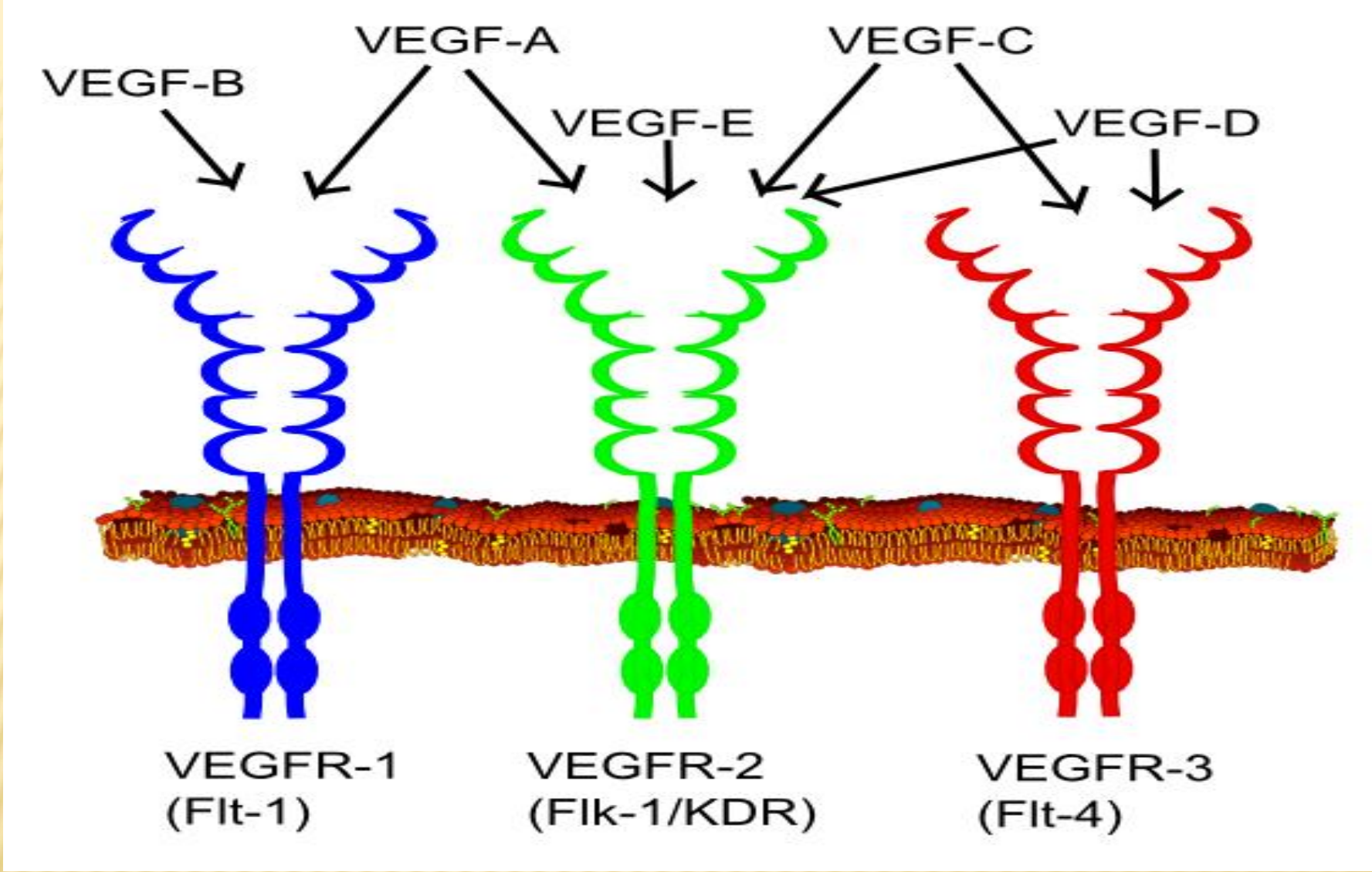
- More than 90 protein tyrosine kinases (PTKs) have been found in the human genome. They are divided into two classes, receptor and non-receptor tyrosine kinases.

RECEPTOR TYROSINE KINASE

- **Receptor tyrosine kinases** (**RTKs**) are the high-affinity cell surface receptors for many polypeptide growth factors, cytokines, and hormones. Of the 90 unique tyrosine kinase genes identified in the human genome, 58 encode receptor tyrosine kinase proteins.Receptor tyrosine kinases have been shown not only to be key regulators of normal cellular processes but also to have a critical role in the development and progression of many types of cancer. Mutations in receptor tyrosine kinases lead to activation of a series of signalling cascades which have numerous effects on protein expression. Receptor tyrosine kinases are part of the larger family of protein tyrosine kinases, encompassing the receptor tyrosine kinase proteins which contain a transmembrane domain, as well as the non receptor tyrosine kinases which do not possess transmembrane domains.

EXAMPLES OF RECEPTOR TYROSINE KINASE

- Regulatory molecules that stimulate cell proliferation are known as **growth factors.** Certain growth factors also guide the differentiation of cells into specialized cell types. Many growth factors signal via a signal transduction pathway involving **receptor tyrosine kinases.** Notable examples are:
- **Vascular-endothelial growth factor** (**VEGF**) This growth factor promotes new blood vessel growth, but is also important for maintenance of endothelial cells in the delicate filtration membrane of the kidney.
- **Neurotrophins** Neurotrophins promote the survival and differentiation of neurons.
- **Insulin-like growth factor-1** (**IGF-1**) This hormone/paracrine is produced in response to growth hormone, and is responsible for the majority of its growth-promoting effects.



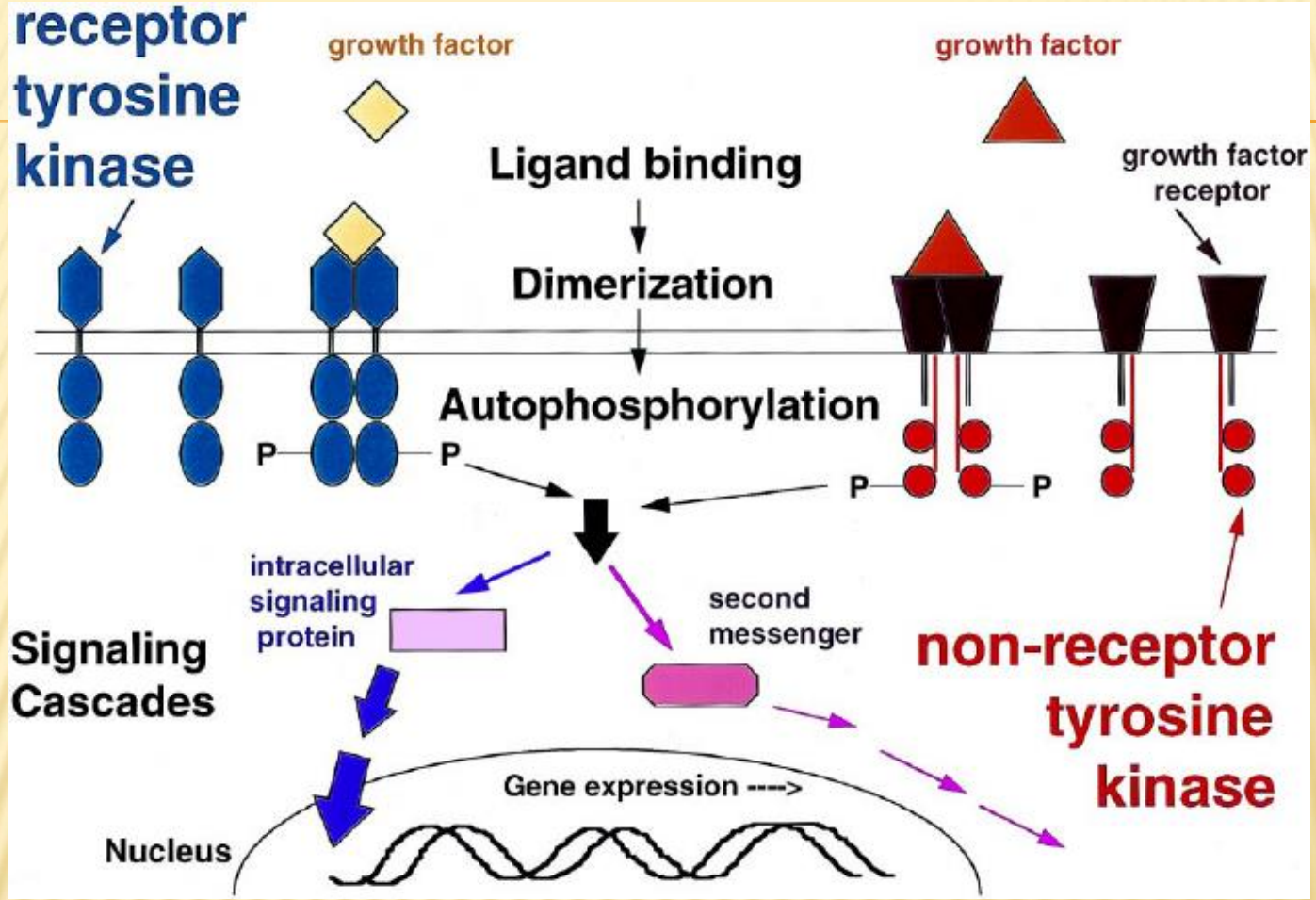
RECEPTOR TYROSINE KINASE

NON-RECEPTOR PROTEIN-TYROSINE KINASE

- Non-receptor protein-tyrosine kinase that regulates reorganization of the actin cytoskeleton, cell polarization, cell migration, adhesion, spreading and bone remodeling. Plays a role in the regulation of the humoral immune response, and is required for normal levels of marginal B-cells in the spleen and normal migration of splenic B-cells. Required for normal macrophage polarization and migration towards sites of inflammation. Regulates cytoskeleton rearrangement and cell spreading in T-cells, and contributes to the regulation of T-cell responses. Promotes osteoclastic bone resorption; this requires both PTK2B/PYK2 and SRC. May inhibit differentiation and activity of osteoprogenitor cells. Functions in signaling downstream of integrin and collagen receptors, immune receptors, G-protein coupled receptors (GPCR), cytokine, chemokine and growth factor receptors, and mediates responses to cellular stress. Forms multisubunit signaling complexes with SRC and SRC family members upon activation; this leads to the phosphorylation of additional tyrosine residues, creating binding sites for scaffold proteins, effectors and substrates. Regulates numerous signaling pathways. Promotes activation of phosphatidylinositol 3-kinase and of the AKT1 signaling cascade. Promotes activation of NOS3. Regulates production of the cellular messenger cGMP. Promotes activation of the MAP kinase signaling cascade, including activation of MAPK1/ERK2, MAPK3/ERK1 and MAPK8/JNK1. Promotes activation of Rho family GTPases, such as RHOA and RAC1. Recruits the ubiquitin ligase MDM2 to P53/TP53 in the nucleus, and thereby regulates P53/TP53 activity, P53/TP53 ubiquitination and proteasomal degradation. Acts as a scaffold, binding to both PDPK1 and SRC, thereby allowing SRC to phosphorylate PDPK1 at 'Tyr-9, 'Tyr-373', and 'Tyr-376'. Promotes phosphorylation of NMDA receptors by SRC family members, and thereby contributes to the regulation of NMDA receptor ion channel activity and intracellular Ca^{2+} levels. May also regulate potassium ion transport by phosphorylation of potassium channel subunits. Phosphorylates SRC; this increases SRC kinase activity. Phosphorylates ASAP1, NPHP1, KCNA2 and SHC1. Promotes phosphorylation of ASAP2, RHOU and PXN; this requires both SRC and PTK2/PYK2

NON-RECEPTOR TYROSINE KINASE

- **Non-receptor tyrosine kinases** (**nRTKs**) are cytosolic enzymes that are responsible for catalysing the transfer of a phosphate group from a nucleoside triphosphate donor, such as ATP, to tyrosine residues in proteins. Non-receptor tyrosine kinases are a subgroup of protein family tyrosine kinases, enzymes that can transfer the phosphate group from ATP to a tyrosine residue of a protein (phosphorylation). These enzymes regulate many cellular functions by switching on or switching off other enzymes in a cell.
- Unlike the receptor tyrosine kinases (RTKs), the second subgroup of tyrosine kinases, the non-receptor tyrosine kinases are cytosolic enzymes. Thirty-two non-receptor tyrosine kinases have been identified in human cells .Non-receptor tyrosine kinases regulate cell growth, proliferation, differentiation, adhesion, migration and apoptosis, and they are critical components in the regulation of the immune system.


receptor tyrosine kinase
growth factor
growth factor
Ligand binding
growth factor receptor
Dimerization
Autophosphorylation
P
P
P
P
intracellular signaling protein
second messenger
Signaling Cascades
non-receptor tyrosine kinase
Gene expression ---->
Nucleus

NRTKS & RTKS

EXAMPLE OF NON-RECEPTOR TYROSINE KINASE

- SRC

DISCOVERY

- In 1979, J. Michael Bishop and Harold E. Varmus discovered that normal chickens possess a gene that is structurally closely related to *v-Src*. The normal cellular gene was called *c-src* (cellular-src). This discovery changed the current thinking about cancer from a model wherein cancer is caused by a foreign substance (a viral gene) to one where a gene that is normally present in the cell can cause cancer. It is believed that at one point an ancestral virus mistakenly incorporated the *c-Src* gene of its cellular host. Eventually this normal gene mutated into an abnormally functioning oncogene within the Rous sarcoma virus. Once the oncogene is transfected back into a chicken, it can lead to cancer.

C SRC

Protein names[i]	*Recommended name:* **Proto-oncogene tyrosine-protein kinase Src** (EC:2.7.10.26 Publications)*Alternative name(s):* Proto-oncogene c-Src pp60c-src*Short name:* p60-Src
Gene names[i]	*Name:*SRCSynonyms:SRC1
Organism[i]	Homo sapiens (Human)
Taxonomic identifier[i]	9606 [NCBI]
Taxonomic lineage[i]	Eukaryota › Metazoa › Chordata › Craniata › Vertebrata › Euteleostomi › Mammalia › Eutheria › Euarchontoglires › Primates › Haplorrhini › Catarrhini › Hominidae › Homo
Proteomes[i]	•UP000005640 Component[i]: Chromosome 20

Amino acid modifications

Feature key	Position(s)	DescriptionActions	Graphical view	Length
Lipidation[i]	2	N-myristoyl glycine1 Publication		1
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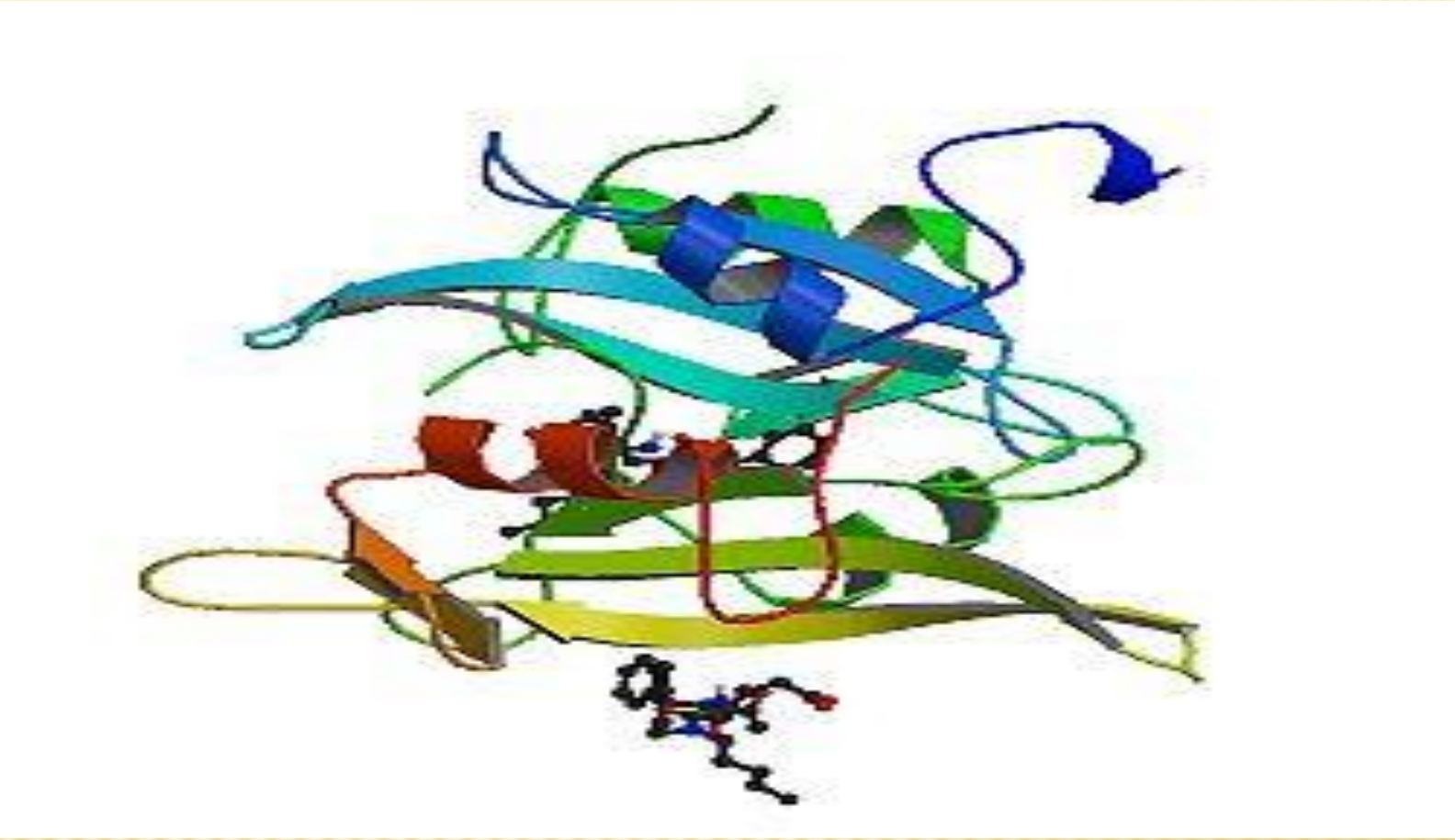
Modified residue[i]	75	Phosphoserine; by CDK5Combined sources1 Publication		1
Modified residue[i]	187	PhosphotyrosineBy similarity		1
Modified residue[i]	419	Phosphotyrosine; by autocatalysis5 Publications		1
Modified residue[i]	419	Phosphotyrosine; by FAK2By similarity		1
Modified residue[i]	530	Phosphotyrosine; by CSKCombined sources4 Publications		



C-SRC

PROTO-ONCOGENE TYROSINE-PROTEIN KINASE SRC

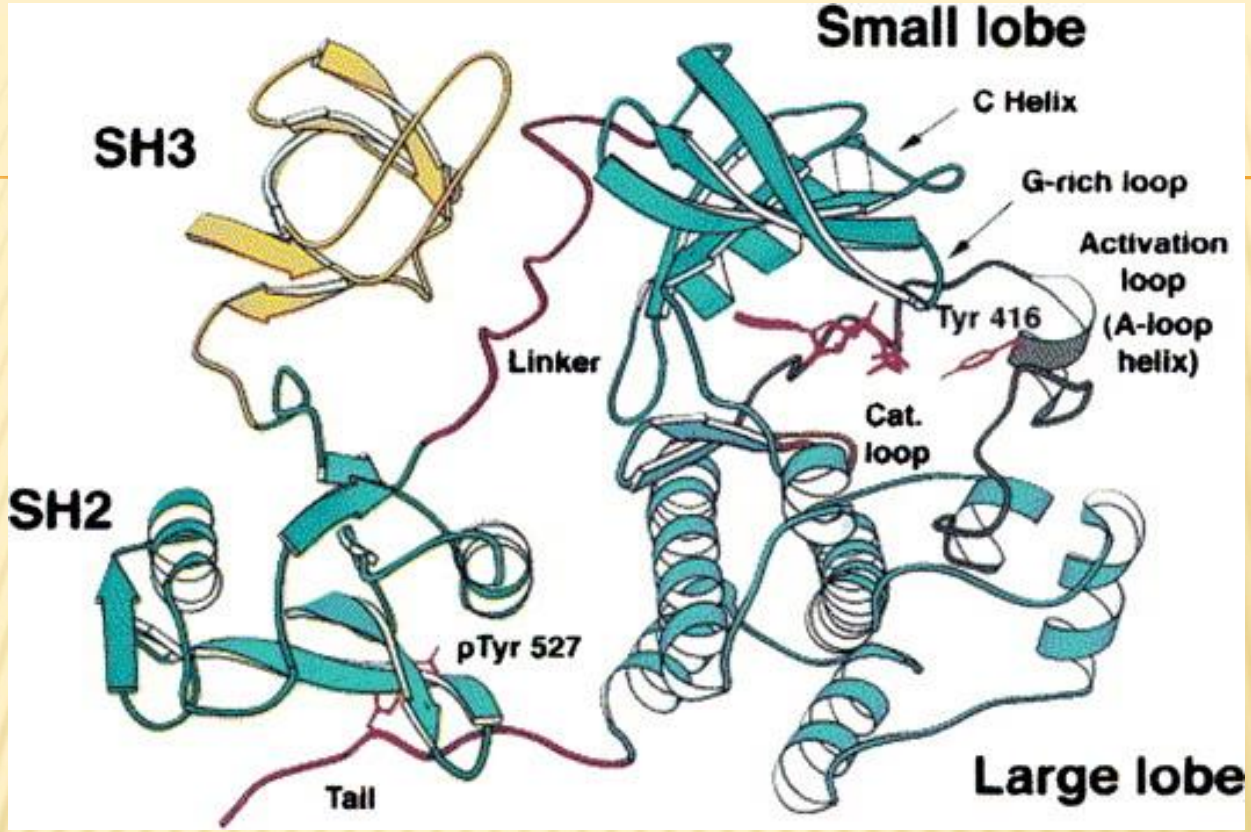
- **Proto-oncogene tyrosine-protein kinase Src**, also known as **proto-oncogene c-Src** or simply **c-Src** , is a non-receptor tyrosine kinase protein that in humans is encoded by the SRC gene. This protein phosphorylates specific tyrosine residues in other proteins.
- c-Src stands for "cellular Src kinase" and should not be confused with "C-terminal Src kinase" (CSK) which is an enzyme which phosphorylates c-Src at its C-terminus and provides negative regulation of Src's enzymatic activity. Similarly, c-Src should not be mistaken for v-Src, a viral (hence the prefix *v-*) gene that shares similarity with c-Src and is also an oncogene, which can be found in Rous sarcoma virus.
- An elevated level of activity of c-Src tyrosine kinase is suggested to be linked to cancer progression by promoting other signals.[5] Mutations in this gene could be involved in the malignant progression of colon cancer.
- This proto-oncogene may play a role in the regulation of embryonic development and cell growth.
- c-Src includes an SH2 domain, an SH3 domain, and a tyrosine kinase domain.
- **Src** (pronounced "sarc" as it is short for sarcoma) was originally discovered by J. Michael Bishop and Harold E. Varmus, for which they were awarded the 1989 Nobel Prize in Physiology or Medicine.[6] It belongs to a family of Src family kinases.This gene is similar to the v-Src gene of Rous sarcoma virus.
- Two transcript variants encoding the same protein have been found for this gene

STRUCTURE

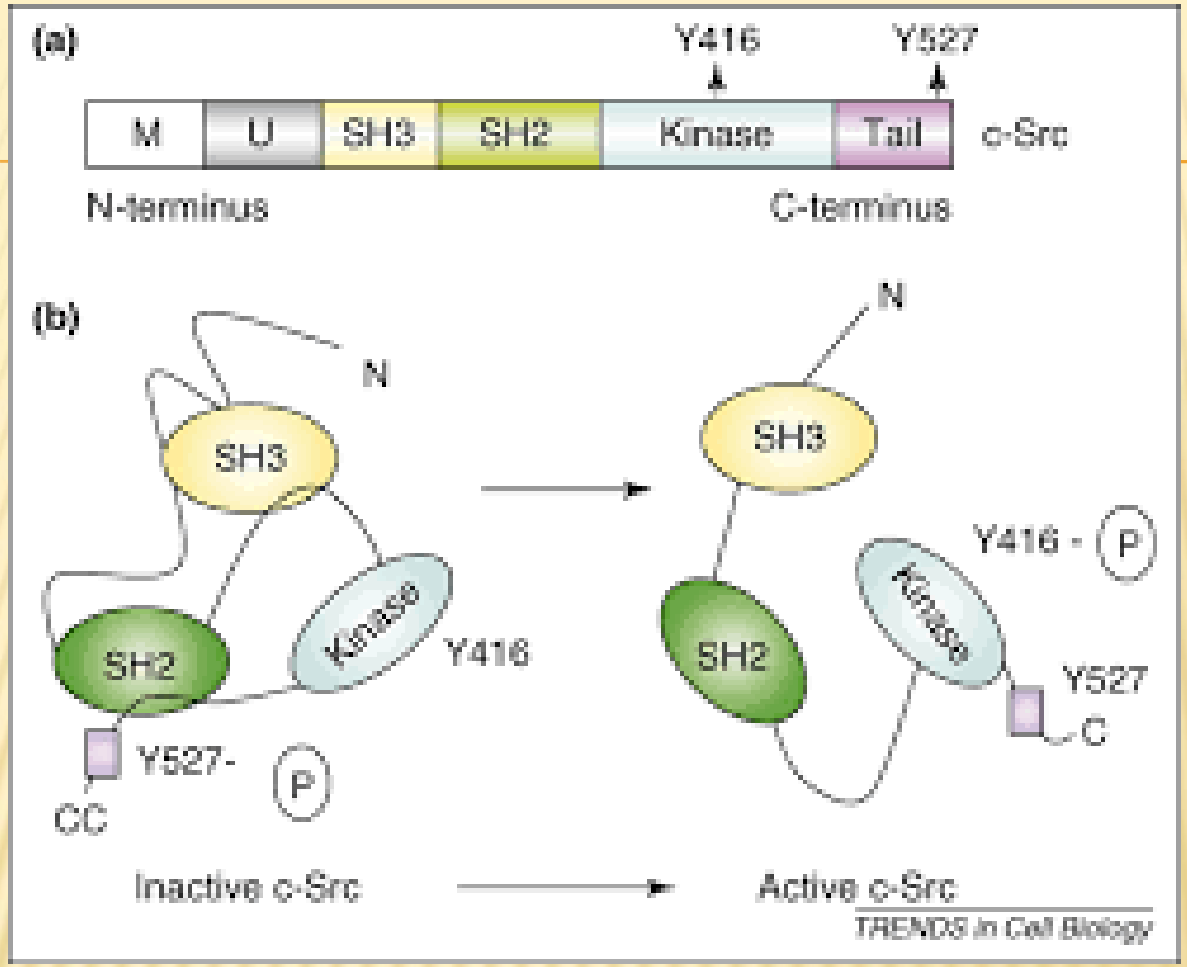
- There are 9 members part of the Src family kinases: **c-Src**, Yes, Fyn, Fgr, Yrk, Lyn, Blk, Hck, and Lck. The expression of these Src family members are not the same throughout all tissues and cell types. Src, Fyn and Yes are expressed ubiquitously in all cell types while the others are generally found in hematopoietic cells.
- c-Src is made up of 6 functional regions: Src homology 4 domain (SH4 domain), unique region, SH3 domain, SH2 domain, catalytic domain and short regulatory tail. When Src is inactive, the phosphorylated tyrosine group at the 527 position interacts with the SH2 domain which helps the SH3 domain interact with the flexible linker domain and thereby keeps the inactive unit tightly bound. The activation of c-Src causes the dephosphorylation of the tyrosine 527. This induces long-range allostery via protein domain dynamics, causing the structure to be destabilized, resulting in the opening up of the SH3, SH2 and kinase domains and the autophosphorylation of the residue tyrosine 416.
- The autophosphorylation of Y416 as well as phosphorylation of selected Src substrates is enhanced through dimerization of c-Src. The dimerization of c-Src is mediated by the interaction of the myristoylated N-terminal region of one partner and the kinase domain of another partner.Both the N-terminally attached myristic acid and the peptide sequences of the unique region are involved in the interaction. Given the versatility inherent in this intrinsically disordered region, its multisite phosphorylations, and its divergence within the family, the unique domain likely functions as a central signaling hub overseeing much of the enzymatic activities and unique functions of Src family kinases.
- c-Src can be activated by many transmembrane proteins that include: adhesion receptors, receptor tyrosine kinases, G-protein coupled receptors and cytokine receptors. Most studies have looked at the receptor tyrosine kinases and examples of these are platelet derived growth factor receptor (PDGFR) pathway and epidermal growth factor receptor (EGFR).
- Src contains at least three flexible protein domains, which, in conjunction with myristoylation, can mediate attachment to membranes and determine subcellular localization

DOMAINS AND REPEATS

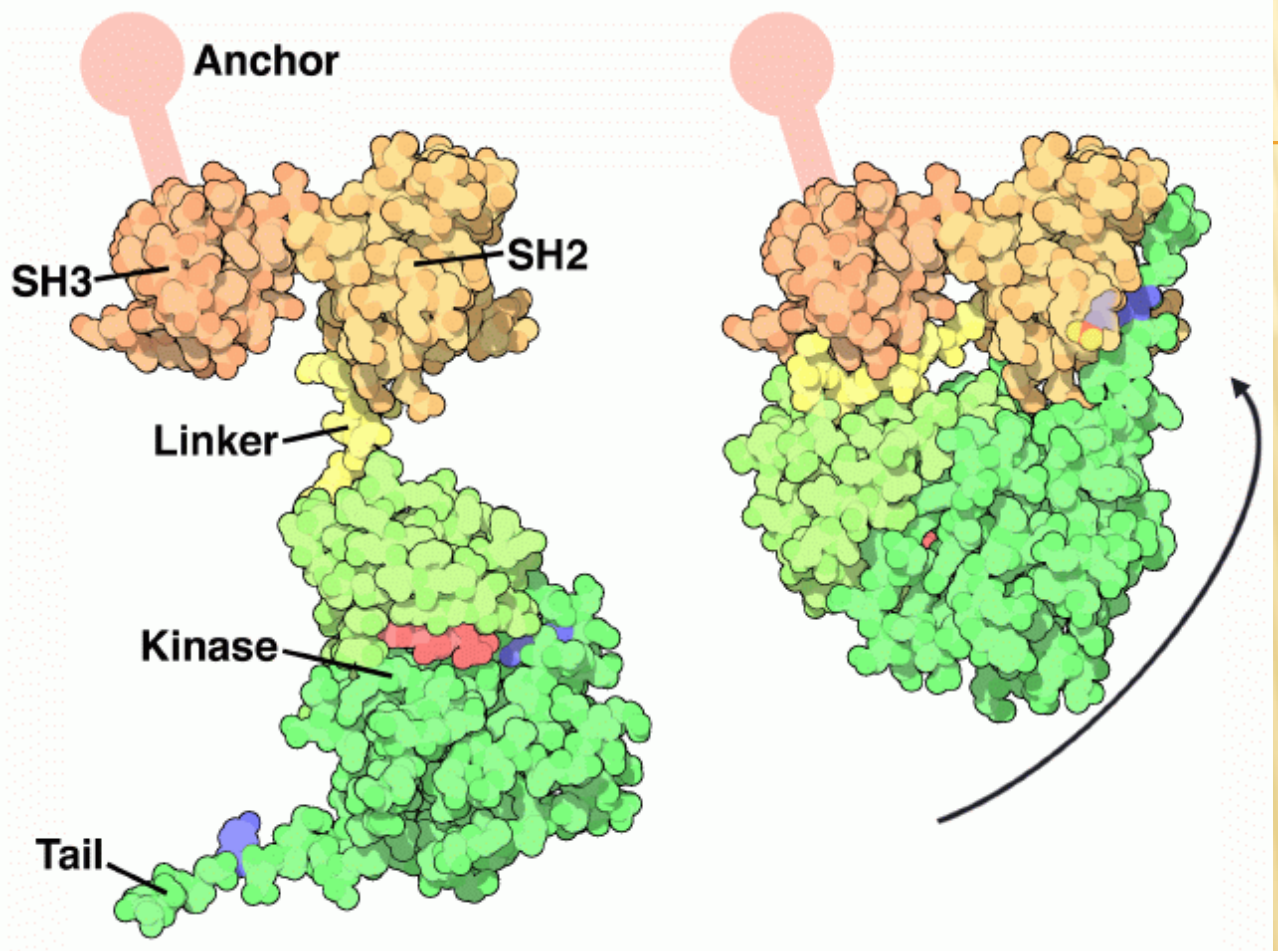
Feature key	Position(s)	DescriptionActions	Graphical view	Length
Domain[i]	84 – 145	SH3PROSITE-ProRule annotationAdd BLAST		62
Domain[i]	151 – 248	SH2PROSITE-ProRule annotationAdd BLAST		98
Domain[i]	270 – 523	Protein kinasePROSITE-ProRule annotation		



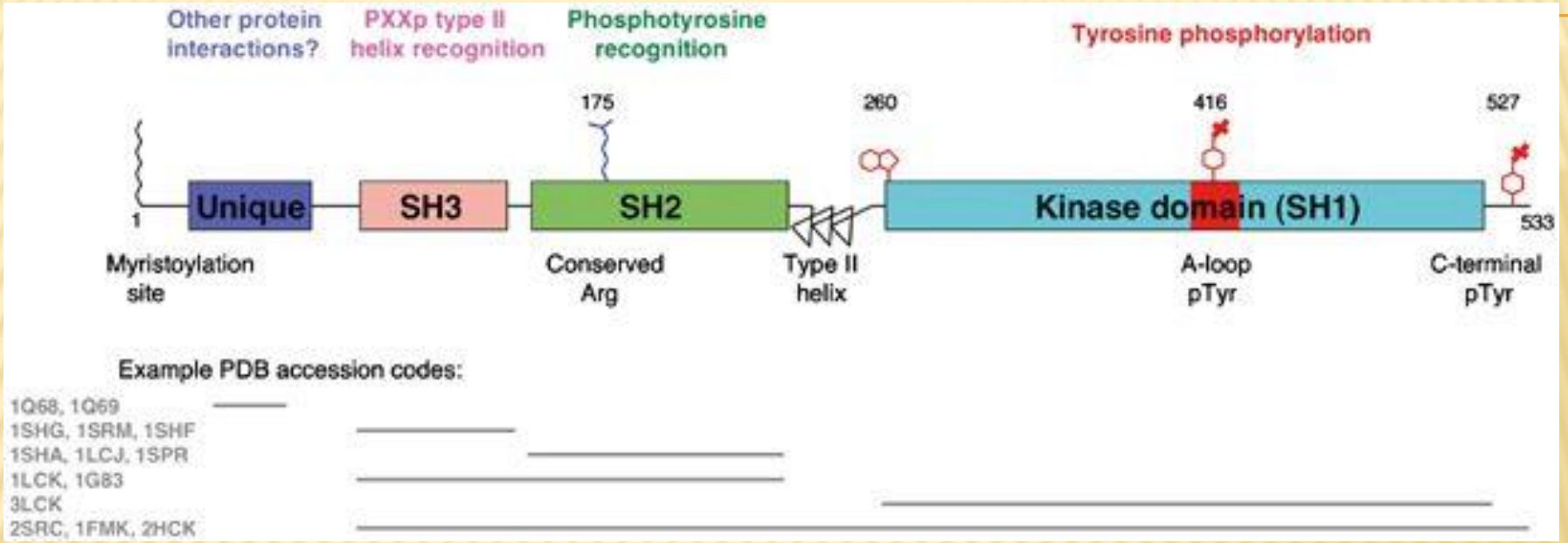
STRUCTURE OF CSRC



STRUCTURE OF CSRC

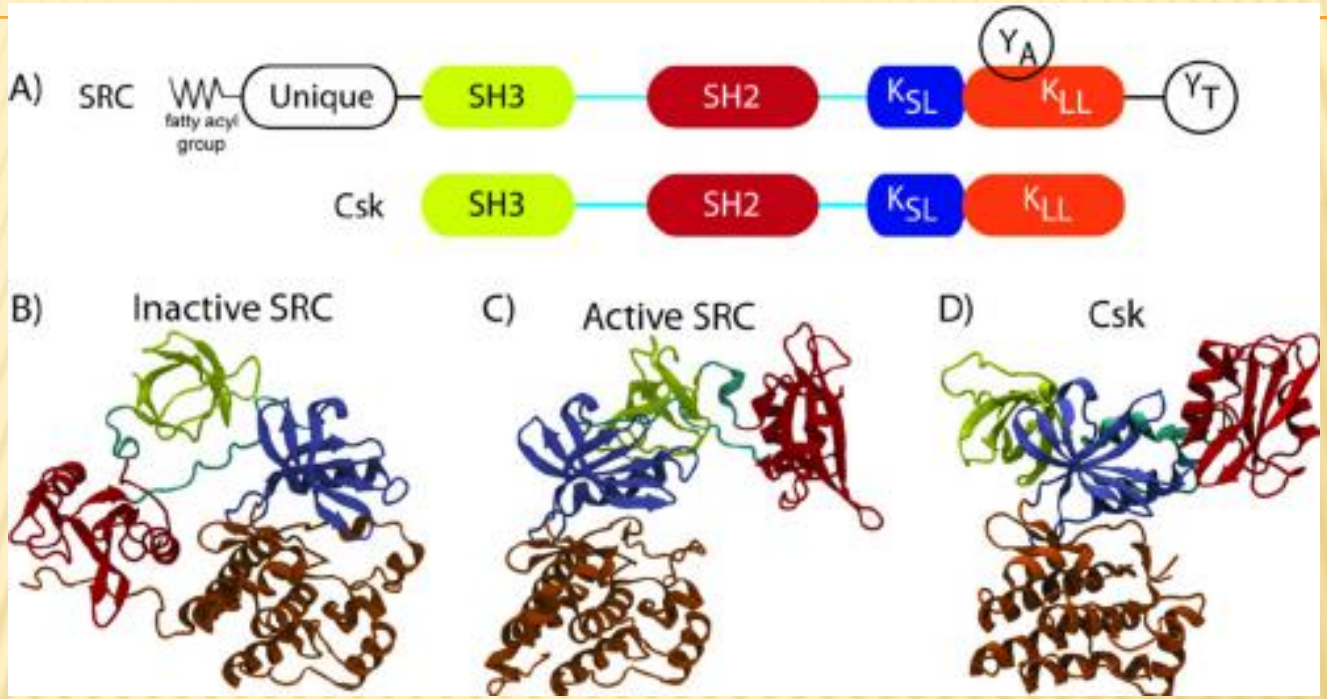


STRUCTURE OF CSRC



STRUCTURE OF CSRC

STRUCTURE OF CSRC



FUNCTION

- This proto-oncogene may play a role in the regulation of embryonic development and cell growth.
- When src is activated, it promotes survival, angiogenesis, proliferation and invasion pathways. It also regulates angiogenic factors and vascular permeability after focal cerebral ischemia-reperfusion,and regulates matrix metalloproteinase-9 activity after intracerebral hemorrhage

ROLE IN CANCER

- The activation of the c-Src pathway has been observed in about 50% of tumors from colon, liver, lung, breast and the pancreas. Since the activation of c-Src leads to the promotion of survival, angiogenesis, proliferation and invasion pathways, the aberrant growth of tumors in cancers is observed. A common mechanism is that there are genetic mutations that result in the increased activity or the overexpression of the c-Src leading to the constant activation of the c-Src.

MUTAGENESIS

Feature key	Position(s)	DescriptionActions	Graphical view	Length
Mutagenesis[i]	298	K → M: Kinase inactive. Abolishes ubiquitination promoted by CBLC. 1 Publication		1
Mutagenesis[i]	302	P → E: Kinase active. Interacts with PDLIM4; when associated with E-307 and F-419. 1 Publication		1
Mutagenesis[i]	307	P → E: Kinase active. Interacts with PDLIM4; when associated with E-302 and F-419. 1 Publication		1
Mutagenesis[i]	419	Y → F: Loss of kinase activity. Loss of interaction with PDLIM4. 1 Publication		

COLON

- The activity of c-Src has been best characterized in colon cancer. Researchers have shown that Src expression is 5 to 8 fold higher in premalignant polyps than normal mucosa.The elevated c-Src levels have also been shown to have a correlation with advanced stages of the tumor, size of tumor, and metastatic potential of tumors.

BREAST

- EGFR activates c-Src while EGF also increases the activity of c-Src. In addition, overexpression of c-Src increases the response of EGFR-mediated processes. So both EGFR and c-Src enhance the effects of one another. Elevated expression levels of c-Src were found in human breast cancer tissues compared to normal tissues.
- Overexpression of Human Epidermal Growth Factor Receptor 2 (HER2), also known as erbB2, is correlated with a worse prognosis for breast cancer.Thus, c-Src plays a key role in the tumor progression of breast cancers.

PROSTATE

- Members of the Src family kinases Src, Lyn and Fgr are highly expressed in malignant prostate cells compared to normal prostate cells. When the primary prostate cells are treated with KRX-123, which is an inhibitor of Lyn, the cells in vitro were reduced in proliferation, migration and invasive potential.So the use of a tyrosine kinase inhibitor is a possible way of reducing the progression of prostate cancers

AS A DRUG TARGET

- A number of tyrosine kinase inhibitors that target c-Src tyrosine kinase (as well as related tyrosine kinases) have been developed for therapeutic use. One notable example is dasatinib which has been approved for the treatment of chronic myeloid leukemia (CML) and Philadelphia chromosome-positive (PH+) acute lymphocytic leukemia (ALL).Dasatinib is also in clinical trials for the use in non-Hodgkin's lymphoma, metastatic breast cancer and prostate cancer. Other tyrosine kinase inhibitor drugs that are in clinical trials include bosutinib,bafetinib, AZD-530, XLI-999, KX01 and XL228.

REFRENCES

- *lehninger* book
- https://www.researchgate.net/figure/Structural-and-regulatory-features-of-c-Src-and-Csk-A-domain-organization-c-Src_fig1_46036193
- http://emboj.embopress.org/content/16/24/7261.figures-only
- https://courses.washington.edu/conj/bess/rtk/rtk.htm
- https://www.uniprot.org/uniprot/P12931