Tutorial for

KEGG Online Database

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Acknowledgment:

Thanks to Dr. Peter Abuja for his expert knowledge in the area of metabolisms and pathways.

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This document will be available at:

http://www.icg.tu-graz.ac.at/research/CGIS/GENVIEW/

Snapshots from KEGG web interface

http://www.genome.jp/kegg/pathway.html visited 17-08-2006

Example: Pathway „***metionine metabolism - Reference pathway*** „

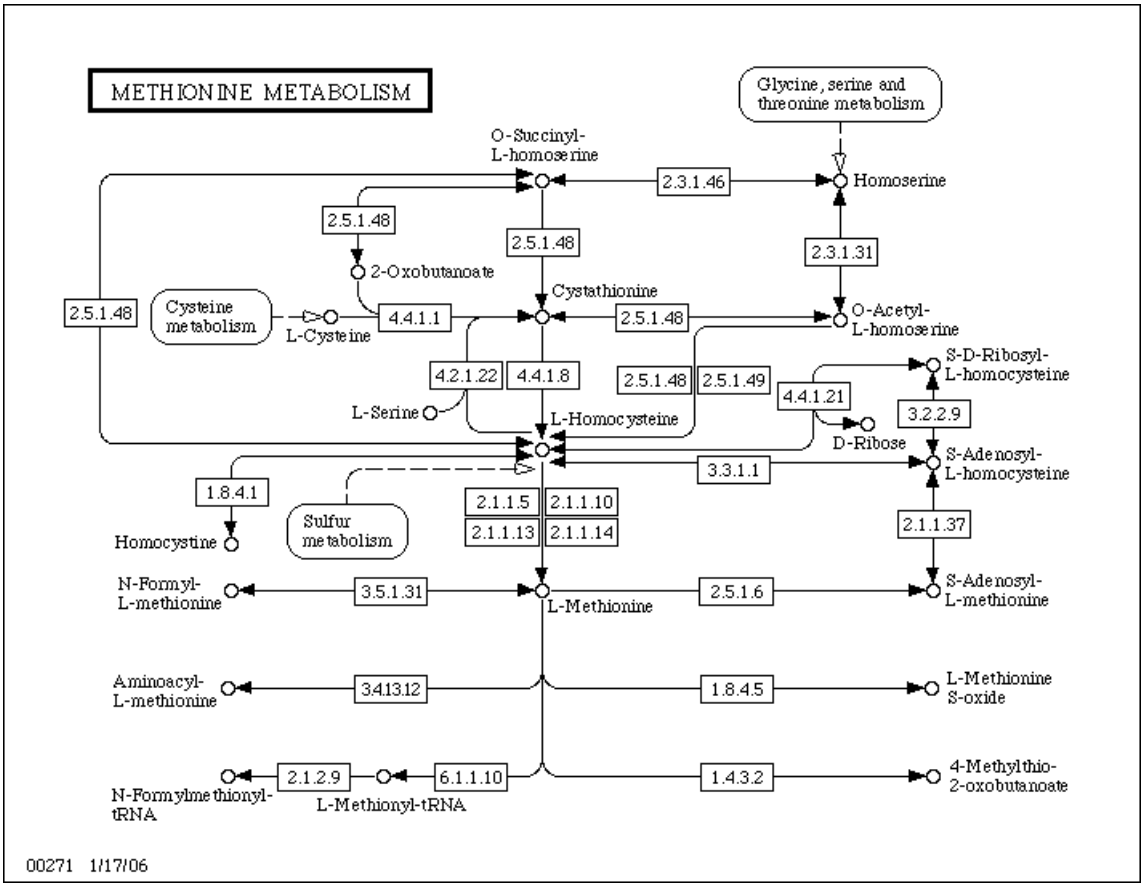


Figure 1:

Klick on [2.5.1.48] opens this:

ENZYME: 2.5.1.48 Help

Entry
EC 2.5.1.48 Enzyme

Name cystathionine gamma-synthase;
O-succinyl-L-homoserine succinate-lyase (adding cysteine);
O-succinylhomoserine (thiol)-lyase;
homoserine O-transsuccinylase;
O-succinylhomoserine synthase;
O-succinylhomoserine synthetase;
cystathionine synthase;
cystathionine synthetase;
homoserine transsuccinylase

Class Transferases
Transferring alkyl or aryl groups, other than methyl groups
Transferring alkyl or aryl groups, other than methyl groups

Sysname O4-succinyl-L-homoserine:L-cysteine
S-(3-amino-3-carboxypropyl)transferase

Reaction O-succinyl-L-homoserine + L-cysteine = L-cystathionine + succinate
[RN:R00999 R01288 R02508 R03132 R03260 R04944 R04945 R04946]

Substrate O-Succinyl-L-homoserine [CPD:C01118];
L-Cysteine [CPD:C00097]

Product L-Cystathionine [CPD:C02291];
Succinate [CPD:C00042]

Cofactor Pyridoxal phosphate [CPD:C00018]

Comment A pyridoxal-phosphate protein. Also reacts with hydrogen sulfide and methanethiol as replacing agents, producing homocysteine and methionine, respectively. In the absence of thiol, can also catalyse beta,gamma-elimination to form 2-oxobutanoate, succinate and ammonia.

Pathway PATH: map00271 Methionine metabolism
PATH: map00272 Cysteine metabolism
PATH: map00450 Selenoamino acid metabolism
PATH: map00920 Sulfur metabolism

Ortholog KO: K01739 cystathionine gamma-synthase

Genes XLA: 494673(LOC494673)
XTR: 394634(MGC75946)
ATH: At3g01120(T4P13.19)
CME: CMF156C
SCE: YJR130C(STR2) YML082W
AGO: AER164C(AER164Cp)
CAL: orf19.1033(STR2) orf19.7297
SPO: SPBC15D4.09c
CNE: CNC01220
LMA: LmjF35.3230
EHI: 132.t00018 389.t00003 395.t00003
ECO: b3939(metB)
ECJ: JW3910(metB)
...: reduced the list of 191 gene in total

Reference 1 [PMID:5340123]
Flavin M, Slaughter C.
Enzymatic synthesis of homocysteine or methionine directly from

O-succinyl-homoserine.
Biochim. Biophys. Acta. 132 (1967) 400-5.
2 [PMID:5922970]
Kaplan MM, Flavin M.
Cystathionine gamma-synthetase of Salmonella. Catalytic properties
of a new enzyme in bacterial methionine biosynthesis.
J. Biol. Chem. 241 (1966) 4463-71.
3 [PMID:6016326]
Wiebers JL, Garner HR.
Homocysteine and cysteine synthetases of Neurospora crassa.
Purification, properties, and feedback control of activity.
J. Biol. Chem. 242 (1967) 12-23.
4
Wiebers, J.L. and Garner, H.R. Acyl derivatives of homoserine as
substrates for homocysteine synthesis in Neurospora crassa, yeast,
and Escherichia coli. J. Biol. Chem. 242 (1967) 5644-5649.
5 [PMID:9843488]
Clausen T, Huber R, Prade L, Wahl MC, Messerschmidt A.
Crystal structure of Escherichia coli cystathionine gamma-synthase
at 1.5 A resolution.
EMBO. J. 17 (1998) 6827-38.
6 [PMID:9531508]
Ravanel S, Gakiere B, Job D, Douce R.
Cystathionine gamma-synthase from Arabidopsis thaliana: purification
and biochemical characterization of the recombinant enzyme
overexpressed in Escherichia coli.
Biochem. J. 331 (Pt 2) (1998) 639-48.

Other DBs IUBMB Enzyme Nomenclature: 2.5.1.48
ExPASy - ENZYME nomenclature database: 2.5.1.48
ERGO genome analysis and discovery system: 2.5.1.48
BRENDA, the Enzyme Database: 2.5.1.48
CAS: 9030-70-0

LinkDB All DBs

=> **Original format**

DBGET integrated database retrieval system, GenomeNet

Table 1

Klick on pathway „PATH: map00271 metionine metabolism“ shows this:

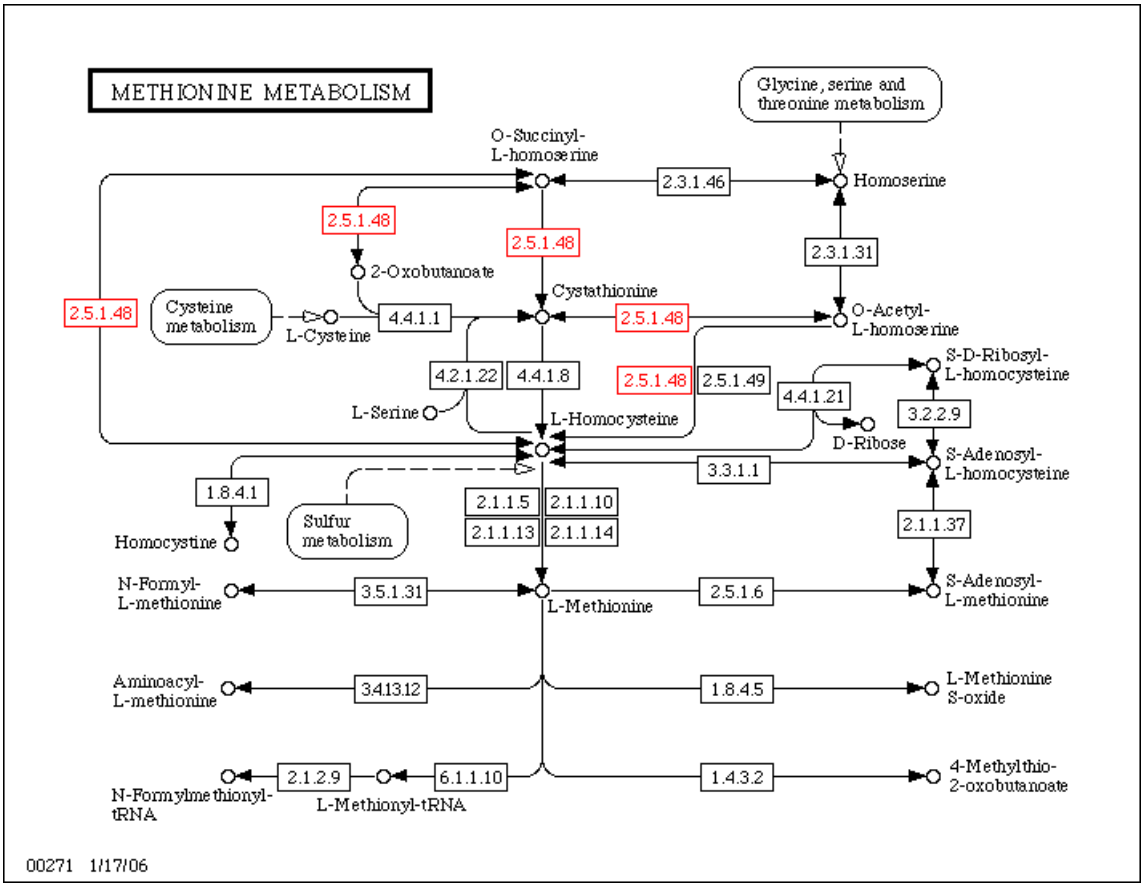


Figure 2:

Klick on Cystathionine opens this page:

=> **Original format**

DBGET integrated database retrieval system, GenomeNet

Table 2

Example for a reaction:

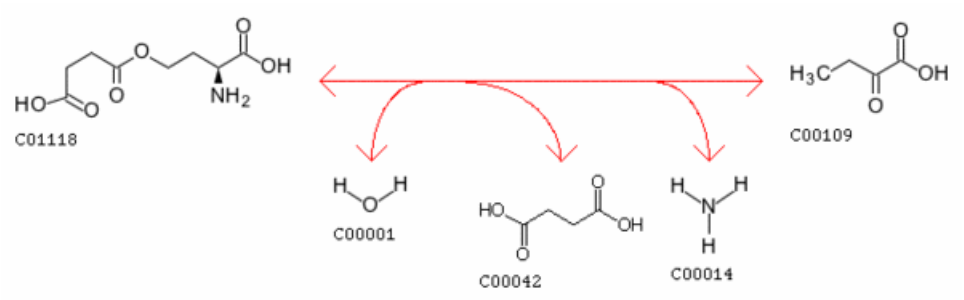
REACTION: R00999 Help

Entry R00999 Reaction

Name O-Succinyl-L-homoserine succinate-lyase (adding cysteine)

Definition O-Succinyl-L-homoserine + H2O <=> 2-Oxobutanoate + Succinate + NH3

Equation C01118 + C00001 <=> C00109 + C00042 + C00014



RPair RP: A00132 C00042_C01118 main
RP: A01229 C00109_C01118 main

Pathway PATH: rn00271 Methionine metabolism

Enzyme 2.5.1.48

Ortholog KO: K01739 cystathionine gamma-synthase

LinkDB All DBs

=> **Original format**

DBGET integrated database retrieval system, GenomeNet

Table 3

Flow Charts:

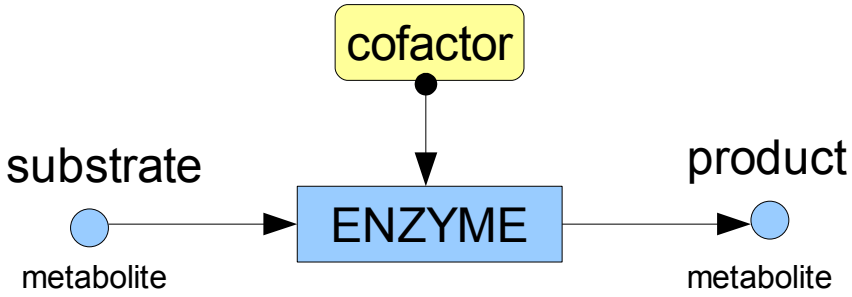


Figure 3: Basic graph with metabilite and enzyme regulated by cofactor

Regulation of enzymes and metabolites

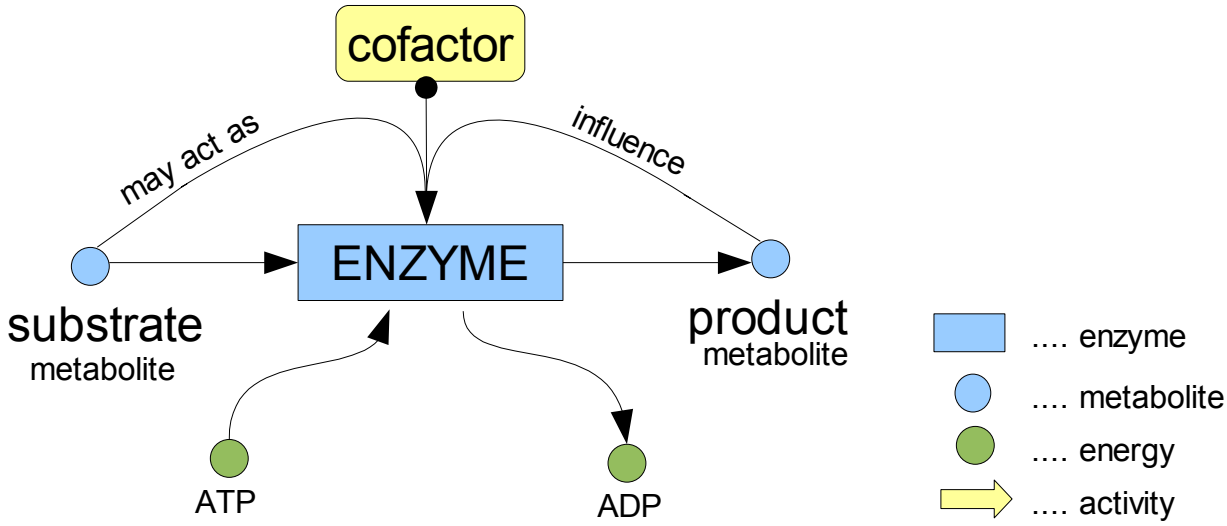


Figure 5: Exdented model of pathway

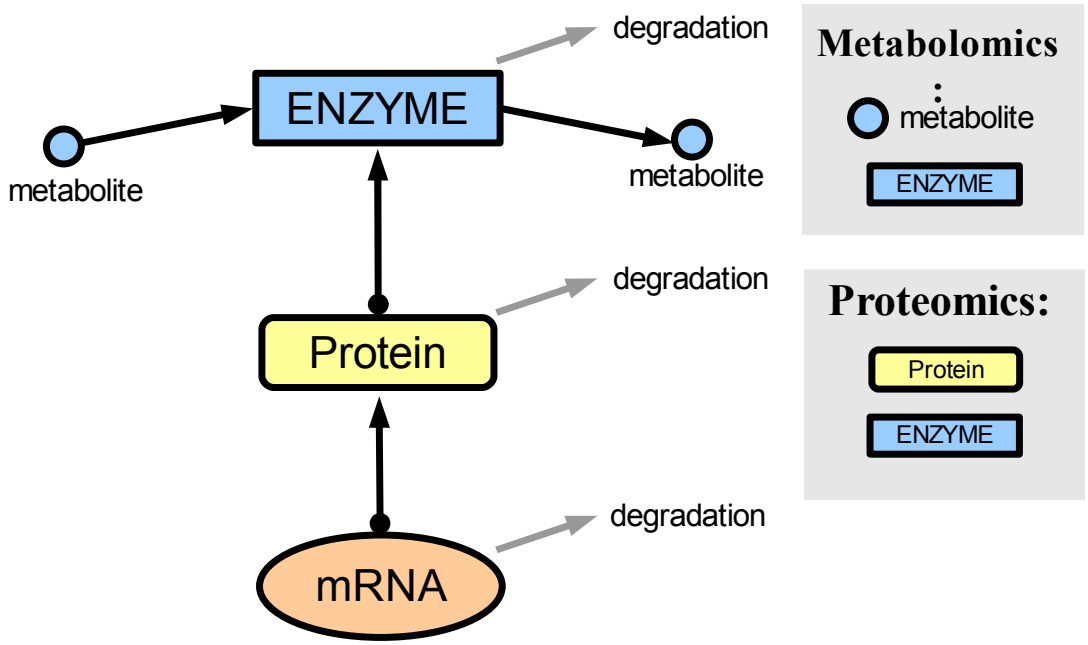


Figure 6: Interrelationship of mRNA, protein, enzyme and metabolism

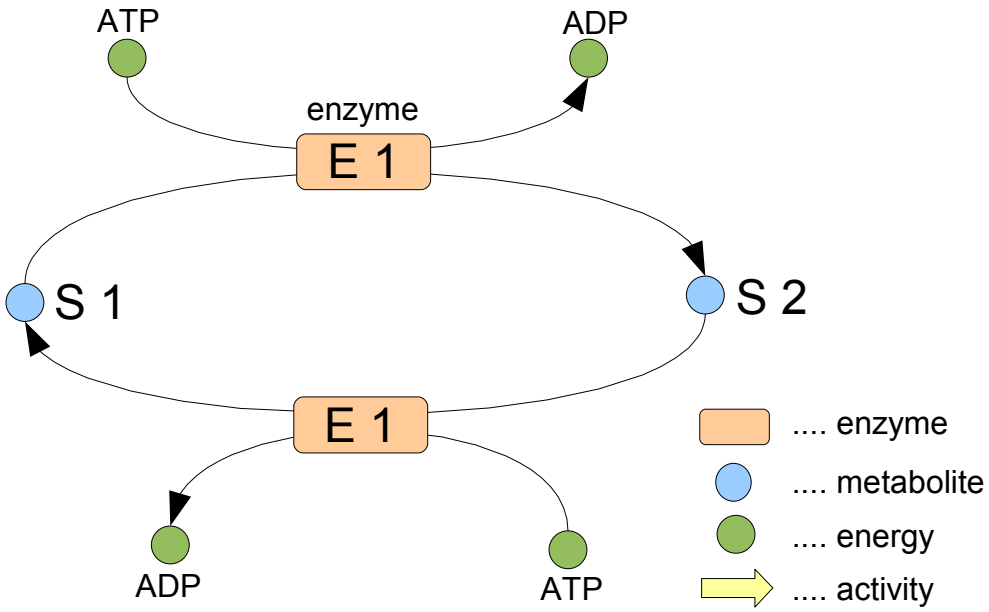


Figure 7: Two metabolits and two enzymes form a switch, that consumes energy

Concepts:

- Inside pathway: 2 step, 3 step relations inside the graph fram a given Protein or metabolit
- Take connectivity of metabolit into account (i.e. google principle)
- Use connectivity of metabolit as wight for its importance inside pathways
  (Spring mass modell / Graviation modell)
- Interrelations/ interaction:
  - Kinase ==> Protein
  - Protein <==> Protein interrelations
- Visualize flows from:

  Substrate ==>
  (Enzyme/ Protein) <-- using cofactor ==>
  metabolit A ==>
  (Enyzm / Protein) ==>
  metabolit B

A possibility here could be the application of Petri-Nets (http://en.wikipedia.org/wiki/Petri_net)

- Semantic Zooming:
  - hide / show metabolits
  - collapse protein chains
  - collapse pathway inside another pathway
  - magic lense for sematic zooming
- Messure activity of metabolids:
  required for detailed analysis, but not done within Micro Array analysis
- Position of enzyme inside cell
- Compartments the enzyme is present in
- Handle enzyme casscades ==> if top element of enzyme-casscade is active, the casscade might be active to. Thus only the result of the casscade is active, but not its intermediate enzyms.

- Important fields of „Enzymes“ in KEEG (see table 1):

  Entry: unique international standardized identifyier
  Name: known names for this molecule and aliases for it
  Reaction: chemical reaction
  Substrate: Input to the Enzyme
  Product: Output of the Enzyme
  Cofactor: if set, it is nesseccary to activate the Enzyme
  Pathway: shows only active pathways linked to one Enzyme but not pathways influenced by this enzyme!
  Ortholog: special genes (see glossary)
  Genes: ???

- Important fields of „Compounds“ in KEGG (see table 2):

  Entry: unique code (used by KEGG? Only or also international unique?)
  Name: known names for this molecule and aliases for it
  Mass: mass of this molecule (mass unit?)
  Structur: chemical structure of thie molecule
  Reaction: chemical reaction, were this compound is part of
  Pathway: list of active pathways, were this compound can be found
  Enzyme: list of enzymes capeable of handling this compound, either as input or as output; also know as „Substrate“ and „Product“

- Important fields of „Reaction“ in KEGG (see table 3):

  Entry: unique name (probably only in KEGG)
  Name: name that defines this reaction
  Definition: define this reaction
  Equation: link to compunds of this reaction
  Rpair: ??
  Pathway: list of pathways were this reaction is part of
  Enzyme: list of enzymes that do this reaction
  Ortholog: special genes (see glossary)

Other databases aside from KEGG:

(suggested by Dr. Abuja)

Biomodal DB:	http://www.ebi.ac.uk/biomodals/
Reactome „A curated knowledgebase of biological pathways“	http://www.reactome.org/
Brookhaven Protein Database for structure of enzymes	http://cds.dl.ac.uk/cds/pdb.html
EMBL „European Molecular Biology Laboratoy“	http://www.embl.org/
EMBO „European Molecular Biology Organization“	http://www.embo.org/
SMBL „Systems Biology Markup Language“	http://sbml.org/

Other important websites:

RCSB „Research Collaboratory for Structural Bioinformatics “	http://home.rcsb.org/
RCSB PDB Protein Data Base	http://www.rcsb.org/pdb/

Software we should look at:

- „Panther“ (Applied Biosoftware)
- google: „Pathway“ as well as „Pathway Heatmap“

Books:

David Fell, „Regulation of Metabolism“ („Kinetische Regulation“)

Homepage of David Fell:

http://www.brookes.ac.uk/bms/research/fell.html

Glossary:

Term	Description	Link
Substrate	a molecule upon which an enzyme acts. Enzymes catalyze chemical reactions involving the substrate(s).	http://en.wikipedia.org/wiki/Substrate_(biochemistry)
Kinase	a type of enzyme that transfers phosphate groups from high-energy donor molecules, such as ATP, to specific target molecules (substrates); the process is termed *phosphorylation*.	http://en.wikipedia.org/wiki/Kinase
Inhibitor	???	
Effektor	???	
Kinetische Regulation	???	
EC-Number	Enzyme Commission number (EC number)	http://en.wikipedia.org/wiki/EC_number
Homocysteine		http://en.wikipedia.org/wiki/Homocysteine
Ortholog	**Orthologs** are genes in different species which evolved from a common ancestral gene.	http://en.wikipedia.org/wiki/Ortholog
Proteomics		http://en.wikipedia.org/wiki/Proteomics
Metabolomics		http://en.wikipedia.org/wiki/Metabolomics
mRNA	Messenger RNA, transcribed DNA that leaves the cell core. A ribosom transcribed the mRNA to a Protein.	http://en.wikipedia.org/wiki/MRNA
Nucleotide	A,G,T,C (U=T) Adenosine Guanin Thymidine (DNA only) Cytidne Urdine (RNA only) = T	http://en.wikipedia.org/wiki/Nucleotide

Term	Description	Link
Codon	Group of three Nucleotides. A ribosom translates Codons of the mRNA to amino acids, that are linked to gethe to form a protein.	http://en.wikipedia.org/wiki/Codon
Amino acids	Amino acids build a Protein.	http://en.wikipedia.org/wiki/List_of_standard_amino_acids
Ribosom	Translates the mRNA to a Protein by reading Codons and translating them to amion acids, that are linked together forming the protein	http://en.wikipedia.org/wiki/Ribosome