the process in which the genetic code carried by mRNA directs the synthesis of proteins from amino acids.[66] Through constant modification of the protein, organisms accommodate radically different protein expression in different parts of the body and in different stages of the life cycle. Although amino acids can be predicted from nucleotide sequences, posttranslational modifications to proteins, in general, cannot.

Once synthesized on the ribosomes, proteins are subject to a multitude of modification steps. Because they are cleaved (thus eliminating signal sequences, transit or pro-peptides and initiator methionines), many simple chemical groups (for example acetyl, methyl, phosphoryl) as well as more complex molecules (such as sugars and lipids) can associate with them. Moreover, they can be internally or externally cross-linked (example: disulfide bonds). So far, over 200 different modifications have been described. The complexity due to all these modifications is compounded by the high level of diversity that alternative splicing[67] can produce at the level of the sequence. Many PTM have well described roles in signal transduction and the regulation of cellular processes. In contrast, other modifications are much less well documented but are also likely to play very important roles within the cell. Identifying the type and location of these proteins is a first step in understanding their regulatory potential. The complex study of posttranslational modifications is one major objective of proteomics and is referred to as 'PTM proteomics'. [68]

V. Role of Enzymes and their chemical activity

One important function performed by proteins is the ability to catalyze chemical reactions.[69] The biological catalysts were named enzymes.[70] Enzymes are usually specific to the reaction they catalyze and the chemical substances involved in the reaction. Many enzymes are composed of several proteins acting together as a unit.

66 Human Genome Project Information, Glossary of the Human Genome Project, available at http://www.ornl.gov/TechResources/Human_Genome/glossary/. Recent advances in mimicking PTMs are helping to elucidate the role of the modifications and are the subject of high expectations for future pharmaceuticals, Davis, Benjamin G., Mimicking Posttranslational Modifications of Proteins, 303 Science 2004, 480.

67 Alternative splicing of mRNA permits that many gene products with different functions are produced from a single coding sequence, see Brett, David/Pospisil, Heike et al., Alternative splicing and genome complexity, Nature Genetics 30, 2 (2001).

68 MacCoss, Michael J./Hayes McDonald; Saraf,/Saraf, Anita/Sadygov, Rovshan/Clark, Judy M./Tasto, Joseph J./Gould, Kathleen L./Wolters, Dirk/Washburn, Michael/Weiss, Avery/Clark, John I./Yates, John R., Shotgun Identification of Protein Modifications from Protein Complexes and Lens Tissue, 99 Proceedings of the National Academy of Science of the United States of America 2002, 7900, 7901.

69 Catalytic function was amongst the first biological roles recoginzed in proteins through the work of Eduard Buchner and Emil Fischer., Whitford, David, Proteins: Structure and Function, Chichester, West Sussex, U.K., 2005, 189.

70 The name derived from the Greek for 'in yeast' - 'en' 'zyme', Whitford, David, Proteins: Structure and Function, Chichester, West Sussex, U.K., 2005, 189.

Most parts of an enzyme have regulatory or structural functions. The catalyzed reaction takes place in only a small part of the enzyme called the active site. It is determined by a 3-D cleft formed by groups originating from different parts of the amino acid sequence. The active site, also described as a "binding pocket", takes up a relatively small part of the total volume of the enzyme.

Enzyme-substrate binding was historically described through the "lock-and-key model". The binding depends on the precisely defined arrangements of atoms in an active site. Complementary structural properties of the enzyme and substrate are responsible for this specificity. Most enzymes are highly selective with respect to the substrates that they bind, since their catalytic specificity depends partly on the specificity of binding. The active site contains the residues directly involved in the breaking and formation of bonds. These residues are referred to as catalytic groups. The catalytic power of enzymes originates from their ability to unite substrates in favorable orientations so as to promote the formation of the transitional states in enzyme-substrate complexes.[71] In 1958, Daniel Koshland introduced the "induced fit model", a modification of the lock and key model. It is based on the understanding that enzymes are flexible structures, in which the active side is continuously reshaped by its interaction with the substrate. Consequently, the amino acid side chains constituting the active side are molded into the exact position to start the catalytic function of the enzyme.[72] The enzyme-substrate complex according to the "induced fit model" is illustrated in Figure 5:[73]

71 Berg, Jeremy M./Tymoczko, John L./Stryer Lubert, Biochemistry, New York, NY, 2005, 200; Wikipedia, Enzymes, available at http://en.wikipedia.org/wiki/Image:Two_substrates_b.png, last checked on January 22, 2008.

72 Koshland D. E., Application of a Theory of Enzyme Specificity to Protein Synthesis, Proceedings of the National Academy of Science 44 (2), (1958), 98.

73 Based on Figure 5 provided by Wikipedia, Enzymes, available at: http://en.wikipedia.org/wiki/Imag e: Two_substrates_b.png, last checked on January 22, 2008.

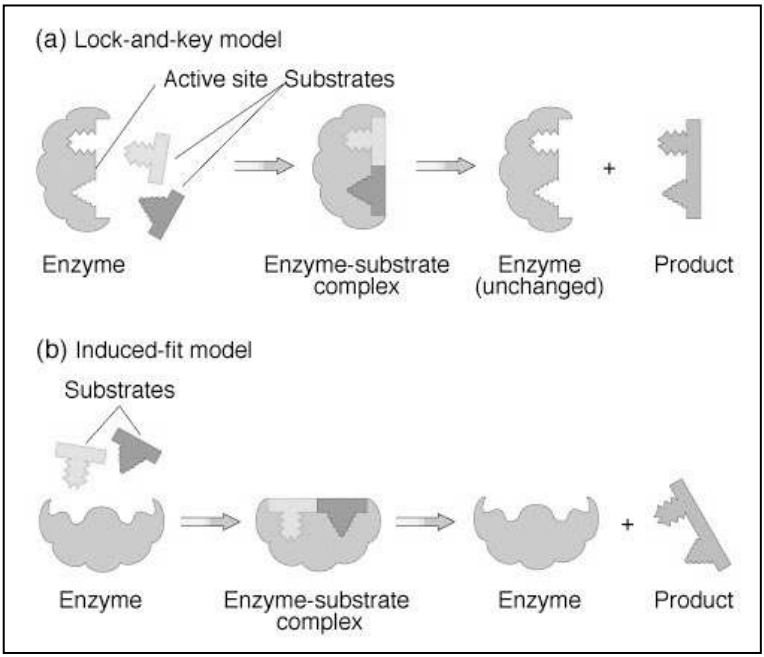

Figure 5: Enzyme-substrate complex

The primary function of enzymes is to enhance rates of reaction. Thereby they are compatible with the needs of the organism. Chemical reactions require a certain amount of activation energy to take place. Enzymes can increase the reaction speed by favoring or enabling a different reaction path with lower activation energy, making it easier for the reaction to occur. They can also serve to associate two or more reactions together, such that a thermodynamically favorable reaction can be used to "drive" a thermodynamically unfavorable one. The most common examples are enzymes that use the dephosphorylation of ATP to drive some otherwise unrelated chemical reactions.[74] Enzymes conduct up to several million catalytic reactions per second. In order to understand how enzymes work, a kinetic description of their activity is used. The maximum speed of an enzymatic reaction is determined by the so called "Michaelis-Menten equation". Here, the substrate concentration is increased until a constant rate of product formation is achieved. This is the maximum velocity (V_{max}) of the reaction catalyzed by the enzyme. In this state, all enzyme active sites are saturated with substrate. Since the substrate concentration at V_{max} cannot be precisely determined, enzymes are characterized by the substrate concentration at which

74 Figure available at http://lc.brooklyn.cuny.edu/smarttutor/core3_21/energy.html, last checked on January 22, 2008.

the rate of reaction is half its maximum. This substrate concentration is called the Michaelis-Menten constant (K_M).[75]

The activities of many enzymes can be anticipated by the binding of specific small molecules and ions. Inhibitor activity serves as a major control mechanism in biological systems. Many drugs act in this fashion. Inhibition by specific substrates can give a valuable insight into the mechanism of enzyme action. Enzyme inhibition occurs either reversibly or irreversibly. An irreversible inhibitor disconnects slowly from its target enzyme, because it binds tightly to it. Some important drugs are irreversible inhibitors. Penicillin acts by modifying the enzyme and thereby inhibiting the synthesis of bacterial cell walls, thus killing the bacteria. Aspirin has the ability to suppress the production of prostaglandins and thromboxanes by enzyme modification.[76]

Reversible inhibition, in contrast, describes the rapid dissociation of the enzyme-inhibitor complex. By competitive inhibition, an enzyme is not only bound by its natural substrate, but also by a further substrate ("inhibitor") which does not trigger the catalytic reaction. In this way, further binding of the natural substrate and its subsequent reaction are inhibited. The competitive inhibitor is similar to the substrate and associates to the active center of the protein. Thus, it prevents the substrate from binding to the same active site and diminishes the rate of catalysis by reducing the proportion of enzyme molecules bound to a substrate. Increasing substrate concentration leads to a decrease of competitive inhibitions.[77] Competitive inhibition[78] and non-competitive inhibition[79] are illustrated in Figures 6 and 7:

75 Berg, Jeremy M./Tymoczko, John L./Stryer Lubert, Biochemistry, New York, NY 2005, 200-203.

76 Prostaglandins are hormone-like substances, which are produced in the body and have various effects, including the transmission of pain information to the brain, modulation of the hypothalamic thermostat, and inflammation. Thromboxanes act to promote the aggregation of platelets that form blood clots. The effects of aspirin were discovered in 1971 by the British pharmacologist, John R. Vane. He was awarded a Nobel Prize in Physiology and Medicine for his research in 1982. Berg, Jeremy M./Tymoczko, John L./Stryer Lubert, Biochemistry, New York, NY 2005, 209.

77 For a detailed overview of the specific chemical procedures see Berg, Jeremy M./Tymoczko, John L./Stryer Lubert, Biochemistry, New York, NY, 2005, 209-219.

78 See: http://en.wikipedia.org/wiki/Enzyme, last checked on January 22, 2008.

79 The diagram showing the mechanism of non-competitive inhibition is taken from Wikipedia http://en.wikipedia.org/wiki/Enzyme, last checked on January 21, 2008.

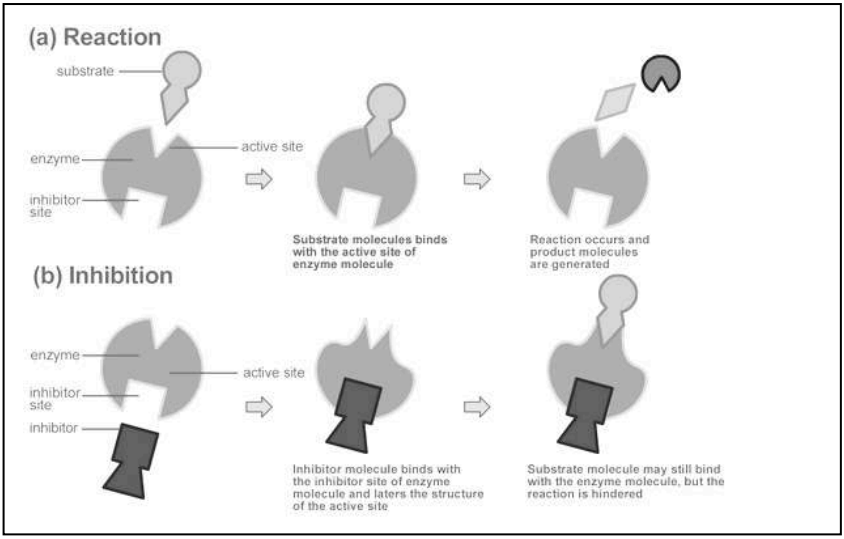

Figure 6: Competitive inhibition

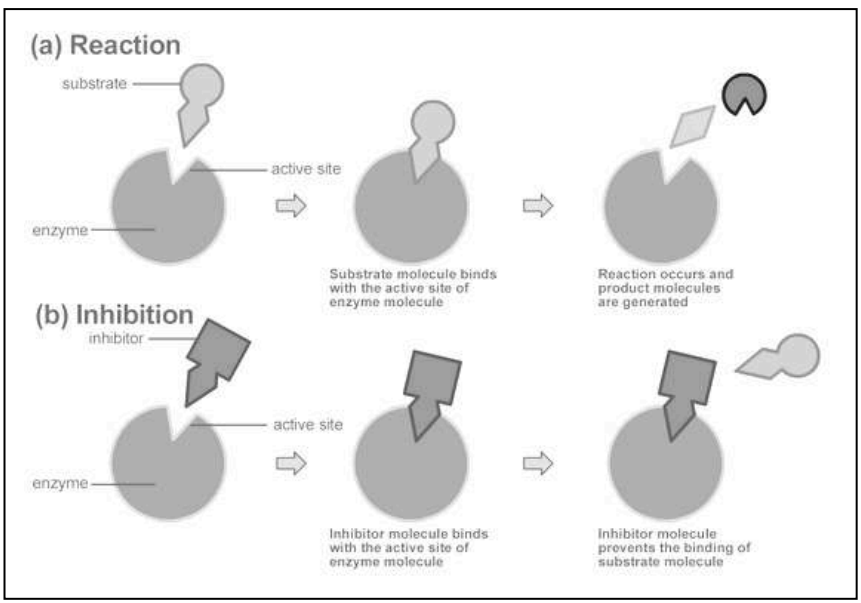

Figure 7: Non-competitive inhibition

C. Genetic coding of proteins[80]

In order to produce a protein, a cell requires information about the sequence in which the amino acid must be assembled.[81] The cell utilizes a long polymeric molecule, DNA (deoxyribonucleic acid), to store this information. The amino acid sequence of a protein is genetically determined by the sequences of bases in a DNA molecule. The subunits of the DNA are called nucleotides.[82] DNA encompasses four nucleotides that are distinguishable from the base regions of the molecule. The four bases are adenine, guanine, cytosine, and thymine (referred to as A, G, C and T). The sequence of these bases along the DNA molecule determines which amino acids will be inserted in sequence into the polypeptide chain of a protein. DNA is synthesized in extremely long strands (called chromosomes) encompassing information encoding for the sequence of many proteins. The region of DNA on the chromosome that determines the sequence of a single protein is called a gene.[83] The process in which the data in a gene is utilized to synthesize a new protein is called gene expression. To express a gene, a copy of the gene as a molecule of RNA (ribonucleic acid) is made. RNA is a molecule very similar to DNA. One difference, however, is that RNA contains a different sugar (ribose instead of deoxyribose). Furthermore, the base thymine (T) of DNA is replaced in RNA by the structurally similar base, uracil (U).[84] The process of making an RNA copy of DNA is called transcription.[85] The transcribed RNA copy contains sequences of A, U, C, and G having the same information as the sequence of A, T, C and G in the DNA. The RNA molecule, referred to as messenger RNA (mRNA), then progresses to a location in the cell where proteins are synthesized. The information encoding the sequence of amino acids in a protein (the "genetic code") is composed of serially reaching groups of three contiguous nucleotides. Each combination of three contiguous nucleotides, called a codon, determines one amino acid. The four bases A, G, C and U can be specified as triplets in 64 different ways, but there are only 20 amino acids to be translated. Thus,

80 Alberts, Bruce/Johnson, Alexander/Lewis, Julian, Molecular Biology of the Cell (4th ed.), New York 2002, 111-112.

81 CAFC decisions often provide a useful and clear illustration of the scientific background. The process of genetic coding and translation is explained in: In re O'Farrell 853 F.2d 894, 895-899 (Fed. Cir. 1988); for a detailed overview of the genetic coding of proteins see Vossius, Volker/Jaenichen, Hans-Rainer, Zur Patentierung biologischer Erfindungen nach Europäischem Patentübereinkommen und Deutschem Patentgesetz - Formulierung und Auslegung von Patentansprüchen, GRUR 1985, 821.

82 Alberts, Bruce/Johnson, Alexander./Lewis, Julian, Molecular Biology of the Cell, New York 2002, 98.

83 For a brief overview of the basics of genetics, see also Kraßer, Rudolf, Patentrecht: ein Lehr- und Handbuch zum deutschen Patent- und Gebrauchsmusterrecht, europäischen und internationalen Patentrecht, 5. Aufl., München 2004, 222.

84 Alberts, Bruce/Johnson, Alexander./Lewis, Julian, Molecular Biology of the Cell, New York 2002, 104-105.

85 Alberts, Bruce/Johnson, Alexander./Lewis, Julian,Molecular Biology of the Cell, New York 2002, 104.