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Phase II-(I) Glucoronic acid conjugation, sulphate conjugation, amino acid conjugation,

Phase II-(II) glutathione conjugation, acetyl and methyl conjugation.

Phase II-(III) Physicochemical and stereochemical aspects in relation to biological activity, drug receptor interactions.

Hello friends I am Dr. Ankur Vaidya, Asst. Professor from Pharmacy College, UPRIMS & R, Saifai, Etawah (U.P.). Today we are going to deal with an interesting and learning episode of B. PHARM on the important title Phase II-Drug metabolism. So Let's start our episode while taking a look at what we are going to learn today.

We will begin with the genral introduction of the topic followed by Phase-II drug-conjugation.

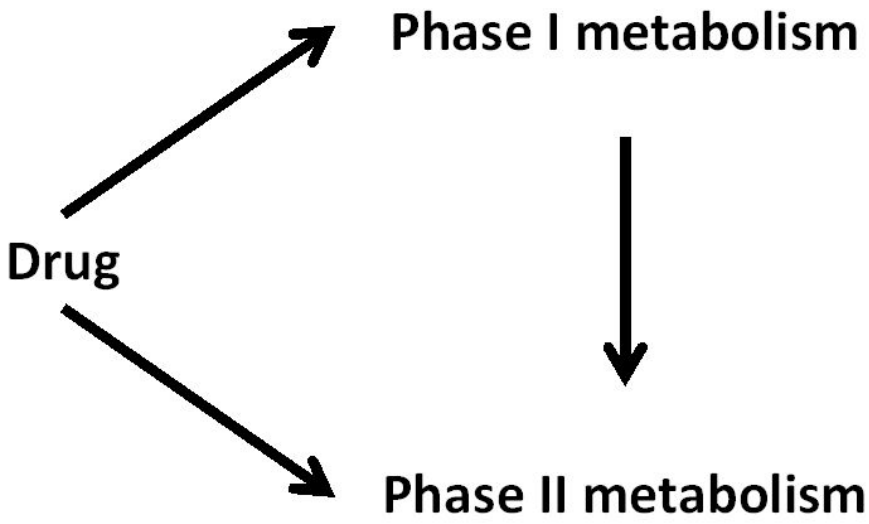
After discussing it our lecture will proceed further to

Physicochemical and stereochemical aspects in relation to biological activity and drug receptor interactions..

So lets start our today's lecture with introduction of this topic …

Introduction

Biotransformation of substances or xenobiotics is divided into two phases i.e. Phase I and Phase II. Phase I reactions include modification of a parent compound to more polar metabolite(s) by unmasking or de novo formation of functional groups (e.g. -OH, -NH_2, -SH). The phase I reactions are chiefly catalyze by the enzymes cytochromes P450 (CYPs) mediated hydroxylations and hence acting as monooxygenases, dioxygenases and hydrolases. These cytochromes P450 enzymes constitute a superfamily of heme enzymes responsible for the metabolism of xenobiotics and endobiotics.



In spite of Phase I biotransformation, the phase II biotransformation is to perform conjugating reactions by means of glucuronidation, sulfation, methylation, acetylation, glutathione and amino acid conjugation. The respective conjugates are more hydrophilic than the parent compounds. The enzymes catalyzing Phase II reactions include: UDP-glucuronosyltransferases (UGTs), sulfotransferases (SULTs), N-acetyltransferases (NATs), glutathione S-transferases (GSTs) and various methyltransferases (mainly thiopurine S-methyl transferase (TPMT) and catechol O-methyl transferase (COMT)). Conjugation reactions catalyzed by the superfamily of these enzymes serve as the most important detoxification pathway for broad spectrum of drugs and dietary chemicals.

These produced conjugates in Phase II reactions may also play an essential role in the toxicity of many chemicals due to the formation of toxic metabolites (carcinogenic conjugate metabolite product of benzylic alcohols, polycyclic aromatic hydrocarbons, aromatic hydroxylamines, hydroxamic acid and nitroalkanes by sulphotransferases).

Phase II conjugation reactions usually involve metabolite activation by a high–energy intermediate and lead to classified into two general categories:

Category I (e.g., glucuronidation and sulfonation), in which an activated conjugating agent combines with substrate to yield the conjugated product.

Category II (e.g., amino acid conjugation), in which the substrate is activated and then combined with an amino acid to yield a conjugated product.

In the present topic, we will focus on the most important conjugation reactions, namely glucoronic acid conjugation, sulphate conjugation, amino acid conjugation, glutathione conjugation, acetyl and methyl conjugation. In spite of these conjugation reactions we will also discuss about physicochemical and stereochemical aspects in relation to biological activity and drug receptor interactions.

Module 1

Glucuronic acid conjugation

Glucuronidation is the most important conjugation of Phase II reaction. This reaction is catalyzed by UDP–glucuronosyltransferases (UGTs) enzyme which belongs among the key enzymes of metabolism of various exogenous as well as endogenous compounds. In this reaction a huge supply of glucuronic acid is required. In humans, about 40–70% of clinically used drugs are subjected to glucuronidation. A number of drugs are found to be a substrate for UDP–glucuronosyltransferases (UGTs) enzyme. Drugs carrying nucleophilic O-, N-, S-, or C-atom are the primary target for UGTs mediates glucuronide conjugation. Glucuronidation lead to formation of a chemical bond between a nucleophilic O-, N-, S-, or C-atom with uridine- 5'-diphospho-α-D-glucuronic acid (UDPGA). These newly formed β–D–glucuronides exhibit increased water–solubility or very polar compound lead to enhance urinary or bile excretion.

All UDPGA are capable of forming O-linked glucuronides conjugate with the drugs having aliphatic alcohols, phenols and carboxylic acids in the presence of UGT enzymes.

Around 19 human UGTs have been reported to conjugate different nitrogen-containing compounds, and forming N-glucuronides. N-Glucuronidation exhibits marked differences across species. As an example, the ability to form quaternary ammonium glucuronides from tertiary amines is a reaction largely, but not completely, restricted to humans. Among the all the known UGTs, UGT1A4 has been considered the enzyme "specializing" in N-glucuronidation.

S-glucuronide conjugate was reported for sulfur containing drugs. Benzothiazole-2-sulfonamide was found to be first metabolized to its corresponding mercapturic acid, which subsequently formed the *S*-glucuronic acid conjugate.

A carbon atom is not considered to be a nucleophilic atom; however, xenobiotics containing strongly acidic carbons or enolic acids are known to form *C*-linked glucuronides. A number of *C*-glucuronide conjugates have been reported for drugs

carrying carbon atom. Although it may be speculated that *C*-glucuronides may be formed at acidic carbon atoms, the mechanism of formation of *C*-glucuronides has not yet been fully understood.

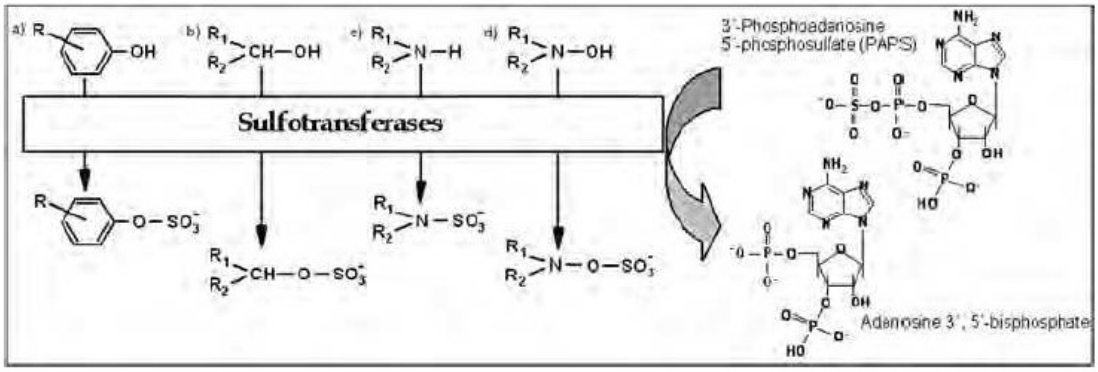
These UGTs are also involved in the regulation of several active endogenous compounds such as bile acids or hydroxysteroids due to their inactivation via glucuronidation. Similar to cytochromes P450 enzymes these UGTs are also membrane–bound enzymes with subcellular localization in the endoplasmic reticulum (ER) but the active site of these enzymes is embedded in the lumenal part of ER (distinct from cytochromes P450). UGT gene superfamily consist of 117 members while in humans, four UGT families have been identified: UGT1, UGT2 (divided into subfamilies, 2A and 2B), UGT3 and UGT8. First two families, UGT1 and UGT2, use UDPGA to glucuronidate endo- and xenobiotics. This is not valid for the UGT8 and UGT3 family. The UGT8 enzyme has a biosynthetic role in the nervous system and uses the UDP-galactose to galactosidate ceramides. The function of the UGT3 family was unclear for a long time.

Module 2

Sulphate Conjugation

Sulphate conjugation (or sulfonation) is an important pathway in the metabolism of numerous xenobiotics. The sulfonation reaction was first recognized by Baumann in 1876 by detecting phenyl sulfate in the urine of a patient who had been administered phenol. Sulfate conjugation gives a polar and ionized conjugate by means of the esterification of a hydroxyl group with sulfate ion (transferred from 3'-phosphoadenosine-5'-phosphosulfate or PAPS). Sulfation sometimes gives rise to reactive intermediates that may undergo further reactions to yield electrophilic metabolites. In the case of 2-acetaminofluorene, the *O*- sulfate moiety is a facile leaving group, and this cleavage produces nitrenium ions, which act as alkylating agents for DNA. The reaction is catalyzed by a family of enzymes called sulfotransferases (SULTs) which catalyze the transfer of sulfonate (SO^{3-}) from the universal sulfonate donor 3'–phosphoadenosine 5'–phosphosulfate (PAPS) to the hydroxyl or amino group of an acceptor molecule (Figure). Transfer of sulfonate moiety to the molecule leads to formation of a water–soluble compound which is then easily eliminated from the body. PAPS is a common donor of sulfonate moiety in sulfonation reactions and has been shown to by synthesized by almost

all tissues in mammals from inorganic sulfate. Depletion of PAPS due to lack of inorganic sulfate or due to genetic defects of enzymes participating in PAPS synthesis may lead to reducing of sulfonation capacity which could affect the metabolism of xenobiotics or disrupt the equilibrium between synthesis and degradation of active endogenous compounds.

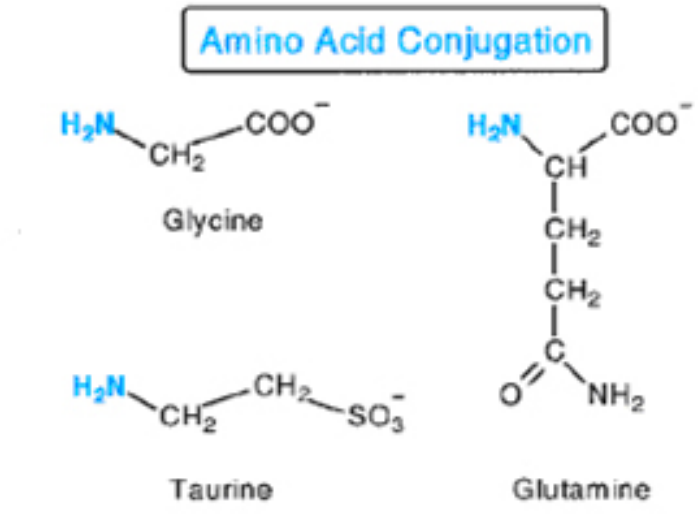


Sulfotransferases (SULTs) enzymes are categorized into two categories depending on their location or presence. The first group includes membrane–bound enzymes having no intrinsic xenobiotic–metabolising activity. These enzymes are found in the Golgi apparatus and they are involved in metabolism of endogenous peptides, proteins, glycosaminoglycans, and lipids.

The second category of sulfotransferases (SULTs) enzymes are cytosolic sulfotransferases which plays a major role in conjugation of a broad spectrum of xenobiotics including environmental chemicals, natural compounds, drugs as well as endogenous compounds such as steroid hormones iodothyronines, catecholamines, eicosanoids, retinol or vitamin D.

Amino acid conjugation

The amino acids include glycine, taurine, glutamine, arginine, and ornithine are commonly used in amino acid conjugation. Amino acids are found in protein-rich foods and help to excrete many toxic chemicals, called xenobiotics, from the environment.



Two principal amino acids conjugation pathways have been reported. The first conjugation pathway involves activation of the xenobiotic by conjugation with CoA, which produces an acyl-CoA thioether that reacts with the amino group of an amino acid to form an amide linkage. The second pathway involves conjugation of xenobiotics containing an aromatic hydroxylamine (N-hydroxy aromatic amine) with the carboxylic acid group of amino acids as serine and proline. This pathway involves activation of an amino acid by aminoacyl-tRNA-synthetase, which reacts with an aromatic hydroxylamine to form a reactive N-ester.

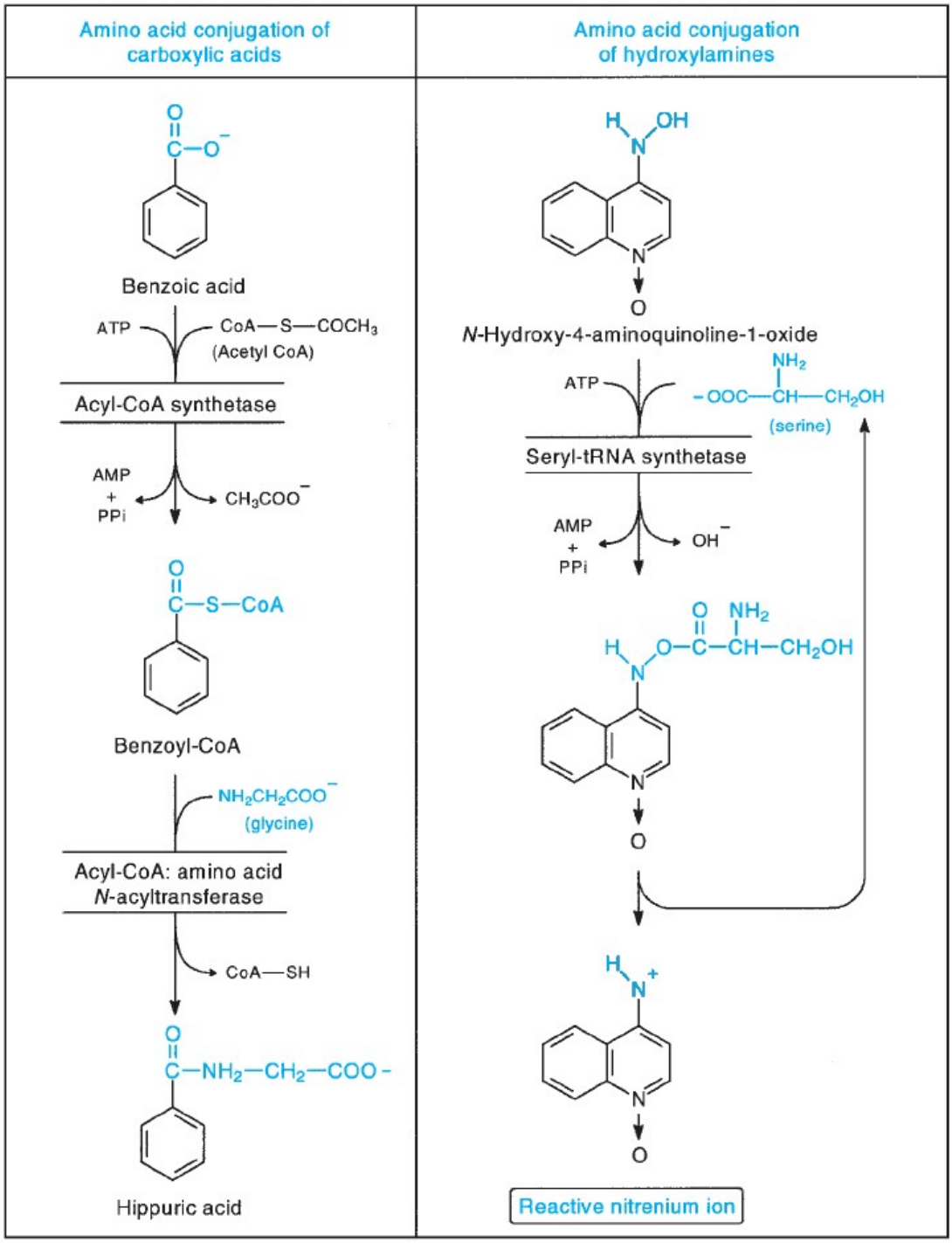


Figure 6-54. Conjugation of xenobiotics with amino acids.

Amino acid conjugation of exogenous carboxylic acids is a special form of acetylation and leads to amide bond formation. The most common amino acid in such reactions is glycine, and its prototypical substrate is benzoic acid, more precisely its benzoyl–CoA

cofactor. Bile acids are also conjugated by a similar sequence of reactions involving a microsomal cholyl–CoA synthetase and a cytosolic enzyme bile acid–CoA: amino acid *N*–acyltransferase. Xenobiotic carboxylic acids and amino acid conjugation involves enzymes located principally in the mitochondria of liver and kidney while conjugation of bile acids is extramitochondrial, involving enzymes located in the endoplasmic reticulum and peroxisomes.

Glycine conjugates are the most common amino acid conjugates in animals and conjugation with L-glutamine is most common in humans and other primates.

Steric effect may affect the conjugation of xenobiotics to amino acids. For example diphenylacetic acid cannot be conjugated with an amino acid due to steric effect.

Bile acids are endogenous substrates for glycine and taurine conjugation. The activation of bile acids to an acyl-CoA thioester is catalyzed by a microsomal enzyme, cholyl-CoA synthetase, while conjugation of glycine or taurine is catalyzed by a single cytosolic enzyme, bile acid-CoA:amino acid N-acyltransferase. In contrast, the activation of xenobiotics occurs mainly in mitochondria, which appear to contain multiple acyl-CoA synthetases. An important difference between the amino acid conjugates of xenobiotics and bile acids is their route of elimination: Bile acids are secreted into bile whereas amino acid conjugates of xenobiotics are eliminated primarily in urine. The addition of an endogenous amino acid to xenobiotics may facilitate this elimination by increasing their ability to interact with the tubular organic anion transport system in the kidney.

Module 3

Glutathione conjugation

Glutathione conjugation is the addition of glutathione by glutathione-S-transferase (GST). Glutathione is a co-substrate tripeptide (Gly-Cys-Glu) present within the body for removal of potentially toxic electrophilic compounds. Many drugs metabolized in phase I are, strong electrophiles, react with glutathione to form non- toxic conjugates. Glutathione conjugates may be excreted directly in urine or bile, but are usually metabolized further. Glutathione S–transferases (GSTs) constitute an important intracellular defence against oxidative stress and they appear to be involved in synthesis and metabolism of several derivatives of arachidonic acid and steroids. In general, these glutathione S– transferases enzymes catalyze a nucleophilic attack of reduced glutathione

on lipophilic compounds containing an electrophilic atom (C–, N– or S–). In spite of these nucleophilic substitutions, transferases also account for Michael additions, isomerations, and hydroxyperoxide reductions. In most cases, more polar glutathione conjugates are eliminated into the bile or are subsequently subjected to other metabolic steps eventually leading to formation of mercapturic acids.

In glutathione conjugate reaction a thioether linkage occur with drug or xenobiotic to the cysteine moiety of the tripeptide. Usually, all GST substrates contain an electrophilic atom and are hydrophobic, which will associate with cellular proteins. Since the concentration of glutathione in cells is usually very high, typically ~7 μmol/g of liver, or in the 10 mM range, many drugs and xenobiotics can react nonenzymatically with glutathione.

GSTs have been divided into two subfamilies on the basis of their locality/presence:

(i) cytosolic GSTs and

(ii) microsomal GSTs

The cytosolic and microsomal GSTs have been differentiate on the basis of their selection of substrates for conjugation; the cytosolic GSTs have more importance in the metabolism of drugs and xenobiotics, whereas the microsomal GSTs are important in the endogenous metabolism of leukotrienes and prostaglandins. The cytosolic GSTs are divided into seven classes termed as alpha (GSTA1 and 2), mu (GSTM1 through 5), omega (GSTO1), pi (GSTP1), sigma (GSTS1), theta (GSTT1 and 2), and zeta (GSTZ1). GSTs of alpha and mu classes can form heterodimers, allowing for a large number of active transferases to form. The cytosolic forms of GST catalyze conjugation, reduction, and isomerization reactions.

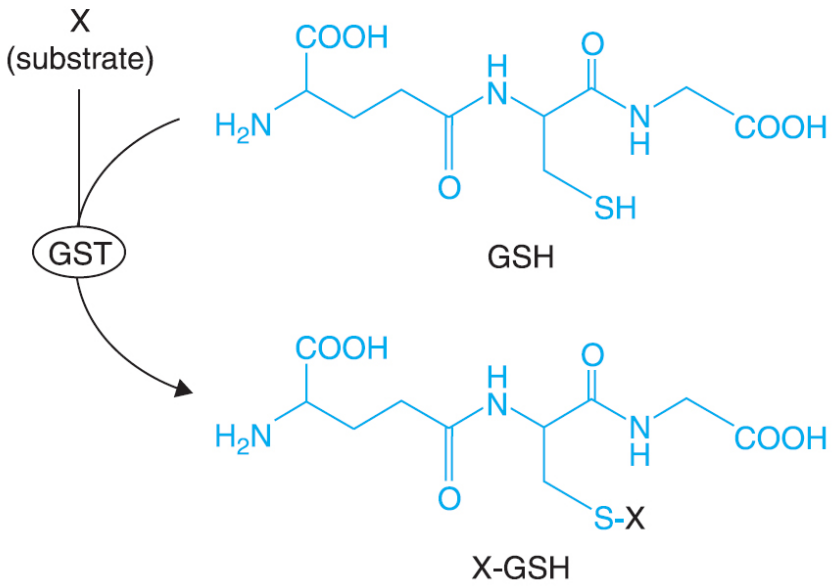


Figure. *Glutathione as a co-substrate in the conjugation of a drug or xenobiotic (X) by glutathione-S-transferase (GST).*

Acetyl Conjugation

Acetyl conjugation or acetylation is a very common Phase II metabolic reaction, which occurs with amino, hydroxyl or sulfhydryl groups. The acetyl group is transferred from acetyl-Coenzyme A (a cofactor) to a primary amino function accepting chemical group, by enzyme acetyltransferases. Drugs and other foreign compounds that are acetylated are either aromatic amines or hydrazines, which are converted to aromatic amides and aromatic hydrazides respectively. Usually three general reactions of acetylation have been reported, namely N–, O–, and N,O–acetylations. The first N–acetylation observe in aromatic amine, is recognized as a major detoxification pathway in arylamine metabolism in experimental animals and humans. However, O– and N, O–acetylations occur in alternative metabolic pathways following activation by N–hydroxylation. The resulting N–acetoxyarylamines are highly unstable, spontaneously forming arylnitrenium ions that bind to DNA and ultimately lead to mutagenesis and carcinogenesis. N–acetyltransferase isoenzymes (NATs) are cytosolic enzymes are of two types, N–acetyltransferase 1 (NAT1) and 2 (NAT2). Enzyme NAT1 has been found in adult liver, bladder, digestive

system, blood cells, placenta, skin, skeletal muscles, gingiva, mammary tissue, prostate, and lung while NAT2 protein is present mainly in the liver and in intestine. A notable difference between these two isoenzymes is the presence during birth. NAT1 activity in fetal and neonatal tissue, such as lungs, kidneys, and adrenal glands while NAT2 is not evident until about 12 months after birth.

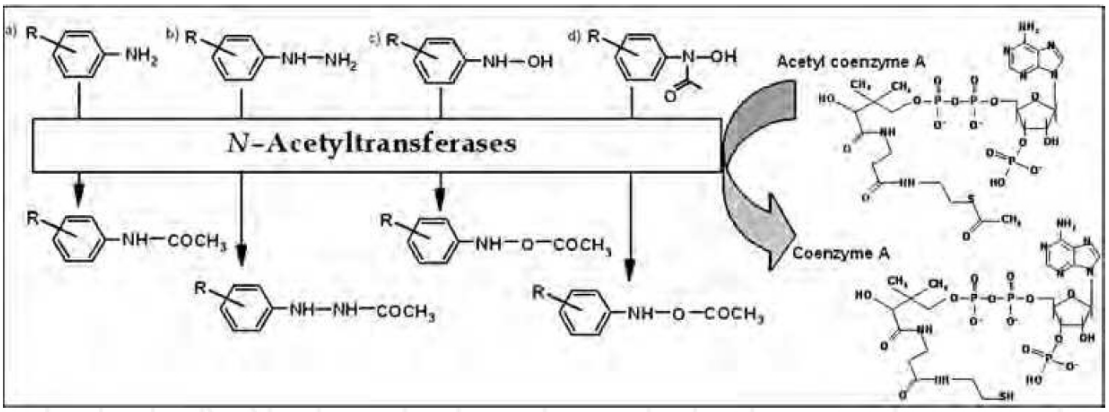


Fig. Reactions catalysed by *N*-acetyltransferases. (a,b) *N*-acetylation of arylamine and arylhydrazine, (c) *O*-acetylation of *N*-arylhydroxylamine, (d) *N, O*-acetyltransfer of an *N*-hydroxamic acid. These reactions use acetyl-coenzyme A as acetyl donor.

Module 4

Methyl Conjugation

Methylation is a common but relatively minor conjugative pathway in drug metabolism. Methylation differs from other phase II reactions in that, it reduces the water solubility or polarity of xenobiotics and masks functional groups that might otherwise be conjugated by other phase II enzymes. Exceptions of this rulse include *N*-methylation of pyridine-containing xenobiotics, such as nicotine, which produces quaternary ammonium ions that are water-soluble and readily excreted and the *S*-methylation of thioethers to form positively charged sulfonium ions, a reaction catalyzed by thioether methyltransferase (TEMT) lead to produce hydrophilic metabolite.

Methylation conjugation involved in the metabolism of small endogenous compounds such as neurotransmitters as well as metabolism of macromolecules includes nucleic acids and in the biotransformation of certain drugs. A large number of both endogenous and exogenous compounds can undergo *N*–, *O*–, *S*– and arsenic–methylation during their metabolism and the functional groups involved in methylation reactions are phenols,

catechols, aliphatic and aromatic amines, *N*-heterocyclics, and sulfhydryl-containing compounds. The co–factor involved in methylation is *S*–adenosylmethionine (SAM), which is primarily formed by the condensation of ATP and L–methionine.

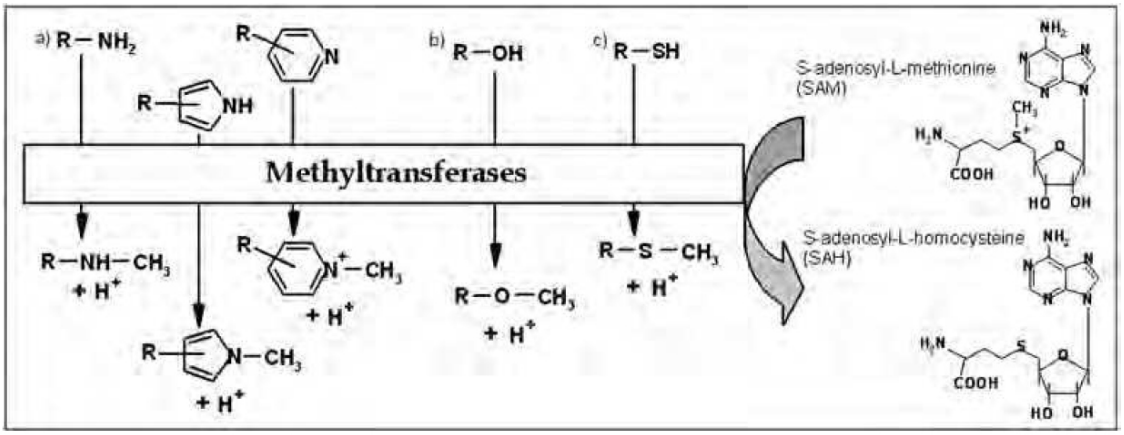


Fig. Methylation reactions catalyzed by methyltransferases.

Physicochemical and stereochemical Properties for Drug Action

Drug action results from the interaction of drug molecules with either normal or abnormal physiological processes.

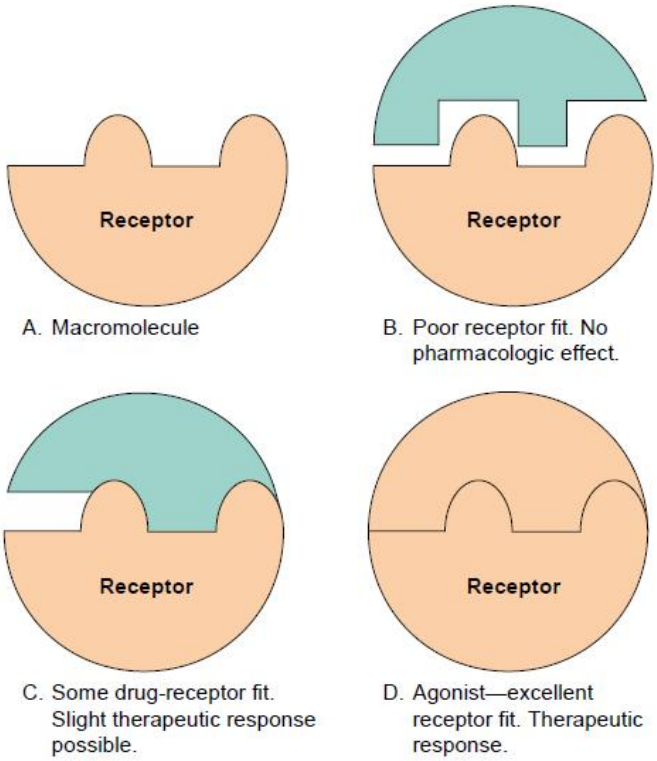
Drugs normally interact with targets or receptors (which they are proteins, enzymes, cell lipids, or pieces of or DNA RNA) and produce their action. The ability of a drug to elicit a pharmacologic / therapeutic effect is related to the influence of its various physical and chemical (physicochemical) properties. Number of factors of drug including water solubility, lipid solubility, partition coefficient, acid-base properties, steric factors and stereochemistry can affect both pharmacokinetic and pharmacodynamic properties. By integrating the knowledge of physicochemical properties, we are able to make informed decisions regarding formulation development and the ultimate performance of the drug product. This knowledge also provides valuable insight into process development and manufacturing. The pharmacokinetic properties include absorption, distribution, metabolism and excretion. Pharmacodynamic properties represent drug-receptor interaction lead to produce activity and toxicity.

Stereochemical features like electronic distribution, stereochemical properties, and surface properties of drug play significant role in the mediation of activity. It is important to note that drug's with different diastereomers produce different pharmacologic effects, as is the case with labetalol. Labetalol a sympatholytic drug is a mixture of four stereoisomers. One isomer is an α1 antagonist (like prazosin), another is a nonselective β antagonist with partial agonist activity (like pindolol), and the other two isomers are inactive. This example clearly shows that stereochemistry play an important role in pharmacologic action.

Module 5

Drug–Receptor Interactions

Receptors are macromolecules located on the cell surface membrane or within the cytoplasm. These receptors involved in chemical signaling between and within cells and directly or indirectly regulate cellular biochemical processes. Molecules or drugs interact with receptors which have protein-like properties and specific three dimensional shapes. Interaction may increase or decrease a particular cell function. Each ligand may interact with multiple receptor subtypes, but the interaction is relative selective. Selectivity is the degree to which a drug acts on a given site relative to other sites; selectivity relates largely to physicochemical binding of the drug to cellular receptors.



A. Macromolecule

B. Poor receptor fit. No pharmacologic effect.

C. Some drug-receptor fit. Slight therapeutic response possible.

D. Agonist—excellent receptor fit. Therapeutic response.

For drug-receptor interaction a specific chemical structure is required for the receptor site and a complementary drug structure. Slight changes in the molecular structure of the drug may drastically change specificity. A minimum three point attachment of a drug to a receptor site is required. Usually reversible-temporary drug- receptor bond formation occur. Covalent bonds would be very tight and practically irreversible and rather rare except in a rather toxic situation. Ionic bonds, hydrogen bonds, wander-wall interactions hydrophobic bonds and polar-polar interactions are most common drug-receptor interaction. Steric effect may alter the drug-receptor interaction. The pharmacologic effects depend on the duration of time that the drug-receptor complex persists. The lifetime of the drug-receptor complex is affected by dynamic processes (conformation changes) that control the rate of drug association and dissociation from the target. Fcators including drugs, aging, genetic mutations and disorders can either increase (up-regulate) or decrease (down-regulate) the number and binding affinity of receptors.

Summary

Phase II biotransformation reactions is also called 'conjugation reactions' generally serve as a detoxifying step in drug metabolism. Phase II drug metabolizing enzymes play an important role in biotransformation of endogenous compounds and xenobiotics to more easily excretable forms as well as in the metabolic inactivation of pharmacologically active compounds. Phase II drug metabolising enzymes are mainly transferases and number of enzymes including UDP-glucuronosyltransferases, sulfotransferases, N-acetyltransferases, glutathione S-transferases and methyltransferases catalyze phase II metabolism. A number of physiochemical and stereochemical properties of drug alter the pharmacological activity.

Assignment

Q 1. Define the term biotransformation.

Q 2. What is phase II biotransformation? Explain it.

Q 3. Classify phase II biotransformation.

Q 4. Explain sulphate conjugation biotransformation with suitable examples.

Q 5. Explain amino acids conjugation biotransformation with suitable examples.

Q 6. Explain glutathione conjugation biotransformation with suitable examples.

Q 7. Write an exhaustive note on physicochemical and stereochemical properties for drug action.

Q 8. Write an exhaustive note on drug receptor interaction.

LOR's with answer

Q 1. Define the term biotransformation.
Ans: Biotransformation is a major mechanism for drug elimination by means of production of metabolites that are more polar than the parent drug. Biotransformation usually terminates the pharmacologic action of the parent drug

Q 2. Explain Phase II biotransformation.
Ans: Phase II biotransformation is to perform conjugating reactions by means of glucuronidation, sulfation, methylation, acetylation, glutathione and amino acid conjugation. The respective conjugates are more hydrophilic than the parent compounds.

Q 3. What is Glucuronic acid conjugation? Explain it.

Ans: Glucuronidation is the most important conjugation of Phase II reaction. In humans, about 40–70% of clinically used drugs are subjected to glucuronidation. A number of drugs are found to be a substrate for UDP–glucuronosyltransferases (UGTs) enzyme. Glucuronidation lead to formation of a chemical bond between a nucleophilic O-, N-, S-, or C-atom with uridine- 5'-diphospho-α-D-glucuronic acid (UDPGA). These newly formed β–D–glucuronides exhibit increased water–solubility or very polar compound lead to enhance urinary or bile excretion.

FAQ's with answer

Q 1. Explain sulphate conjugation of Phase II biotransformation.

Ans: Sulphate conjugation (or sulfonation) is an important pathway in the metabolism of numerous xenobiotics. The sulfonation reaction was first recognized by Baumann in 1876. Sulfate conjugation gives a polar and ionized conjugate by means of the esterification of a hydroxyl group with sulfate ion. The reaction is catalyzed by a family of enzymes called sulfotransferases (SULTs) which catalyze the transfer of sulfonate (SO^{3-}) from the universal sulfonate donor 3'–phosphoadenosine 5'– phosphosulfate (PAPS) to the hydroxyl or amino group of an acceptor molecule.

Q 2. Define amino acid conjugation, with its various pathway.

The amino acids include glycine, taurine, glutamine, arginine, and ornithine are commonly used in amino acid conjugation. Two principal amino acids conjugation pathways have been reported. The first conjugation pathway involves activation of the xenobiotic by conjugation with CoA, which produces an acyl-CoA thioether that reacts with the amino group of an amino acid to form an amide linkage. The second pathway involves conjugation of xenobiotics containing an aromatic hydroxylamine (N-hydroxy aromatic amine) with the carboxylic acid group of amino acids as serine and proline.

Quiz with answer

1. Biotransformation of substances or xenobiotics is divided into (a)-
    a. Two phase
    b. Three phase
    c. Four phase

d. None of these

2. The enzymes catalyzing Phase II reactions include (d)-
   a. UDP-glucuronosyltransferases (UGTs)
   b. Sulfotransferases (SULTs)
   c. N-acetyltransferases (NATs)
   d. All of above
3. Phase II conjugation reactions are classified into (a)-
   a. Two phase
   b. Three phase
   c. Four phase
   d. None of these
4. Glucuronic acid conjugation is catalyze by enzyme (a)-
   a. UDP-glucuronosyltransferases (UGTs)
   b. Sulfotransferases (SULTs)
   c. N-acetyltransferases (NATs)
   d. All of above
5. Among the all the known UGTs, the enzyme "specializing" in N-glucuronidation is (c)-
   a. UGT2 has been considered
   b. UGT1A2 has been considered
   c. UGT1A4 has been considered
   d. UGT1B4 has been considered
6. In humans, most common amino acid conjugation is (c) -
   a. Glycine
   b. Taurine
   c. L-Glutamine
   d. Arginine
7. Glutathione conjugates may be excreted directly in (c)–
   a. Urine
   b. Bile
   c. Both a and b
   d. None of these
8. The co–factor involved in methy-conjugation is (a)–
   a. *S*–adenosylmethionine (SAM)
   b. N-acetyltransferases (NATs)
   c. Sulfotransferases (SULTs)
   d. UDP-glucuronosyltransferases (UGTs)
9. Labetalol a sympatholytic drug is a mixture of ……stereoisomers (c)-
   a. Two
   b. Three
   c. Four

d. None of these

10. Diphenylacetic acid cannot be conjugated with an amino acid possibly due to (c)-
    a. Polarity of Diphenylacetic acid
    b. Presence of +ve charge on Diphenylacetic acid
    c. Steric effect
    d. None of these

Glossary

Acetylation Acetylation is a very common Phase II metabolic reaction, which occurs with amino, hydroxyl or sulfhydryl groups. The acetyl group is transferred from acetyl-Coenzyme A (a cofactor) to a primary amino function accepting chemical group, by enzyme acetyltransferases.

Glucuronidation Glucuronidation is the most important conjugation of Phase II reaction. This reaction is catalyzed by UDP–glucuronosyltransferases (UGTs) enzyme which belongs among the key enzymes of metabolism of various exogenous as well as endogenous compounds. In this reaction a huge supply of glucuronic acid is required.

Methylation Methylation is a common but relatively minor conjugative pathway in drug metabolism. Methylation differs from other phase II reactions in that, it reduces the water solubility or polarity of xenobiotics and masks functional groups that might otherwise be conjugated by other phase II enzymes.

Sulfonation Sulfonation is an important pathway in the metabolism of numerous xenobiotics. Sulfate conjugation gives a polar and ionized conjugate by means of the esterification of a hydroxyl group with sulfate ion (transferred from 3'-phosphoadenosine-5'-phosphosulfate or PAPS).

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