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HEXOSE MONOPHOSPHATE AND OTHER PATHWAYS TO DRUG METABOLISM

***Ohadoma S. C., Lawal B. A. S., Udoh Ekaette S. and Eban L. K.**

Department of Pharmacology, College of Medical Sciences, University of Calabar, Nigeria.

***Corresponding Author: Ohadoma S. C.**
Department of Pharmacology, College of Medical Sciences, University of Calabar, Nigeria.

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INTRODUCTION

The hexose monophosphate pathway also called the pentose phosphate pathway occurs in the cytosol of a cell, and includes two irreversible oxidative reactions, followed by a seriesof reversible sugar–phosphate interconversions (Larry, 2015). No adenosine triphosphate(ATP) is directly consumed or produced in this cycle, instead two reduced nicotinamide adenine dinucleotide phosphates(NADPHs) are produced for each glucose 6-phosphate molecule entering the oxidativepart of the pathway (Larry, 2015). The rate and direction of the reversible reactions of the pentosephosphate pathway are determined by the supply of and demand for intermediates of the cycle(Chayen et al.,1986; LeBlanc, 2008; Larry, 2015). The pathway provides a major portion of the body's NADPH, which functions as biochemical reductant. It also produces ribose 5-phosphate, required for the biosynthesis of nucleotides, and provides a mechanism for the metabolic use of 5-carbonsugars obtained from the diet or the degradation of structural carbohydrates (Larry, 2015).The oxidative portion of the pentose phosphate pathway consists of three reactions that lead to the formation of ribulose 5-phosphate, CO2, and two molecules of NADPH for each molecule of glucose 6-phosphate oxidized (Lehninger *et al*., 2008; Larry 2015). This portion of the pathway isparticularly important in the liver, lactating mammary glands, and adipose tissue, whichare active in the NADPH-dependent biosynthesis of fatty acids.Also in the testes,ovaries, placenta, and adrenal cortex, which are active in the NADPH-dependentbiosynthesis of steroid hormones; and in red blood cells, whichrequire NADPH to keep glutathione reduced (Larry, 2015).Thus NADPH production is regulated by the relative levels of NADP and NADPH, which alter the activity of glucose-6-phosphate dehydrogenase enzyme (Lehninger et al., 2008). All of the HMP enzymes after the 6-phosphogluconate dehydrogenase step are fully reversible. These enzymes allow for the synthesis of ribose-5-phosphate from the glycolytic intermediates fructose-6-phosphate and glyceraldehyde-3-phosphate, and the conversion of ribose-5-phosphate into glycolytic intermediates (Lehninger et al., 2008; Larry, 2015). NADPH (and therefore the hexose monophosphate pathway) is part of the mechanism that red blood cells use to prevent oxygen-mediated damage to their membranes and proteins, and to maintain haemoglobin in the oxidation state required for oxygen transport (Larry, 2015). Experimental studies by Chayen et al., 1986, showed thatHMP activity is also high in mature RBCs, the lens and cornea, all of which need NADPH for reduced glutathione production; which in turn protects them from oxidative damage. An erythrocytic deficiency in Glucose-6-Phosphate deficiency can cause an increase in the concentration of methemoglobin, which is a decrease in the amount of reduced glutathione, an increase in hydrogen peroxide (H2O2), and increased fragility of red blood cell membranes (LeBlanc , 2008; Larry, 2015). The net result is **haemolysis,** which can be exacerbated when patients are given excessive amounts of oxidizing agent drugs such as Aspirin or sulfonamide antibiotics. On the other hand, a relative deficiency of Glucose-6-Phosphate in erythrocytes may protect some persons from certain parasitic infestations (e.g., falciparum malaria), since the parasites that cause this disease require NADPH for its survival cycle (LeBlanc,2008; Larry, 2015).

KEYWORDS: Hexose Monophosphate, Xenobiotic, phase II reactions, nonmicrosomal oxidation, induction/inhibition drug Metabolism.

A. Oxidative Phase Of Hmp Pathway

As earlier mentioned, oxidative phases of HMP are irreversible reactionsthat consists of three reactions that leads to the formation of Ribose 5-phosphate, CO2, and two molecules of NADPH for eachmolecule of glucose 6-phosphate oxidized. They include (Larry, 2015)

Glucose-6-phosphate Dehydrogenase Reaction

Glucose-6-phosphate dehydrogenase is the **rate-limiting step** and **primarycontrol point** of the NADPH-generating portion of the hexose monophosphatepathway (Lehninger, 2008). This enzyme catalyzes the irreversible oxidation of glucose 6-phosphate to 6-

phosphogluconolactone in a reaction that is specific for oxidized NADP (NADP+) as the coenzyme; thus its stimulated by $NADP^+$ and inhibitedby NADPH (Chayen et al., 1986).

The levels of glucose-6-phosphate dehydrogenase enzyme are also increased bythe hormone insulin; because insulin stimulates anabolic processes, and many anabolic reactions requireNADPH (Lehninger et al., 2008; Larry, 2015). Stiochemically, the reaction is represented as:

Glu-6-P + $NADP^+$ ⟶ 6-Phosphogluconolactone + NADPH + H^+

Gluconolactone Hydrolase Reaction

The 6-phosphogluconolactone produced by G6P dehydrogenase enzyme is quite unstable, and will undergo spontaneous hydrolysis to 6-Phosophogluconate (Lehninger et al., 2008). The hydrolase enzyme merely accelerates the process. Hydrolase reaction is effectively irreversible underphysiological conditions (Lehninger et al., 2008; Mark Brandt, Thesis).

6-Phosphogluconolactone + H_2O ⟶ 6-Phosphogluconate

6-Phosphogluconate Dehydrogenase Reaction

6-Phosphogluconate dehydrogenase catalyzes the formation of the second NADPH.It has a reaction mechanism similar to that of isocitrate dehydrogenase, and likeisocitrate dehydrogenase, yields an unstable product (3-keto-6-phosphogluconate)that rapidly decarboxylates to release Ribulose-5-phosphate (Lehninger et al., 2008).

In most cells, NADPH levels are much higher than NADP levels. NADPH has amuch higher free energy than $NADP^+$, both as a result of the concentrationdifference, and of the fact that, like NADH, NADPH is a more energetic moleculethan its oxidized counterpart (Chayen et al., 1986; Lehninger et al., 2008). While most dehydrogenase reactions are reversible,net synthesis of NADPH is possible because the products of both dehydrogenasereactions in this pathway are rapidly and irreversibly converted to compounds thatcannot act as substrates for the reverse reactions (Lehninger et al., 2008; Larry, 2015).

Therefore**,** *the G6P dehydrogenase and 6-phosphogluconate dehydrogenase reactions are usually termed irreversible reactions because their product concentrations are never high enough to allow the reverse reactions to proceed. The reactions stiochiochemistry are*

6-Phosphogluconolactone + NADP ⟶ 3-keto-6-phosphogluconate + NADPH

3-keto-6-phosphogluconate ⟶ Ribulose-5-phosphate+ CO_2

The enzyme; Phosphopentose isomeraseconverts Ribulose 5-phosphate to its aldose isomer,ribose 5-phosphate. In some tissues, the pentosephosphate pathway ends at this point (Lehninger et al., 2008), and its overallequation is**:**

G6P+ 2$NADP^+$+ H2O ⟶ Ribose 5-phosphate + CO2 + 2NADPH + 2H^+

The net result is the production of NADPH, a reductant for biosynthetic reactions, and ribose 5-phosphate, a precursor for nucleotide synthesis (Lehninger et al., 2008; Larry, 2015).

B. Nonoxidative Phase of Hmp Pathway

The nonoxidative reactions of the HMP pathway occur in all cell types synthesizing nucleotides and nucleic acids (Leblanc, 2008; Larry 2015). These reactions catalyze the interconversions of sugars containing three to seven carbons. These reversible reactions permit ribulose 5-phosphate) to be converted either to ribose 5-phosphate or to intermediates of glycolysis (Lehninger et al., 2008, Mark Brandt, Ph.D Thesis) For example, many cells that carry out reductive biosynthetic reactions have a greater need for NADPH than for ribose 5-phosphate. In this case, transketolase and transaldolase enzymes convert the ribulose 5-phosphate produced as an end product of the oxidative reactions to glyceraldehyde 3-phosphate and fructose 6-phosphate, which are glycolytic intermediates(Chayen et al.,1986; Leblanc 2008, Larry, 2015). In contrast, in tissues that require NADPH, the pentose phosphates produced in the oxidative phase are recycled into glucose 6-phosphate. In this nonoxidative phase, ribulose 5-phosphate is first epimerized to xylulose 5-phosphate. Then, in a series of rearrangements of the carbon skeletons, six 5-carbon sugar phosphates are converted to five 6-carbon sugar phosphates, completing the cycle and allowing continued oxidation of glucose (Lehninger et al., 2008).

Ribulose-5-phosphate isomerase Reaction:

The ribulose-5-phosphate formed in the 6-phosphogluconate dehydrogenase reactionacts as a substrate for two different enzymes.The Isomerase enzyme also converts the keto-pentose ribulose-5-phosphate to an aldo-pentoseribose-5-phosphate (Leblanc, 2008).

The product, ***ribose-5-phosphate is theprecursor for all nucleotide synthesis***, and its production is an importantfunction of the hexose monophosphate pathway (Leblanc, 2008).

Ribulose-5-phosphate ⟷ Ribose-5-Phosphate

Ribulose-5-phosphate Epimerase Reaction

Ribulose-5-phosphate epimerase converts ribulose-5-phosphate to its epimer; xylulose-5-phosphate (Lehninger et al., 2008). Because the isomerase and epimerase reactions are reversible, excess ribose-5-

phosphate (resulting from the hexose monophosphate pathway or from nucleotidebreakdown) can be converted to other compounds via the hexose monophosphatepathway (Lehninger et al., 2008, Leblanc, 2008). i.e:

Ribulose-5-phosphate
←−−−−−−−−−−−−−−−→*Xylulose-5-phosphate*

Transketolase Reaction
Transketolase enzyme catalyzes **two-carbon transfer** reactions (Chayen et al., 1986). It requires **thiamine pyrophosphate** as a cofactor, which is used in two separate reactions in the hexose monophosphatepathway. (Lehninger et al., 2008). Where it transfers two carbons from xylulose-5-phosphate to ribulose-5-phosphate to release the 7-carbon sedoheptulose-7-phosphate (Chayen et al., 1986). Alternatively, it can transfer the 2-carbons to erythrose-4-phosphate to produce fructose-6-phosphate. In both cases, the remaining carbons from the xylulose-5-phosphate arereleased as glyceraldehyde-3-Phosphate (Leblanc, 2008).

Thus, Thiamin deficiency therefore prevents the entry of ribose into glycolysis, and results in accumulation of ribose, in addition to other important effects (such as inhibition of pyruvate dehydrogenase and a-ketoglutarate i.e (Chayen et al., 1986; Leblanc, 2008; Lehninger et al., 2008; Larry, 2015):
Xylulose-5-P + Ribulose-5-P −−−−−→*Sedoheptulose-7-P + Glyceraldehyde-3-P*
OR
Xylulose-5-P +Erythrose-4-P−−−−−−−→ *Fructose-6-P + Glyceraldehyde-3-P*

Transaldolase Reaction
Transaldolase catalyzes a **3-carbon transfer** from sedoheptulose-7-phosphate to glyceraldehyde-3-P, leaving erythrose-4-P, and forming fructose-6-P (Leblanc, 2008). Note that the erythrose-4-P produced in this reaction is a substrate for one of the transketolase reactions. The transaldolase reaction also involves the formation of a covalent intermediate between the enzyme and substrate. In the case of transaldolase, however, the covalent intermediate isformed between the substrate and a lysine side chain of the enzyme (Lehninger et al., 2008; Larry, 2015).

Pathway variations
Intermediates can enter the hexose monophosphate pathway from glycolysis (which bypasses NADPH synthesis) (Leblanc, 2008). This is especially important in muscle and other tissues that lack significant amounts of glucose-6-phosphate dehydrogenase; these tissues can therefore synthesize ribose-5-phosphate for nucleotide biosynthesis, although they produce little NADPH (Lehninger et al., 2008). Alternatively, ribose-5-phosphate can enter the pathway to be broken down for energy or converted into other compounds(Chayen et al.,1986; Lehninger et al., 2008; Larry, 2015).

Uses of Nadph
Chemically NAD and $NADP^+$ are very similarmolecules. The two molecules are structurallynearly identical (the only difference is thephosphate on the 2´-position of the adenosineribose of NADP instead of the free hydroxyl atthis position in NAD (Larry, 2015).

Physiologically, however, the two nicotinamide cofactors are quite different. NAD isused to accept electrons in catabolic processes (such as the TCA cycle), whileNADPH is primarily used to donate electrons for synthetic reactions (Lehninger et al., 2008; Leblanc, 2008).

Most dehydrogenase enzymes are extremely specific, using either NADP(H) or NAD(H), but not both. Under most conditions, the cellular concentration of NADPH is much higher than the concentration of NADP (Leblanc, 2008). NADPH can therefore act as a strong driving force for otherwise unfavorable reactions because of both the concentration differential and the fact that NADPH contains more energy than NADP (in the same way that NADH contains more energy than NAD)(Chayen et al, 1986).

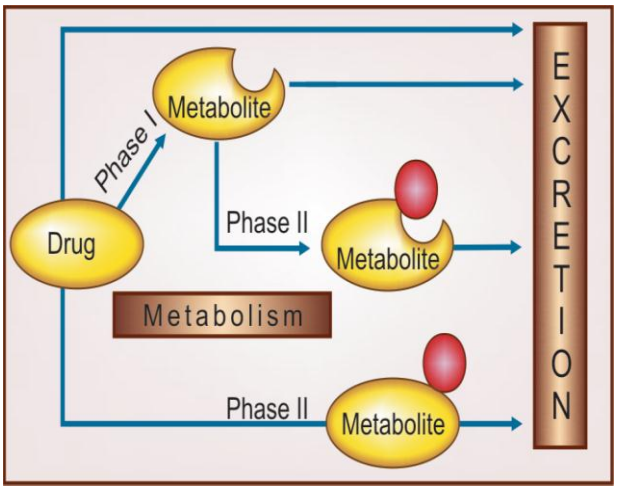
In contrast, the reduction of NAD associated with a number of catabolic reactions is assisted by the fact that NAD is normally maintained at much higher concentrations than NADH (Leblanc, 2008).NADPH is used for a variety of electron donation reactions. In addition to its involvement in synthetic processes, in some cell types, NADPH is required to maintain normal functioning (Lehninger et al 2008).

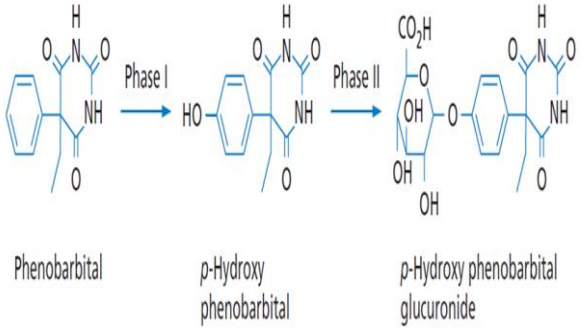
NADPH in Red Blood Cells
NADPH plays two critical roles in red blood cells. Both of these roles are related to the oxygen-transport function of the red blood cell (Lehninger et al., 2008). The first role is related to glutathione: erythrocytes require NADPH to maintain their levels of reduced glutathione (Lehninger et al., 2008). The second role of NADPH in red blood cells is related to haemoglobin. Oxygen tends to oxidize the haemoglobin iron from +2 to the more stable +3 oxidation state (resulting in methemoglobin) (Lehninger et al., 2008). This is a problem: the +3 state of heme iron binds oxygen very poorly. NADPH is used to supply reducing equivalents to methemoglobin reductase, the enzyme that returns the haemoglobin to the +2 oxidation statecycle(Chayen et al.,1986; LeBlanc , 2008; Lehninger et al., 2008; Larry, 2015).In Glucose-6-phosphate dehydrogenase deficiency; a genetically-mediated reduction in glucose-6-phosphate dehydrogenase levels is somewhat protective against malaria (Larry, 2015).The result of the deficiency is a decreased ability to form NADPH in red blood cells. Malaria parasites live inside red blood cells. Red blood cells normally have a lifespan of about 120 days (Larry, 2015). A number of mutations (glucose-6-phosphate dehydrogenase deficiency, sickle cell anemia, and the thalassemias) shorten the lifespan of the red blood cell,

resulting in death of the malaria parasites that have not reached maturity. In the case of glucose-6-phosphate dehydrogenase deficiency, the most common mutations appear to decrease the half-life of the enzyme, which is effective in reducing the life-span of the cell because red blood cells lack the ability to synthesize new proteins (Lehninger et al 2008; Leblanc, 2008). The erythrocytes in individuals with glucose-6-phosphate dehydrogenase deficiency are asymptomatic unless challenged with oxidants; oxidant exposure, which however, results in haemolytic anemia (Chayen et al., 1986). The haemolysis occurs because, as the membrane lipids become oxidized, the membrane becomes less flexible. Less flexible red blood cell membranes are a serious problem, because the red blood cell is larger than the typical capillary, and if insufficiently flexible, either cannot enter the capillary or lyses as it attempts to do so(Chayen et al.,1986).

Xenobiotic (Drug) Metabolism

Evolution of Drug Metabolism as a Science had pioneers such as Richard Tecwyn Williams (1942) from Great Britain; who worked on the metabolism on TNT with regard to toxicity in munitions workers (Luzzatto *et al.*, 2001). He developed the concept of Phase 1 and Phase 2 reactions which entailed Biotransformation (involving metabolic oxygenation, reduction, or hydrolysis) and Conjugation, which in almost all cases result in the detoxification of the drug(Benedetti ,2001**;**; Lehninger et al., 2008; Goodman and Gilman, 2009).





Majority of drug metabolism occur in the liver, an organ devoted tothe synthesis of many important biologically functional proteins, which also has thecapacity to mediate chemical transformations of xenobiotics (Nelson, 2003). Most drugs thatenter the body are lipophilic, a property that enables them to bind to lipid membranesand to be transported by lipoproteins in the blood. After their entrance into the liver, as well as in other organs, drugs may undergo one or two phases of metabolism. Phase I reaction of a drug biotransformation is also referred to as the Non-synthetic / Functional reaction and the Phase II is also known as the Synthetic or Conjugation reaction of drug biotransformationdrug (Benedetti ,2001). In Phase I, a polar reactive group is introduced into the molecule, rendering it a suitable substrate for Phase II enzymes. Enzymes typically involvedin Phase I metabolism include the CYPs (Cytochromes), FMOs (flavin monooxygenases), and hydrolases etc (Luzzatto et al., 2001; Nelson, 2003; Lehninger et al., 2008; Goodman and Gilman; 2009). In Phase II, following the introduction of a polar group, conjugating enzymestypically add endogenous substituents, such as sugars, sulfates, or amino acids whichresult in substantially increasing the water solubility of the drug, making it easily excreted. Although this process is generally a detoxification sequence, reactive intermediates may be formed that are much more toxic than the parent compound (Lehninger et al., 2008; Goodman and Gilman, 2009).The role of the transport proteins, known collectively as *transporters* is often referred to as the Phase III reaction in some articles (Goodman and Gilman, 2009). Also, according to Triparti et al., 2008; there is another infamous type of Drug biotransformation known as *Hofmann Elimination*. In this type of biotransformation; inactivationof the drugs in the body fluidsoccurs by spontaneous molecular rearrangement without the aid of any enzyme. Example of such a drug is Atracurium. Biotransformation of drugs may lead to thefollowing (Benedetti ,2001; Triparti, 2009):

(i) *Inactivation*

Most drugs and their activemetabolites are rendered inactive or less active,*e.g. ibuprofen, paracetamol, lidocaine, chloramphenicol, propranolol.*(Triparti, 2009)

(ii) *Active metabolite from an active drug*

Many drugs have been found to be partiallyconverted to one or more active metabolite; theeffects observed are the sumtotal of that due tothe parent drug and its active metabolite(s).

Examples: Active drug *Active metabolite*
Procainamide — *N-acetyl procainamide*
Diazepam — *Desmethyl-diazepam, oxazepam*
Digitoxin — *Digoxin*
Imipramine — *Desipramine*
Codeine — *Morphine*

(iii) *Activation of inactive drug*

Few drugs are inactive as such and need conversion in the body to one or more active metabolites. Such a drug is called a *prodrug*. Prodrugs may offer advantages over the active form in being more stable, having better pharmacokinetic properties. Examples include-

Prodrug (Inactive Drug)	*Active Drug*
Levodopa —	*Dopamine*
Enalapril —	*Enalaprilat*
Proguanil —	*Cycloguanil*
Prednisone —	*Prednisolone*

Phase I Reactions

Phase I reactions include Microsomal Monooxygenations, Cytosolic and Mitochondrial oxidations, Co - oxidations in the Prostaglandin synthetase reaction, Reductions, Hydrolysis, and Epoxide hydration. All of these reactions, with the exception of reductions, introduce polar groups to the molecule that, in most cases, can be conjugated during a Phase II metabolism (Koukouritaki, et al. 2002; Hodgson et al, 2008; Goodman and Gilman, 2009).

A, The Endoplasmic Reticulum, Microsomes, and Monooxygenations

Monooxygenations of drugs are catalyzed either by the CYP – dependent monooxygenase system or by the FMOs (Triparti, 2009). Both are located in the smooth endoplasmic reticulum of a cell while microsomes are derived from the endoplasmic reticulum as a result of tissue homogenization and are isolated by centrifugation of the post mitochondrialsupernatant fraction (Triparti, 2009). The endoplasmic reticulum is an anastomosing network of lipoprotein membranes extending from the plasma membrane to the nucleus and mitochondria, whereas the microsomal fraction derived from it consists of membranous vesicles contaminated with free ribosomes, glycogen granules, and fragments of other subcellular structures such as mitochondria and Golgi apparatusdrug (Notaro et al., 2000; Benedetti ,2001; Nelson, 2003).

Monooxygenations, previously known as mixed - function oxidations, are those oxidations in which one atom of a molecule of oxygen is incorporated into the substrate while the other is reduced to water: (Triparti, 2009) i.e

$$RH+O + NADPH+H+ \rightarrow NADP^{+} + ROH + H_2O$$

The CYP - Dependent Monooxygenase System

In vertebrates, the liver is the richest source of CYPs and is most active in the monooxygenation of drugs. CYP and other components of the CYP - dependent monooxygenase system are also found in the skin, nasal mucosa, lung, and gastrointestinal tract. In addition to these organs, CYP has been demonstrated in the kidney, adrenal cortex and medulla, placenta, testes, ovaries, fetal and embryonic liver, corpus luteum, aorta, blood platelets, and the nervous system (Koukouritaki , et al. 2002; Hodgson et al, 2008). Although CYPs are found in many tissues, the function of the particular subset of isoforms in a particular organ, tissue, or cell type does not appear to be the same in all cases. In the liver, CYPs oxidize a large number of drugs as well as some endogenous steroids and bile pigments. Whereas the CYPs of a lung appears to bemore concerned primarily with drug oxidations, although the range of substrates is more limited than that of the liver. The skin and small intestine also carry out drug oxidations, but their activities have been less well documented. And in normal pregnant females, the placental microsomes display little or no ability to oxidize foreign compounds, appearing to function as a steroid hormone metabolizing systemdrug (Notaro et al., 2000; Benedetti 2001; Koukouritaki, et al. 2002; Hodgson et al, 2008; Goodman and Gilman, 2009). The CYP1 family contains three known human members, CYP1, CYP2, and CYP3 with their respective subset of isoforms with overlapping substrate specificities (Guengerich, 2008):

CYP1 family members are closely associated with metabolic activation of many procarcinogens and mutagens including benzo(a)pyrene, aflatoxin B1, dimethylbenzanthracene,β - naphthylamine , 4 - aminobiphenyl, 2 - acetylaminofl uorene, and benzidine.

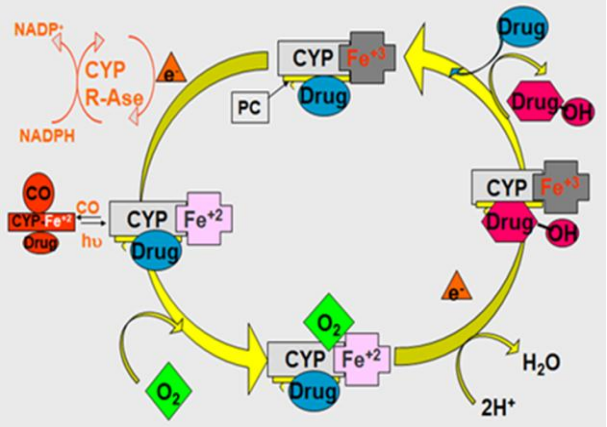
CYP2 family consists of 10 subfamilies, five of which are present in mammalian liver. Some of the more important isoforms found in humans within this family are CYP2A6, CYP2B6, CYP2C8, CYP2C9, CYP2C19, CYP2D6, and CYP2E1. The enzyme CYP2A6 is expressed primarily in liver tissue, where it represents 1 – 10% of total CYP content. CYP2A6 is responsible for the 7 – hydroxylation of the naturally occurring plant compound coumarin, and its activity is often phenotyped by monitoring this particular metabolic pathway. Other drugs metabolized by CYP2A6 include nicotine, 2 - acetylaminofluorine, methoxyflurane, halothane, valproic acid, and disulfiram. Procarcinogens likely activated by CYP2A6 includeaflatoxin B1, 1,3 butadiene, 2,6 - dichlorobenzonitrile, and a number of nitrosamines (Oritz, 2004).

Because CYP2A6 is responsible for up to 80% of the human metabolism of nicotine, a number of studies have been conducted to determine whether individuals with CYP2A6 polymorphisms have reduced risk of lung cancers (Cashman and Zhang, 2006). Like CYP2A6, some common drugs for CYP2B6 include *cyclophosphamide, nevirapine, S - mephobarbital, artemisinin, bupropion, propofol, ifosfamide, ketamine, selegiline, and methadone*.CYP2B6 has also been demonstrated to have a role in the activation of the *organophosphorus insecticide, chlorpyrifos, and in the degradation of the commonly used insecticide repellant, diethyl toluamide (DEET*). Historically, it was thought that CYP2B6 is found in only a small proportion of livers (< 25%), but more recent data using antibodies prepared from human proteins have demonstrated that

most liversamples have detectable levels of CYP2B6 (Oritz, 2004). In contrast with CYP2A6 and CYP2B6, members of the CYP2C family constitute a fairly large percentage of CYP in human liver (20%) and are responsible for the metabolism of several drugs. All four members of the subfamily in humans exhibit genetic polymorphisms, many of which have important clinical consequences in affected individuals (Triparti, 2009). Genetic polymorphisms in CYP2C19 were shown to be responsible for one of the earliest described polymorphic effects, that involving mephenytoin metabolism. This particular polymorphism significantly reduces the metabolism of mephenytoin resulting in the classification of those individuals possessing this trait as poor metabolizers (Triparti, 2009). Other important drugs affected by these CYP2C19 polymorphisms include the antiulcer drug *omeprazole*, other important *proton pump inhibitors, barbiturates, certain tricyclic antidepressants such as imipramine, and the antimalarial drug proguanil*. Other important members of the CYP2C family in humans include CYP2C8, CYP2C9, and CYP2C18.Substrates metabolized exclusively by CYP2C8 include*retinol, retinoic acid, taxol, and arachidonic acid* (Guengerich, 2008). CYP2C9, the principal CYP2C in human liver, metabolizes several important drugs including the *diabetic agent tolbutamide, the anticonvulsant phenytoin, the anticoagulant warfarin, and a number of anti - inflamatory drugs including ibuprofen, diclofenac*, and others. Both CYP2C9 and CYP2C8are responsible for metabolism of the anticancer drug paclitaxel that have been demonstrated to be polymorphic in recent studies (Oritz, 2004). According to Benedetti, (2001), the largest amount of CYP in human liver is that of the CYP3 family. CYP3A4 is the most abundant CYP in the human liver, on average accounting for approximately 30% of the total amount, and is known to metabolize many important drugs, including (Goodman and Gilman, 2009; Triparti, 2009).

cyclosporin A, nifedipine, rapamycin, estradiol, quinidine, digitonin, lidocaine, erythromycin, midazolam, triazolam, lovastatin, and tamoxifen. Other important oxidations ascribed to the CYP3 family include many steroid hormones, macrolide antibiotics, alkaloids, benzodiazepines, dihydropyridines, organophosphorus insecticides and other insecticides, warfarin, polycyclic hydrocarbon - derived dihydrodiols, and afl atoxin B 1 (Oritz, 2004).

Many chemicals are also capable of inducing CYPs of this family including phenobarbital, rifampicin,and dexamethasone. Because of potential difficulties arising from CYP induction, drugs metabolized by this family must be closely examined for the possibility of harmful drug – drug interactions (Koukouritaki, et al. 2002; Hodgson et al, 2008).



Electron flow in microsomal drug oxidizing system

Listing of important oxidation and reduction reactions of CYPs

1.Epoxidation and Aromatic Hydroxylation- Epoxidation is an extremely important microsomal reaction because not only can stable and environmentally persistent epoxides beformed, but highly reactive intermediates of aromatic hydroxylations,, can also be produced. These highly reactive intermediates are known to be involved in chemical carcinogesis as well as chemically induced cellular and tissue necrosis (Triparti, 2009).

These aromatic epoxidation reactions are also of importance in the metabolism of other xenobiotics that contain an aromatic nucleus, such as the insecticide carbaryl and the carcinogen benzo (a) pyrene (Goodman and Gilman, 2009; Richter, 2009).

2. Aliphatic Hydroxylation- Simple aliphatic molecules such as *n* - butane, *n* - pentane, *n* - hexane and so on, as well as alicyclic compounds such as cyclohexane, are known to be oxidized to alcohols (Guengerich, 2008; Triparti, 2009).

3. Aliphatic Epoxidation-Many aliphatic and alicyclic compounds containing unsaturated carbon atoms that are metabolized to epoxide intermediates. Epoxide formation in the case of aflatoxin is believed to be the final step in formation of the ultimate carcinogenic species and is, therefore, an activation reaction ((Guengerich, 2008; Triparti, 2009).

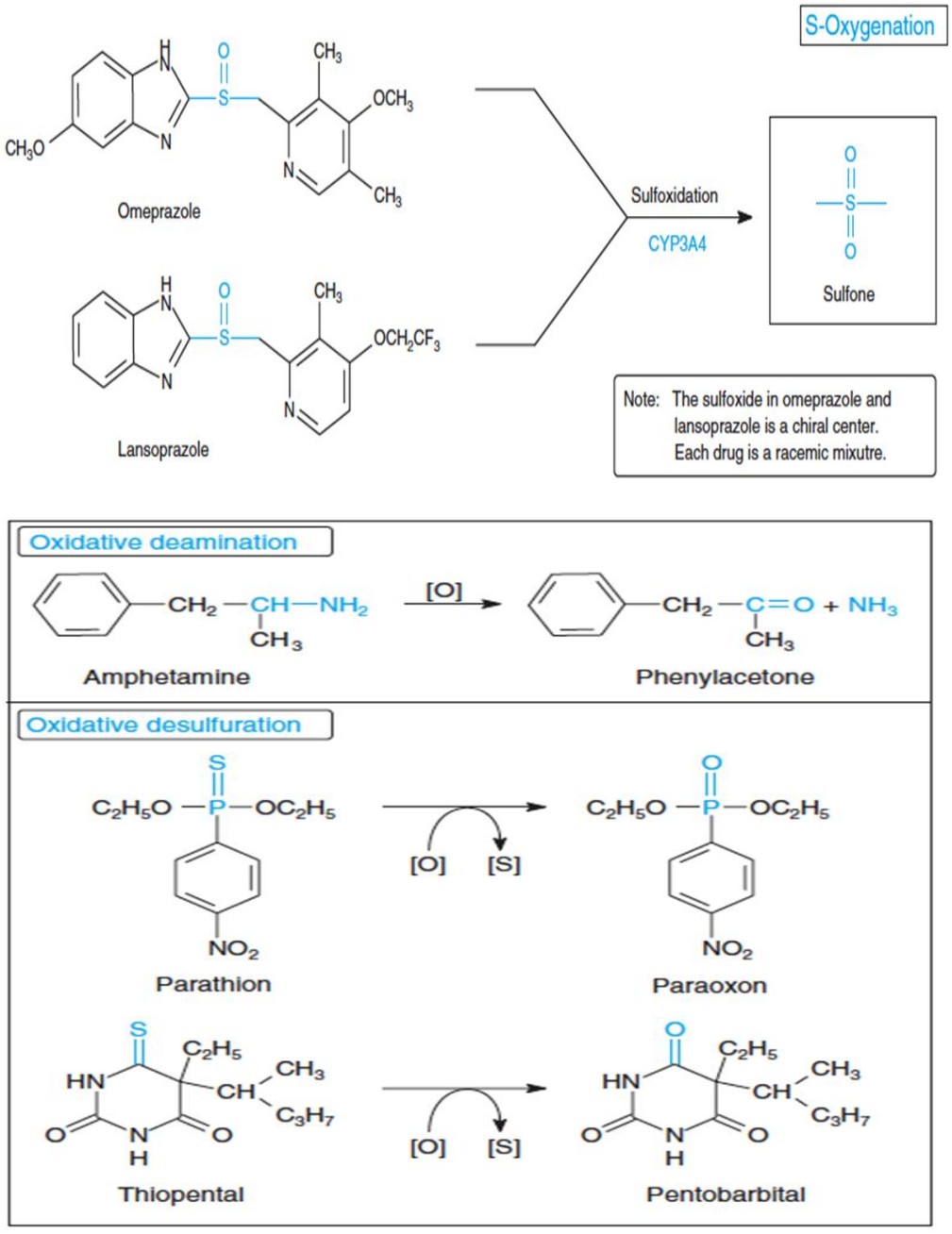
4. O- Dealkylation - the O - dealkylation of organophosphorus triesters differs from that of *p* - nitroanisole in that it involves the dealkylation of an ester rather than ether (Goodman and Gilman, 2009).

5. N – dealkylation- is a common reaction in the metabolism of drugs, insecticides, and other xenobiotics. The drug ethylmorphine and diazepam are useful model compounds for this reaction (Ritter, 2001; Cashman and Zhang, 2006; Richter, 2009).

6. S – dealkylation- is believed to occur with a number of thioethers, including methylmercaptan and 6 – methylthiopurine (Cashman and Zhang, 2006).

7. N - Oxidation- can occur in a number of ways, including hydroxylamine formation, oxime formation, and N - oxide formation, although the latter is primarilydependent on the FMO enzyme (Koukouritaki, et al. 2002; Hodgson et al, 2008; Goodman and Gilman, 2009).

8. S - Oxidation- This reaction is very common among insecticides of several different chemical classes, including carbamates, organophosphorus compounds, and chlorinated hydrocarbons. Drugs, including chlorpromazine, omperazole and solvents, such as dimethyl sulfoxide, arealso subject to S – oxidation (Benedetti, 2001; Ritter, 2001).



9. Epoxide Hydration

Epoxide rings of alkene and arene compounds are hydrated by enzymes known as epoxide hydrolases. Although in general, the hydration of the oxirane ring results in detoxication of the very reactive epoxide, in some cases, such as benzo (a) pyrene, the hydration of an epoxide is the first step in an activation sequence that ultimately yields highly toxic *trans* - dihydrodiol intermediates(Ritter, 2001; Guengerich , 2008; Smart, 2008).

Products of Phase I metabolism and other xenobiotics containing functional groups such as hydroxyl, amino, carboxyl, epoxide, or halogen can undergo conjugation reactions with endogenous metabolites, these conjugations being collectively termed Phase II reactions. The endogenous metabolites in question include sugars, amino acids, GSH, sulfate, and so on(Goodman and Gilman, 2009). Conjugation products, with rare exceptions, are more polar, less toxic, and more readily excreted than are their parent compounds. Conjugation reactions usually involve activation by some high - energy intermediate and have been classified into two general types: type I, in which an activated conjugating agent combines with the substrate to yield the conjugated product, and type II, in which the substrate is activated and then combines with an amino acid to yield a conjugated product. The formation of sulfates and glycosides are examples of type I, whereas type II consists primarily of amino acid conjugation (Ritter, 2001; Ross and Crow, 2007; Lehninger et al., 2008; Smart, 2008; Goodsman and Gilman, 2009).

B. The Flavin-Monooxygenase System

FAD-containing monooxygenases (FMO) oxidize nucleophilic nitrogen, sulfur and phosphorus heteroatoms of a variety of drugs. Some peculiarities of FMOs (Guengerich, 2008; Triparti, 2009).

Include

- FMO's are **not** inducible and are constitutively expressed
- Can be inhibited by other substrates.

C. Non-microsomal Oxidations

In addition to the microsomal monooxygenases, other enzymes are involved in the oxidation of drugs. These enzymes are located in the mitochondria or in thesoluble cytoplasm of the cell; thus there name: non-microsomal oxidation enzymes (Triparti, 2009). They include-

1. Alcohol Dehydrogenase- Alcohol dehydrogenases catalyze the conversion of alcohols to aldehydes or ketones (Goodman and Gilman, 2009).

$$RCH_2OH + NAD^+ \rightarrow RCHO + NADH + H^+$$

Alcohol dehydrogenase can use either NAD or NADP as a coenzyme, but the reaction proceeds at a much slower rate with NADP. In the intact organism, the reaction proceeds in the direction of alcohol consumption, because aldehydes are furtheroxidized to acids. Because aldehydes are toxic and are not readily excreted because of their lipophilicity, alcohol oxidation may be considered an activation reaction, the further oxidation to an acid being a detoxification step (Benedetti ,2001; Tukey and Strassburg; 2008).

Primary alcohols are oxidized to aldehydes, *n* - butanol being the substrate oxidized at the highest rate. Although secondary alcohols are oxidized to ketones, the rate is less than for primary alcohols, and tertiary alcohols are not readily oxidized. Alcohol dehydrogenase is inhibited by a number of heterocyclic compounds such as pyrazole, imidazole, and their derivatives (Guengerich, 2008).

2. Aldehyde Dehydrogenase- Aldehydes are generated from a variety of endogenous and exogenous substrates. Endogenous aldehydes may be formed during metabolism of amino acids, carbohydrates, lipids, biogenic amines, vitamins, and steroids. Metabolism of many drugs and environmental agents produce aldehydes (Cashman and Zhang, 2006). Aldehydes are highly reactive electrophilic compounds and may react with thiol and amino groups to produce a variety of effects. Some aldehydes produce therapeutic effects, but more often effects are cytotoxic, genotoxic, mutagenic, or carcinogenic (Smart, 2008). Aldehyde dehydrogenases are important in helping to alleviate some of the toxic effects of aldehyde generation. This enzyme catalyzes the formation of acids from aliphatic and aromatic aldehydes; the acids are then available as substrates for conjugating enzymes:

$$RCHO + NAD^+ \rightarrow RCOOH + NADH + H^+$$

3. *Amine Oxidases* - The most important function of amine oxidases appears to be the oxidation of amines formed during normal processes. Two types of amine oxidases are concerned with oxidative deamination of both endogenous and exogenous amines (Zeildin, 2008).

4. *Monoamine Oxidases* - The monoamine oxidases are a family of flavoproteins found in the mitochondria of a wide variety of tissues: liver, kidney, brain, intestine, and blood platelets (Zeildin, 2008).

5. Diamine Oxidases - are enzymes that also oxidize amines to aldehydes. The preferred substates are aliphatic diamines (Koukouritaki , et al. 2002; Hodgson et al, 2008).

C. Co - oxidation by Cyclooxygenase (COX)

During the biosynthesis of prostaglandins, a polyunsaturated fatty acid, such as arachidonic acid, is first oxygenated to yield a hydroperoxy endoperoxide, prostaglandinG2. This is then further metabolized to prostaglandin H2, both reactionsbeing catalyzed by the same enzyme, COX, also known as prostaglandin synthase. This enzyme is located in the microsomal membrane and is found ingreatest levels in respiratory tissues such as the lung. It is also common in the kidneyand seminal vesicle (Guengerich, 2008).

Many of the reactions are similar or identical to those catalyzedby other peroxidases and also by microsomal monooxygenases; they include bothdetoxification and activation reactions. This mechanism is important in xenobioticmetabolism, particularly in tissues that are low in CYP and/or the FMO but highin prostaglandin synthase. The COX enzyme is known to exist as two distinct isoforms. COX - 1 is a constitutivelyexpressed

housekeeping enzyme found in nearly all tissues and mediatesphysiological responses. COX - 2 is an inducible form expressed primarily by cells involved in the inflammatory response. Several tissues low in CYP expression arerich in COX, which is believed to have significance in the carcinogenic effects ofaromatic amines in these organs. During co - oxidation, some substrates are activated to become more toxic thanthey were originally. In some cases, substrate oxidation results in the production offree radicals, which may initiate lipid peroxidation or bind to cellular proteins orDNA(Koukouritaki , et al. 2002; Hodgson et al, 2008).

Another activation pathway involves the formation of a peroxyl radical fromsubsequent metabolism of prostaglandin G2. These reactive molecules can epoxidizemany substrates including polycyclic aromatic hydrocarbons, generally resulting inincreasing toxicity of the respective substrate (Benedetti 2001).

D. Reduction Reactions

A number of functional groups, such as nitro, diazo, carbonyl, disulfi desulfoxide,alkene, pentavalent arsenic, and so on, are susceptible to reduction. Examples include-

Ketone and Aldehyde Reduction In addition to the reduction of aldehydeand ketones through the reverse reaction of alcohol dehydrogenase, a family ofaldehyde reductases also reduces these compounds. These reductases are NADPH - dependent, cytoplasmic enzymes of low molecular weight, and have been found inliver, brain, kidney, and other tissues (Guengerich , 2008).

Sulfoxide Reduction The reduction of sulfoxides has been reported to occur in mammalian tissues. Soluble thioredoxin - dependent enzymes in the liver areresponsible in some cases (Zeildin, 2008).

E. Hydrolysis

Enzymes with carboxylesterase and amidases activity are widely distributed in the body, occurring in many tissues and in both microsomal and soluble fractions(Cashman and Zhang, 2006; Goodman and Gilman, 2009).

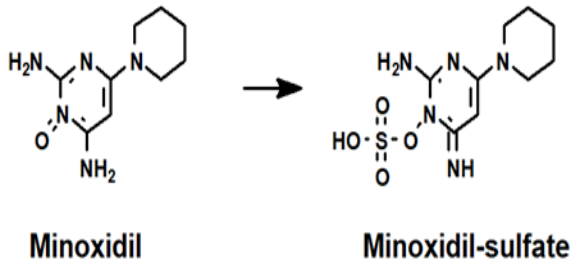
Phase II Reactions

1. Glucuronide Conjugation

The glucuronidation reaction is one of the major pathways for elimination of many lipophilic drugs and endobiotics from the body (Zeildin, 2008). The mechanism for this conjugation involves the reaction of one of many possible functional groups (R - OH, Ar - OH, R- NH2, AR- NH2, R- COOH, Ar - COOH) with the sugar derivative, uridine 5 ′ - diphosphoglucuronic acid (UDPGA) (Goodman and Gilman, 2009). The enzyme involved, the UDP glucuronosyl transferase (UGT), is found in the microsomal fraction of liver, kidney, intestine, and other tissues. Glucuronide conjugation generally results in the formation of products that are less biologically and chemically reactive. This, combined with their greater polarity and greater susceptibility to excretion, contributes greatly to the detoxication of most xenobiotics. However, there are examples of glucuronide conjugation resulting in greater toxicity. Perhaps the best - known example involves the bioactivation of *N* - hydroxy - 2 - acetylaminofluorine. This substrate, unlike 2 - acetylaminofluorine, is unable to bind to DNA without metabolic activation. However, following glucuronide conjugation by linkage of the oxygen through the *N* - hydroxy group, this substrate becomes equipotent as a hepatocarcinogen with 2 – acetylaminofluorine based on its ability to bind to DNA(Benedetti ,2001; (Guengerich , 2008). Another relatively large class of xenobiotics that are often activated by glucuronide conjugation are the acyl glucuronides of carboxylic acids. Useful therapeutic drugs within this class include nonsteroidal anti - inflammatory drugs (NSAIDS), hypolipidemic drugs (clofibrate), and anticonvulsants (valproic acid). The various syndromes associated with the clinical use of some of these drugs (including cytotoxic, carcinogenic, and various immunologic effects) are thought to be the result of the ability of the glucuronide conjugates to react with nucleophilic macromolecules (protein and DNA) (Cashman and Zhang, 2006).

2. Sulfate Conjugation

Sulfation and sulfate conjugate hydrolysis, catalyzed by various members of the sulfotransferases (SULT) and sulfatase enzyme superfamilies, play important roles in the metabolism and disposition of many xenobiotics and endogenous substrates (Zeildin, 2008).



Sulfation may produce active metabolite

3. Methyltransferases

A large number of both endogenous and exogenous compounds can be methylated by several *N* - , *O* - , and *S* - methyltransferases. The most common methyl donor is *S* - adenosylmethionine (SAM), formed from methionine and ATP. Even though these reactions may involve a decrease in water solubility, they are generally detoxication reactions.

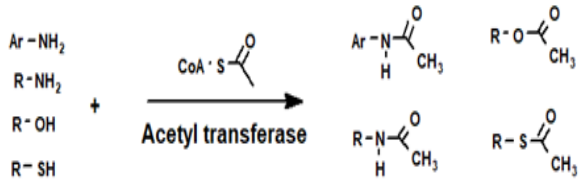
4. Acylation

Acylation reactions are of two general types, the first involving an activated conjugation agent, coenzyme A (CoA), and the second involving activation of the foreign

compounds and subsequent acylation of an amino acid(Cashman and Zhang, 2006)

5. Acetylation

Acetylated derivatives of foreign exogenous amines are acetylated by *N* - acetyltransferase, the acetyl donor being CoA. Acetylation of foreign compounds is influenced by both development and genetics. Newborn mammals generally have a low level of the transferase whereas, due to the different genes involved, fast and slow acetylators have been identified in both rabbit and human populations. Slow acetylators are more susceptible to the effects of compounds detoxified by acetylation (Goodman and Gilman, 2009).



Examples: Procainamide, isoniazid, sulfanilimide, histamine

6. Amino Acid Conjugation

In the second type of acylation reaction, exogenous carboxylic acids are activated to form S - CoA derivatives in a reaction involving ATP and CoA. These CoA derivatives then acylate the amino group of a variety of amino acids. Glycine and glutamate appear to be the most common acceptor of amino acids in mammals. In other organisms, other amino acids are involved(Benedetti ,2001).

Adverse Effects of Drugs (Xenobiotics) Metabolism

1. Acetaminophen-under a normal dosage regimen, acetaminophen undergoes phase II metabolism. However in an over-dose case, its metabolic pathway enters into an alternative pathway that can also be induced by substances such as ethanol, isoniazid. In this alternative pathway; N-acetyl-p-benzoquinone imines are formed that acts as protein adducts to nucleic acids-DNA, RNA, thus leading to oxidative stress and toxicity (Zeildin, 2008).

2. Benzene: benzene metabolites targets liver, kidney, lung, heart, and brain and can cause DNA strand breaks, chromosomal damage, protein binding—can cause bone marrow suppression and leukemia (Zeildin, 2008).

3. Carbon Tetrachloride

$CCL_4$was formerly widely used in fire extinguishers and as a cleaning agent.

CCl_4 is one of the most potent hepatotoxins and causes liver necrosis, and can also affect nervous system and kidneys (Zeildin, 2008).

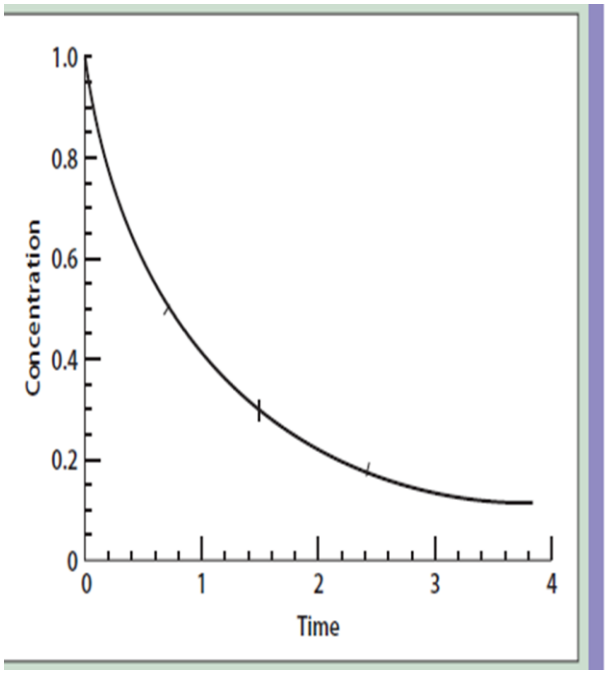
4. Halothane

Halothane is an inhalational general anesthetic and recent studies have shown that its repeated exposure causes severe liver injury. Halothane induces hepatitis and was largely replaced in the 1980s by isoflurane and sevoflurane (Zeildin, 2008).

Also studies have shown that Quinone-Cycling Causes Toxicity through Multiple Mechanisms- (Zeildin, 2008).

Kinectics of Drug Metabolsim

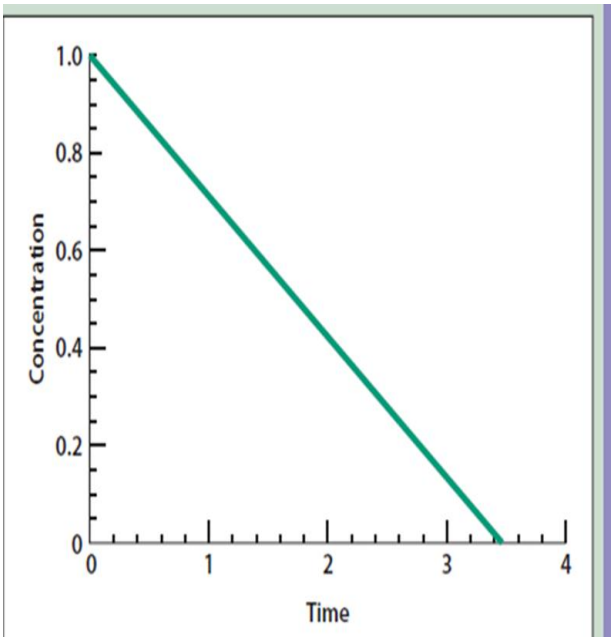
Drugs that are metabolised in such a way that a fixed half-life can be measured from its drug concentration-time graph are said to undergo *first-order reactions* or to follow *first-order kinetics (depicted below)*. Most drugs used at therapeutic dosages tend to follow this pattern and thus fit into the preceding section on drug metabolism (Goodman and Gilman, 2009).



However, a few drugs do not follow first-order kinetics. Instead, these drugs undergo what is termed *zero-order kinetics*. In this case, metabolism takes place at a constant rate, and the metabolic process is measured in the amount of drug metabolised per unit time. For example, ethanol is metabolised at a rate of approximately 10_mL/h. therefore, this is quite different from dealing with half-lives (Guengerich, 2008).

This type of kinetic process is due to the body having a limited amount of enzymes for metabolic processes or better still enzymes that are saturable (i.e Michaelis Menten Enzyme Kinetics), which can consequently quickly become saturated with excess quantities of its substrate. Ethanol does follow first-order kinetics if consumed at a rate of less than 8_g/h by an average-sized man (Goodman and Gilman, 2009).

Drug kinetics further explains why because of enzyme saturation, several drugs, if taken in excess (either intentionally or accidentally) may change their rate of metabolism from first-order to zero-order kinetics. Two common examples of such drugs areaspirinand phenytoin. Other factors that can affect the kinetics of drug metabolism are-genetics, gender and disease conditions (Benedetti,2001).



Induction/ Inhibition Of Drug Metabolism And Conclusion

Drugs can influence the extent of another drug metabolism or itself by activating transcriptionand inducing the expression of genes encoding the drug's metabolizing enzymes. One detriment of this is a decrease in plasmadrug concentration over the course of treatment, resulting in loss of efficacy (Cashman and Zhang, 2006).

A particular nuclear receptor, when activated by a ligand, induces a sequential transcription of targetgenes. Among these target genes are certain CYPs anddrug transporters. Thus, any drug that is a ligand for areceptor that induces CYPs and transporters could leadto drug interactions (Guengerich, 2008).

The aryl hydrocarbon receptor (AHR) is a member of a superfamily of transcription factors with diverseroles in mammals, such as a regulatory role in thedevelopment of the mammalian CNS and modulatingthe response to chemical and oxidative stress. AHR induces the expression of genes encodingCYP1A1 and CYP1A2. Thus, the induction ofthese CYPsby a drug could potentially result in an increase in thetoxicity and carcinogenicity of procarcinogens. Forexample, *omeprazole*, a proton pump inhibitor; is aligand for the AHR and can induce CYP1A1 andCYP1A2, with the possible consequences of toxin/carcinogenactivation as well as drug-drug interactions inpatients receiving agents that are substrates for eitherof these CYPs (Cashman and Zhang, 2006; Goodman and Gilman, 2009).

Another important induction mechanism is due to type2 nuclear receptors that are in the same super family as thesteroid hormone receptors of these receptors are activated by xenobiotics, including drugs. The type 2 nuclear receptors of most importance to drug metabolism and drug therapy include the Pregnane X receptor (PXR), constitutive androstane receptor(CAR), and the peroxisome proliferator activated receptor(PPAR).

PXR, discovered based on its ability to be activatedby the synthetic steroid pregnane 16α-carbonitrile,is activated by a number of drugs including, antibiotics(*rifampicin* and *troleandomycin*), Ca2+ channel blockers(*nifedipine*), statins (*mevastatin*), antidiabetic drugs (*troglitazone*),HIV protease inhibitors (*ritonavir*), and anticancerdrugs (*paclitaxel*).PXR also induces theexpression of genes encoding certain drug transportersand phase 2 enzymes including SULTs and UGTs. Thus, PXR facilitates the metabolism and elimination of xenobiotics, including drugs (Guengerich, 2008).

CAR was discovered based on itsability to activate genes in the absence of ligand. Steroidssuch as *androstanol*, the antifungal agent *clotrimazole*,and the antiemetic *meclizine* are inverse agonists thatinhibit gene activation by CAR, while the pesticide 1,4-bis[2-(3,5-dichloropyridyloxy)]benzene, the steroid 5β-pregnane-3,20-dione, and probably other endogenouscompounds, are agonists that activate gene expressionwhen bound to CAR. Genes induced by CAR includethose encoding several CYPs (CYP2B6, CYP2C9, andCYP3A4), various phase 2 enzymes (including GSTs, and SULTs), and drug and endobiotic transporters.

Peroxisome proliferator activated receptor (PPAR) family is composed of three members, α, β, and γ.PPARαis the target for the fibrate class of hyperlipidemicdrugs, including the widely prescribed *gemfibrozil*and *fenofibrate*. While activation of PPARαresults in induction of target genes encoding fatty acidmetabolizing enzymes that result in lowering of serumtriglycerides, it also induces CYP4 enzymes that carryout the oxidation of fatty acids and drugs with fattyacid–containing side chains (Cashman and Zhang, 2006; Guengerich, 2008)

REFERENCES

1. Benedetti, M. S. Biotransformation of xenobiotics by amine oxidases. *Fundam. Clin. Pharmacol*, 2001; 15: 75 – 84.
2. Cashman, J. R. and J. Zhang Human fl avin - containing monooxygenases *Annu. Rev. Pharmacol. Toxicol*, 2006; 46: 65 – 100.
3. Chayen, J., Howat, D. W., and Bitensky, L. Cellular biochemistry of glucose 6-phosphate and 6-

phosphogluconate dehydrogenase activities. *Cell Biochem. Funct*, 1986; 4: 249–253.
4. Goodman and Gilman. (2009). The Pharmacological basis of therapeutics. Drug Metabolism. *McGraw-Hill Med. Publishing division*. 11ed: 97-119.
5. Guengerich, F. P. Cytochrome P450 and chemical toxicology. *Chem. Res. Toxicol*, 2008; 21: 70 – 83.
6. Hodgson, E., P. K. Das, T. M. Cho, et al. Phase 1 — Metabolism of toxicants and metabolic interactions, 2008.
7. In *Molecular and Biochemical Toxicology*, 4th ed., ed. R. C. Smart and E. Hodgson. Hoboken, NJ: John Wiley and Sons, 2008.
8. Koukouritaki, S. B., P. Simpson, C. K. Yeung, et al. Human hepatic flavin - containing monooxygenases 1 (FMO1) and 3 (FMO3) developmental expression. *Pediatric. Res.*, 2002; 51: 236 –243.
9. Larry, Engelking R. Textbook of Veterinary Physiological Chemistry. Hexose Monophosphate shunt, 2015; 3: 1015 – 1026.
10. LeBlanc, G. A. Phase II — Conjugation of toxicants. In *Molecular and Biochemical Toxicology*, 4th ed., ed. R. C. Smart and E. Hodgson. Hoboken, NJ: John Wiley and Sons, 2008.
11. Luzzato, L., Mehta, A., and Vulliamy, T. Glucose 6-phosphate dehydrogenase deficiency. In *The Metabolic and MolecularBases of Inherited Disease*, 2001; 8: 4517–4553.
12. Nelson, D. R. Comparison of P450s from human and fugu: 420 million years. *Arch. Biochem. Biophys*, 2003; 409: 18 – 24.
13. Notaro, R., Afolayan, A. and Luzzatto, L**.** Human mutations in glucose 6-phosphate dehydrogenase reflect evolutionary history. *FASEB J*., 2000; 14**:** 485–494.
14. Ortiz de Montellano, P. R. *Cytochrome P450: Structure, Mechanism and Biochemistry* .New York: Springer, 2004.
15. Richter, R., G. P. Jarvik, and C. E. Furlong Paraoxonase 1 status and substrate hydrolysis. *Toxicol. Appl. Pharmacol*, 2009; 235: 1 – 9.
16. Ritter, J. K. Roles of glucuronidation and UDP - glucuronosyltransferases in xenobioticbioactivation reactions. *Chem. Biol. Interact*, 2001; 129: 171 – 193.
17. Ross, M. K. and J. A. Crow. Human carboxylases and their role in xenobiotic metabolism. *J. Biochem. Mol. Toxicol*, 2007; 187–196.
18. Smart, R. C. and E. Hodgson. *Molecular and Biochemical Toxicology*, 4th ed. Hoboken, NJ: John Wiley and Sons, 2008.
19. Tukey, R. H. and C. P. Strassburg Human UDP - glucuronosyltransferases: Metabolism, expression, and disease. *Annu. Rev. Pharmacol. Toxicol*, 2000; 40: 581 – 611.
20. Vasiliou, V., A. Pappa, and D. R. Petersen. Role of aldehyde dehydrogenases in endogenous and xenobiotic metabolism. *Chem. Biol. Interact*, 2000; 129: 1 – 19.
21. Zeldin, D. C. and J. M. Seubert Structure, mechanism and regulation of cytochromes P450. In *Molecular and Biochemical Toxicology*, 4th ed., ed. R. C. Smart and E. Hodgson .Hoboken, NJ : John Wiley and Sons, 2008.