Biological oxidations

D'Eustachio, P., Jassal, B.

European Bioinformatics Institute, New York University Langone Medical Center, Ontario Institute for Cancer Research, Oregon Health and Science University.

The contents of this document may be freely copied and distributed in any media, provided the authors, plus the institutions, are credited, as stated under the terms of Creative Commons Attribution 4.0 International (CC BY 4.0) License. For more information see our license.

This is just an excerpt of a full-length report for this pathway. To access the complete report, please download it at the Reactome Textbook.

25/12/2022
Introduction

Reactome is open-source, open access, manually curated and peer-reviewed pathway database. Pathway annotations are authored by expert biologists, in collaboration with Reactome editorial staff and cross-referenced to many bioinformatics databases. A system of evidence tracking ensures that all assertions are backed up by the primary literature. Reactome is used by clinicians, geneticists, genomics researchers, and molecular biologists to interpret the results of high-throughput experimental studies, by bioinformaticians seeking to develop novel algorithms for mining knowledge from genomic studies, and by systems biologists building predictive models of normal and disease variant pathways.

The development of Reactome is supported by grants from the US National Institutes of Health (P41 HG003751), University of Toronto (CFREF Medicine by Design), European Union (EU STRP, EMI-CD), and the European Molecular Biology Laboratory (EBI Industry program).

Literature references


Reactome database release: 83

This document contains 4 pathways (see Table of Contents)

https://reactome.org
All organisms are constantly exposed to foreign chemicals every day. These can be man-made (drugs, industrial chemicals) or natural (alkaloids, toxins from plants and animals). Uptake is usually via ingestion but inhalation and transdermal routes are also common.

The very nature of many chemicals that make them suitable for uptake by these routes, in other words their lipophilicity (favors fat solubility) is also the main reason organisms have developed mechanisms that convert them to hydrophilic (favors water solubility) compounds which are readily excreted via bile and urine. Otherwise, lipophilic chemicals would accumulate in the body and overwhelm defense mechanisms. This process is called biotransformation and is catalyzed by enzymes mainly in the liver of higher organisms but a number of other organs have considerable ability to process xenobiotica such as kidneys, gut and lungs. As well as xenobiotics, many endogenous compounds are commonly eliminated by this process.

This mechanism is of ancient origin and a major factor for its development in animals is plants. Most animals are plant eaters and thus are subject to a huge variety of chemical compounds which plants produce to stop themselves being eaten. This complex set of enzymes have several features which make them ideal for biotransformation;

1. **metabolites of the parent chemical are usually made more water soluble so it favours rapid excretion via bile and urine**

2. **the enzymes possess broad and overlapping specificity to be able to deal with newly exposed chemicals**

3. **metabolites of the parent generally don’t have adverse biological effects.**

In the real world however, all these criteria have exceptions. Many chemicals are transformed into reactive metabolites. In pharmacology, the metabolites of some parent drugs exert the desired pharmacological effect but in the case of polycyclic aromatic hydrocarbons (PAHs), which can undergo epoxidation,
it results in the formation of an electrophile which can attack proteins and DNA.

Metabolism of xenobiotics occurs in several steps called **Phase 1 (functionalization)** and **Phase 2 (conjugation)**. To improve water solubility, a functional group is added to or exposed on the chemical in one or more steps (Phase 1) to which hydrophilic conjugating species can be added (Phase 2). Functional groups can either be electrophilic (epoxides, carbonyl groups) or nucleophilic (hydroxyls, amino and sulfhydryl groups, carboxylic groups) (**see picture**).

Once chemicals undergo functionalization, the electrophilic or nucleophilic species can be detrimental to biological systems. Electrophiles can react with electron-rich macromolecules such as proteins, DNA and RNA by covalent interaction whilst nucleophiles have the potential to interact with biological receptors. That's why conjugation is so important as it mops up these potentially reactive species.

Many chemicals, when exposed to certain metabolizing enzymes can induce those enzymes, a process called **enzyme induction**. The effect of this is that these chemicals accelerate their own biotransformation and excretion. The reverse is also true where some chemicals cause enzyme inhibition. Some other factors that alter enzyme levels are sex, age and genetic predisposition. Between species, there can be considerable differences in biotransformation ability which is a problem faced by drug researchers interpreting toxicological results to humans.

**Literature references**


**Editions**

<table>
<thead>
<tr>
<th>Date</th>
<th>Action</th>
<th>Author</th>
</tr>
</thead>
<tbody>
<tr>
<td>2008-05-19</td>
<td>Authored, Edited</td>
<td>Jassal, B.</td>
</tr>
<tr>
<td>2008-05-28</td>
<td>Reviewed</td>
<td>D'Eustachio, P.</td>
</tr>
</tbody>
</table>
Phase 1 - Functionalization of compounds

Location: Biological oxidations

Stable identifier: R-HSA-211945

Compartments: mitochondrial outer membrane, cytosol, mitochondrial matrix, endoplasmic reticulum lumen, mitochondrial inner membrane, endoplasmic reticulum membrane

Phase 1 of metabolism is concerned with functionalization, that is the introduction or exposure of functional groups on the chemical structure of a compound. This provides a 'handle' for phase 2 conjugating species with which to react with. Many xenobiotics are lipophilic and almost chemically inert (e.g. PAHs) so would not necessarily undergo a phase 2 reaction. Making them more chemically reactive would facilitate their excretion but also increases their chance of reacting with cellular macromolecules (e.g. proteins, DNA). There is a fine balance between producing a more reactive metabolite and conjugation reactions.

There are two groups of enzymes in phase 1 - oxidoreductases and hydrolases. Oxidoreductases introduce an oxygen atom into or remove electrons from their substrates. The major oxidoreductase enzyme system is called the P450 monooxygenases. Other systems include flavin-containing monooxygenases (FMO), cyclooxygenases (COX) and monoamine oxidases (MAO). Hydrolases hydrolyse esters, amides, epoxides and glucuronides.

Literature references


Editions

2008-05-19 Authored, Edited, Revised Jassal, B.
2008-05-28 Reviewed D'Eustachio, P.
Phase II of biotransformation is concerned with **conjugation**, that is using groups from cofactors to react with functional groups present or introduced from phase I on the compound. The enzymes involved are a set of transferases which perform the transfer of the cofactor group to the substrate. The resultant conjugation results in greatly increasing the excretory potential of compounds. Although most conjugations result in pharmacological inactivation or detoxification, some can result in bioactivation. Most of the phase II enzymes are located in the cytosol except UDP-glucuronosyltransferases (UGT), which are microsomal. Phase II reactions are typically much faster than phase I reactions therefore the rate-limiting step for biotransformation of a compound is usually the phase I reaction.

Phase II metabolism can deal with all the products of phase I metabolism, be they reactive (Type I substrate) or unreactive/poorly active (Type II substrate) compounds. With the exception of glutathione, the conjugating species needs to be made chemically reactive after synthesis. The availability of the cofactor in the synthesis may be a rate-limiting factor in some phase II pathways as it may prevent the formation of enough conjugating species to deal with the substrate or it's metabolite. As many substrates and/or their metabolites are chemically reactive, their continued presence may lead to toxicity.

**Literature references**


Aflatoxins are among the principal mycotoxins produced as secondary metabolites by the molds Aspergillus flavus and Aspergillus parasiticus that contaminate economically important food and feed crops (Wild & Turner 2002). Aflatoxin B1 (AFB1) is the most potent naturally occurring carcinogen known and is also an immunosuppressant. It is a potent hepatocarcinogenic agent in many species, and has been implicated in the etiology of human hepatocellular carcinoma. Poultry, especially turkeys, are extremely sensitive to the toxic and carcinogenic action of AFB1 present in animal feed, resulting in multi-million dollar losses to the industry. Discerning the biochemical and molecular mechanisms of this extreme sensitivity of poultry to AFB1 will help with the development of new strategies to increase aflatoxin resistance (Rawal et al. 2010, Diaz & Murcia 2011).

AFB1 has one major genotoxic metabolic fate, conversion to AFXBO, and several others that are less mutagenic but that can still be quite toxic. AFB1 can be oxidised to the toxic AFB1 exo 8,9 epoxide (AFXBO) product by several cytochrome P450 enzymes, especially P450 3A4 in the liver. This 8,9 epoxide can react with the N7 atom of a guanyl base of DNA to produce adducts by intercalating between DNA base pairs. The exo epoxide is unstable in solution, however, and can react spontaneously to form a diol that is no longer reactive with DNA. The diol product in turn undergoes base-catalysed rearrangement to a dialdehyde that can react with protein lysine residues. AFB1 can also be metabolised to products (AFQ1, AFM1, AFM1E) which have far less genotoxic consequences than AFB1. The main route of detoxification of AFB1 is conjugation of its reactive 8,9-epoxide form with glutathione (GSH). This reaction is carried out by trimeric glutathione transferases (GSTs), providing a chemoprotective mechanism against toxicity. Glutathione conjugates are usually excreted as mercapturic acids in urine (Guengerich et al. 1998, Hamid et al. 2013). The main metabolic routes of aflatoxin in humans are described here.
Literature references


Editions

<table>
<thead>
<tr>
<th>Date</th>
<th>Action</th>
<th>Author(s)</th>
</tr>
</thead>
<tbody>
<tr>
<td>2014-05-09</td>
<td>Authored, Edited</td>
<td>Jassal, B.</td>
</tr>
<tr>
<td>2014-05-22</td>
<td>Reviewed</td>
<td>D'Eustachio, P.</td>
</tr>
</tbody>
</table>