

Prediction of Drug Induced Liver Disease, Pre and Post Marketing

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ABSTRACT

Drug induced liver disease is one of the most important causes of drug withdrawals post marketing. The liver is an engine that creates and stores energy, metabolises and detoxifies chemicals through various pathways, each of which can be a target of liver injury. Corroboration with animal studies is not optimal; pre registration studies do not have sufficient numbers to identify injury causing drug candidates, making prediction of liver injury a challenging task. Efforts are on to identify potential hepatic injury causing molecules at the molecular assessment, preclinical, clinical and post marketing stages in a product lifecycle. This article reviews many of these efforts that are ongoing to predict drug induced liver injury

Key Words

Drug induced liver injury; Drug induced liver disease, translational research, MIP DILI, MAQC-II, DILIsym, Hy's law, Genotyping

Introduction

Liver is a large and very complex organ of the body. It undertakes multiple chemical processes and reactions while providing the body with processed nutrients and metabolising/disabling potentially toxic drugs and chemicals. It could also be injured in the process. An estimated 1000 drugs could cause toxicity. This is often difficult to identify as it mimics various forms of hepatic disease and also accounts for 50 % cases of liver failure. Drug Induced Liver Disease (DILD) and Drug Induced Liver Injury (DILI) are often used synonymously [1]. Over the past 50 years, DILD is the most common cause of withdrawal of drugs from the markets. Jaundice appears late in liver injury.

Zimmerman [2] observed that cholestasis or impaired bilirubin excretion was ominous in itself but if jaundice was also present,

it could have 10 to 50 percent mortality due to acute liver failure. This observation was made in the pretransplant days. Today, due to the availability of liver transplantation the mortality is lesser but still a cause of concern. This article reviews the many efforts ongoing to predict DILI.

AETIOPATHOLOGY

In DILI there could be hepatic cellular damage, obstruction to bile flow with or without jaundice, or a mixed presentation of cholestasis and hepatocellular damage. E.g. Isoniazid and Ketoconazole cause hepatitis while high doses of paracetamol lead to acute hepatic necrosis. Estrogenic and anabolic steroids could cause cholestasis and phenytoin and enalapril could cause both cholestasis and hepatitis [3, 4].

In India, Devi et al retrospectively screened 6302 case records for DILI. 25 cases were included in the study. There were 2 definite cases of DILI, with 20 probable and 3 possible cases. 22 cases in the first two groups were analysed. 51 years was the median age; with a male to female ratio of 1.7. Anti-tubercular drugs were responsible for 17 cases with

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cisplatin, metformin, low molecular weight heparin, chlorpromazine and leflunomide contributing one case each. Pattern of acute hepatitis was cell damage in 11, cholestasis in seven and both in four patients. 20 patients recovered, while 2 died [5].

CLINICAL FEATURES

DILI can occur with almost any drug even at low doses and is often unexpected and idiosyncratic. It could be an acute or a delayed reaction over weeks or months.

History of ingestion of a known hepatotoxic drug, the dose administered, duration of exposure and temporal relationship between exposure and presentation are the key elements in diagnosis. If other causes of liver disease like viral hepatitis B or C, alcoholic liver disease, non alcoholic fatty liver disease or metabolic disorders like haemochromatosis can be ruled out, lab signs of hepatitis with jaundice will be confirmatory.

Improvement after discontinuing the drug is further evidence. After stopping therapy, the raised hepatitis associated enzymes will reduce by half in case of hepatocellular injury. They would return to normal by 4 weeks but in cholestatic injury the recovery could take upto a few months [4].

CAN WE PREDICT DILI?

Attempts have been and continue to be made for prediction of DILI. But it is a very challenging task for the following reasons:

DILI has an incidence of about 1 in >10,000. So a standard phase III study fails to raise a suspicion of hepatic toxicity.

Drugs like tacrine, statins, aspirin or heparin cause elevation of (aspartate aminotransferase/alanine aminotransferase) AST/ALT more than 3 times the upper limit of normal (ULN) without increasing risk on DILI. Hence elevated hepatic enzymes cannot be a marker.

In most people the liver can adapt to new drugs, and develop tolerance as in case of isoniazid. Hence a negative rechallenge is also not an indicator [2, 4].

In case of hematological, gastrointestinal and cardiovascular systems, animal studies can predict liver toxicity in humans with a certainty of 91%, 85% and 80% respectively. This figure is as low as 55% for hepatic disease [6].

Many new ideas are being tried to enhance the predictability of DILI. Development of new DILI biomarkers, introduction of high content screening, adoption of more sensitive animal models, and utilization of toxicogenomics are but a few of them [6].

Prediction of DILI is being tried with some amount of success at all stages of drug development.

PRECLINICAL STAGE

Translational Research

Translational science utilizes clinical or animal data to open doors in drug discovery. Lui Z et al [6] have proposed that the side effects observed in clinical trials and post-marketing surveillance could be utilised in drug discovery.

Thirteen types of liver injuries were identified from clinical and post marketing data. A quantitative structure-activity relationship (QSAR) model was developed for every side effect. An in silico model was prepared for each purely based on the structure. All these models were combined into a prediction system. The positive predictive value of this system for DILI in humans was 91%. This DILI predictive system could be used to screen and shortlist new drug candidates or chemicals.

MIP-DILI

Creating models to predict DILI in early development is a highly capital intensive and long drawn task which even large pharma would hesitate to take up on their own. That a collaborative effort is necessary is obvious. Under the Innovative Medicines Initiative (IMI), many large pharma companies, research institutions, NGOs, public bodies and gene research companies from Europe have come

together to start the MIP-DILI (Mechanism-Based Integrated Systems for the Prediction of Drug-Induced Liver Injury)

The team will do an in depth study of the science behind DILI. They will focus on current and new laboratory systems including cultures of human cells in both one dimensional and three dimensional configurations. The project will develop models to address interindividual differences. Due to a lack of human liver cells available to researchers, the group will use induced pluripotent stem cells derived from individuals susceptible to DILI in the modelling. The project will also develop computer models to unravel the complex and interrelated mechanisms behind DILI [7].

MicroArray Quality Control Phase-II (MAQC-II)

Food and Drug Administration has led the MAQC-II effort to develop and validate predictive signatures in DILI. The purpose of the project is to identify genomic biomarkers for liver injury. The project uses gene expression data from two tissues, blood and liver, to test cross tissue predictability of genomic indicators to a form of chemically induced liver injury. The genomic indicators from blood are then used as biomarkers for prediction of drug induced liver injury. Rats were chemically stressed to cause liver necrosis. Necrosis is often due to induction of apoptosis, mitochondrial damage, severe immune response and angiogenesis. The gene expression data acquired from the blood of rats related to these routes of liver necrosis. Assay of peripheral blood for the identification of novel biomarkers of DILI may be a useful diagnostic test in the near future [8].

Multiparametric assay on mouse liver mitochondria

Drug-induced mitochondrial dysfunction can cause several types of hepatotoxicity including cytolytic hepatitis, microvesicular steatosis, steatohepatitis, liver failure, and even cirrhosis. By inducing mitochondrial liability,

compounds can also damage other tissues, such as skeletal muscles, heart, and pancreas. Thus it is essential to screen a compound for its actions on mitochondria.

Authors developed a high-throughput screening platform using isolated mouse liver mitochondria. The broad spectrum multiparametric assay was designed to detect the dysfunctions in mitochondrial systems like global mitochondrial membrane permeabilization (swelling), inner membrane permeabilization (transmembrane potential), outer membrane permeabilization (cytochrome *c* release), and alteration of mitochondrial respiration. About 124 marketed drugs with known DILI potential were screened with this system and it showed a high positive predictive value. Authors believe that this system could be used to evaluate both marketed as well as under development molecules [9].

DILI Sym

Major pharma companies along with The Hammer Institutes and FDA have developed a model for DILI to be shared by the industry during its preclinical phase to eliminate DILI causing molecules. DILIsym® is a multi-scale representation of drug-induced liver injury. It has models of key liver cell like hepatocytes and Kupffer cells, intracellular biochemical systems representing mitochondrial dysfunction, and a whole body system to test drug distribution and metabolism. It simulates physiological data for mice, rats, dogs, and humans. It can also simulate a population of patients and take care of interindividual variations too [10].

Clinical stage

Hy's law

During clinical studies indications of possible liver injury are often seen in stray cases in the form of raised liver enzymes or raised bilirubin. Although this data is a pointer, it is not conclusive evidence of possible liver injury.

In this regard the observation by Zimmerman, mentioned earlier gains significance. On their own, raised transferases or raised bilirubin alone may not be so serious; but being present together could be dangerous.

Since the liver has a large excess of bilirubin-excreting capacity, injury to hepatocytes sufficient to cause jaundice or even mild hyperbilirubinemia (i.e., a bilirubin >2xULN) represents an extent of liver injury so great that recovery may not be possible in some patients. Hy's law is a translation of Zimmerman's observation into understandable numbers. After reviewing all clinical data of a product before marketing, one can identify Hy's law cases.

Guidance for industry Drug Induced Liver Injury, by FDA describes Hy's law cases as having the following components

"1. A higher incidence of 3-fold or greater elevations above the ULN of ALT or AST than the (nonhepatotoxic) control drug or placebo

2. Among trial subjects showing such AT elevations, one or more also show elevation of serum TBL(total bilirubin level) to >2xULN, without initial findings of cholestasis (elevated serum Alkaline Phosphatase,ALP)

3. No other reason can be found to explain the combination of increased ALT and TBL, such as viral hepatitis A, B, or C; pre-existing or acute liver disease; or another drug capable of causing the observed injury"

Finding one Hy's Law case in the clinical trial database is worrisome; finding two is considered highly predictive that the drug has the potential to cause severe DILI when given to a larger population.

Dilevalol showed 2 cases in 1000 exposures hence was not marketed in US. Later, post marketing studies in Portugal revealed fatal liver injury.

Tasosartan showed only one case per 1000, hence a larger premarketing database was asked for. Drug was later abandoned [2].

Post marketing stage

A number of drugs associated with liver injury clinically show a rise in transaminases indicating hepatocellular disease, rise in alkaline phosphatase and gamma-glutamyl transferase indicating cholestasis or a mixed phenotype. A very strong association with HLA (Human Lymphocyte Antigen) alleles indicates an immune aetiology even though there is no clinical support in the form of fever, eosinophilia, and an acute onset of liver injury.

A recent study which utilised a genome wide approach, detected a strong link between the rs2395029, the tag SNP (Single nucleotide Polymorphism) for HLA-B*5701 in the HCP (Hybrid Cluster protein) gene, and flucloxacillin-induced hepatotoxicity. HLA-B genotyping found that all patients with the rs2395029 polymorphism were also positive for HLA-B*5701. The positive predictive value (PPV) of such a test depends on the prevalence of the disorder. A low PPV in this case, would mean that approximately only 1 in 500-1,000 individuals positive for HLA-B*5701 would develop liver damage if exposed to flucloxacillin., pre-treatment genetic testing is not of clinical value in this case. However, where there are competing aetiologies the use of HLA-B*5701 to confirm the diagnosis of flucloxacillin-induced cholestatic hepatitis should be considered.

Another finding is that patients who are carriers of HLA-DRB1*1501 alleles are at risk of developing toxicity to amoxicillin clavulanic acid [11].

Most drugs are metabolized in the liver via the Cytochrome P 450 enzyme system. Genotyping can segregate individuals into fast or slow metabolisers and if doses are adjusted accordingly, side effects can be avoided. In the RAPID GENE trial patients were typed for CYP2C19*2 allele associated with lack of efficacy for Clopidogrel. Patients were randomized to a standard 75 mg/day arm or a genotype guided 10 mg/day arm. The genotype guided arm showed less adverse events [12].

Molecular-targeted cancer agents like imatinib, gefitinib and erlotinib are metabolized in the liver via the cytochrome pathway. Hepatotoxicity is linked to serum concentrations of drugs, such as imatinib. When imatinib is taken with a CYP3A4 inhibitor such as roxithromycin the severity of toxic effects can be increased due to higher serum concentrations. Clinicians should be aware of drug-drug interactions, especially with CYP3A4 and CYP3A5 inducers or inhibitors and drugs metabolized by these enzymes [13].

Classification of molecules involved in DILI

Chen et al utilized package inserts of 287 already approved drugs to create a classification system. It is difficult to gather all published data for a molecule but in a package insert any reaction whether causal or not, has to be mentioned by law. Hence it is the most comprehensive. To consider a drug of no DILI concern the criteria used was 10 years of marketing. This yielded a list of 65 drugs most of which were in the market for over 25 years. The authors believe that the method is transparent and reproducible. It could be used to study the DILI of marketed drugs for supporting drug discovery and biomarker development [14].

Conclusion

DILI accounts for the maximum number of product withdrawals after marketing. The liver is the main engine for assimilation, metabolism, and excretion acting through multitude of physiological processes. Any of these could be a target and a cause of liver injury. In spite of the various methods of screening molecules for DILI at every stage of development, many of them still skip the nets and lead to post marketing drug withdrawals. This is yet an evolving science and we will see many more collaborative efforts worldwide to tackle this issue.

References

- [1] Kalpowitz N, Drug Induced Liver Injury, CID.38,(2004),S44.
- [2] Guidance for industry Drug Induced Liver Injury: Premarketing Clinical Evaluation, US Dept. of health and Human Sciences, FDA (2009).
- [3] <http://livertox.nih.gov/drugliverinjury.html>, last accessed Feb. 2014
- [4] Sovani V, <http://www.pharmawisdom.com/Drug-Induced-Liver-Disease.html>, last accessed July 2014
- [5] Devi D P, Guido SS, Drug induced serious hepatic adverse reactions in a tertiary care centre: A five-year retrospective analysis, Indian J Med Sci.58,(2004),211-2.
- [6] Liu Z, Shi Q, Ding D, Kelly R, Fang H, Tong W, Translating Clinical Findings into Knowledge in Drug Safety Evaluation - Drug Induced Liver Injury Prediction System (Delis), Plopro Computational Biology. 7,(2011) , 12 ,e1002310.
- [7] <http://www.mip-dili.eu/index.php?page=project#1>, last accessed September 2014
- [8] Huang J, Shi W, Zhang J, Chow JW, Pales RS, Garish K et al, Genomic indicators in the blood predict drug-induced liver injury, The Pharmacogenomics Journal, 10, (2010), 267-277.
- [9] Porceddu M, Buron N, Roussel C, Labbe G, Fromenty B, Borgne-sanchez A, Prediction of Liver Injury Induced by Chemicals in Human With a Multiparametric Assay on Isolated Mouse Liver Mitochondria, Toxicological Sciences 129, (2012),332-345.
- [10] <http://www.dilisym.com/Products-Services/dilisym.html> last accessed August 2014
- [11] Alfievic A, Pirmohamed M, Drug Induced Hypersensitivity and the HLA

Complex,Pharmaceuticals, 4,(2011), 69-90

- [12] Carr DF, Alferivic A, Pirmohamed M, Pharmacogenomics: Current state of art.Genes, 5,(2014),430.
- [13] Medscape Multispeciality. Faculty and Disclosures ,
http://www.medscape.org/viewarticle/571554_3, Last accessed September 2014
- [14] Chen M, Vijay V, Shi Q,Liu Z,fang H, Tong W, FDA-approved drug labelling for the study of drug-induced liver injury, Drug Discov Today,05(2011), 007.