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Hepatotoxicity and Antioxidants: An overview

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ABSTRACT

The liver is responsible for several functions in the body including performing and controlling the homeostasis as well as nutrient and energy supply, metabolism and growth. It activates and inactivates exogenous and endogenous xenobiotics due to it has a defense mechanism but this sometimes inadequate, resulting in pathological conditions that influence the function and structure of the liver compartment leading to pathophysiological changes which may results in hepatotoxicity. It is the main reason for withdrawing of some drugs from marketing. Antioxidants are the substances those are present at low levels and significantly delay or prevent oxidation of the oxidizable substrate. They are one solution for controlling the hepatotoxicity. It is necessary to obtain a comprehensive article giving us overview information about hepatotoxicity and antioxidants relationship.

Keywords: Hepatotoxicity – Antioxidants - Pathophysiological changes

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1. INTRODUCTION

Liver is the main internal organ and gland amongst the most important and complex organs, representing about 2.5% of an adult body weight. It's formed from parenchymal cells (hepatocytes) and non-parenchymal cells (Sinusoidal endothelial cells, kupffer cells and hepatic stellate cells). It occupies the upper right part of the abdominal cavity and underneath the diaphragm. It is connected to hepatic artery and hepatic portal vein. In fact, during rest it receives about 25% of the cardiac output through hepatic artery and hepatic portal vein. The hepatic portal vein carries the absorbed blood containing nutrients, toxins, and other substances from the gastrointestinal tract (duodenum) to the liver. The liver filters this blood, after that sends it via the hepatic vein to the heart in order to pump it all over the body [1].

It often performs a central function in more than 500 bodily functions via oxidation, reduction, hydration, hydrolysis, condensation, conjugation, or isomerization [2, 3]. Scientists suppose that, the liver is related to every dysfunction or disease and acts as the laboratory of the body [4]**.** Actually, it is the chief site for intense biotransformation and excretion [5]. Liver adjusts production, storage, and release of fats, sugar and cholesterol. It stores numerous nutrients, including vitamins A, D, B_{12} and K along with iron and copper and converts iron into heme according to body requirements. Besides, it synthesizes several important hormones (angiotensinogen, insulin-like growth factor1, thrombopoietin and glycoprotein),clotting factors (fibrinogen, prothrombin and antithrombin), enzyme catalase and immune factors [6,7]. Moreover, hepatocytes produce and excrete bile fluid (consists of water, bile salts, cholesterol, a phospholipid called lecithin, bile pigments and several ions) that helps digestion of fats, absorption of fat-soluble nutrients and serves as a filter that detaches out toxic substances from blood stream [8].

In addition hepatic cells are imperative for the metabolism, detoxification and activation of endogenous (reactive oxygen/nitrogen species, saturated fatty acids, cholesterol, oxysterols, isoprostanes, eicosanoids and lipopolysaccharide) and exogenous substances (over the counter medications, prescription medications, parasites, bacteria, viruses, heavy metals, pesticides and food preservatives) in existence of various enzymes. Actually, the essential role played by liver in the transformation and clearance of toxins exposes it to hurt and induce hepatotoxicity[9, 10].

Agents which cause hepatotoxicity are named hepatotoxinsor hepatotoxicants[7]. Any damage to liver or disturbance of its functions might result in many implications on one's health [11]. The leading cause of drug

non-approval and drug withdrawal by the Food and Drug Administration (FDA) in the US is drug-induced hepatotoxicity [9, 12]. More than a thousand medicines and chemicals have been accounted to cause liver injury [13]. Drug-induced liver injury may account for about 10% of all cases of acute hepatitis, 5% of all hospital permissions, and 50% of all acute liver failures [12]. It is remarkable that more than 75% of cases of idiosyncratic drug reactions result in liver transplantation or death [9]. Drug-induced liver injury is a somewhat common cause of acute liver disease and carries a mortality of around 10% [14]

So, maintainance of a healthy liver is a vital factor for the overall health and wellbeing [15]. According to Ghori et al. hepatotoxicants damage hepatocytes mainly by inducing lipid peroxidation and further oxidative damages in the liver due to chronic treatment or toxic doses resulting in death [16]. The liver is a chief target organ of the toxicity of xenobiotics, drugs, and oxidative stress resulting from its association to the gastrointestinal tract and unique metabolism [10]. The gut–liver relationship plays an important part in drug-induced liver injury owing to increasing the production of cytokines, arachidonic fatty acid metabolites and reactive oxygen species (ROS) in the hepatocytes resulting from increasing of hepatic endotoxin influx [17]. Thus, liver injury and disease are still the foremost global health problems[18]. There are different factors that improve a person's susceptibility to a potentially hepatotoxic drug [14] and they are represented in Figure (1).

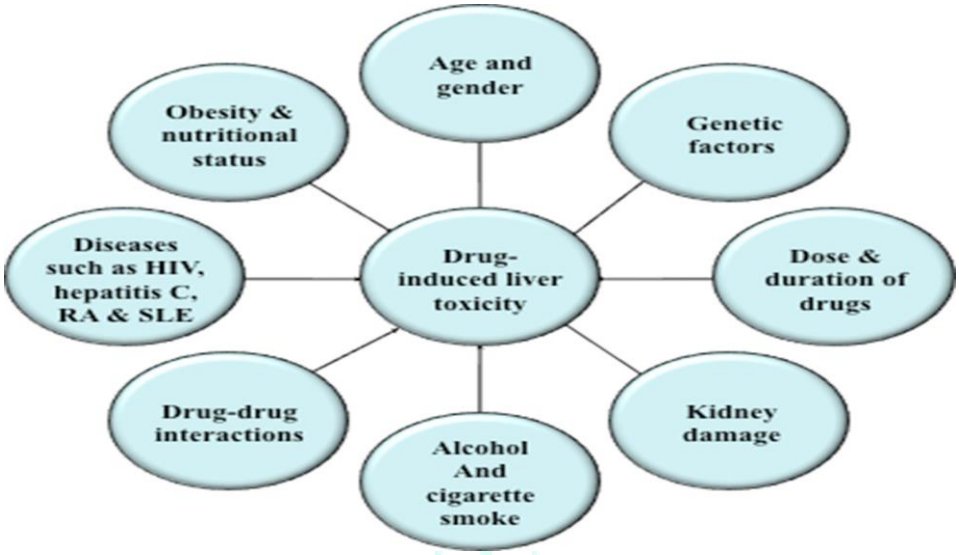

Figure 1: Factors affecting drug induced hepatictoxicity.

2. Pathogenesis of the hepatotoxicity

Drugs and xenobiotic in the liver are in concentrated form because around 75% blood which enters liver by portal vein comes from spleen and gastrointestinal canal. The basic role of the liver is converting of the chemical substance from lipophilic form to hydrophilic form to be easily excreted. Enzymatic metabolism in the liver carried out by cytochrome P-450, the main metabolizing enzymes system which is situated in the endoplasmic reticulum of the hepatocytes [19]. Liver damage results from synthesis of reactive metabolites from drugs. They have an ability to interact with macromolecules of hepatocyte for instance nucleic acids, lipids, proteins and carbohydrates leading to oxidative stress due to DNA damage, lipid peroxidation and protein dysfunction [20].

2.a. Mitochondria

Mitochondria are multifunctional organelles liable mainly for production of energy in the form of ATP from ADP in what is called oxidative phosphorylation [21], and mitochondrial role involved in intracellular stress cascades, apoptosis and necrosis or death[22, 23]. Mitochondria of the cell carry the electron transport chain that reduces oxygen to water by addition of electrons during oxidative phosphorylation. Mitochondria are the basic source of endogenous ROS. It is elicited that the production of endogenous ROS within human mitochondrion is about 10^7 molecules/day throughout normal oxidative phoshorylation[24]. ROS induce more persistent damage to mitochondrial DNA than to nuclear DNA [25]. High levels of intracellular ROS related to depletion of GSH, oxidation or alkylation of protein, binding to enzymes or nucleic acids, peroxidation of lipid, and respiratory chain changes are accompanied by mitochondrial dysfunction. These events are the major reasons for liver diseases and drug induced liver injury [26]. Moreover, parent drug or drug reactive metabolites, after the biotransformation with Phase I (oxidation, reduction, hydroxylation and de-methylation) or Phase II (conjugation of chemicals conjugate with hydrophilic moieties) enzymes, can provoke hepatitis through immune reactions or direct toxicity causing mitochondrial membrane disruption [27].

Drug or its reactive metabolites covalently bind to human leukocyte antigen (HLA) proteins as a carrier forming hapten, which is exposed to cytotoxic T-Lymphocytes and recognized as antigens. Antigens are presented on antigen presenting cells to stimulate synthesis of antibodies against themselves or initiate the specific immune system forming auto-antibodies against cell organelles [28, 29]. Danger signals are necessary for immunological reactions [30]. These signals initiate signal pathways for oxidative stress resulting in immune-mediated hepatocyte damage. The signal may be a separate drug, viruses, bacteria or a cytokine results from an inflammatory reaction [31, 32].

2.b. Endoplasmic reticulum

Endoplasmic reticulum (ER) is a main intracellular organelle in controlling of protein synthesis, folding, modification, and trafficking. In addition to these, the ER has a critical function in calcium maintenance and in regulating carbohydrates, steroids and lipids biosynthesis[33]. In the ER lumen the

concentration of oxidized glutathione (GSSG) is more than reduced glutathione (GSH) creating oxidizing environment that promotes formation of disulfide bond [34]. ER is the major site for producing of ROS from formation of disulphide bond through oxidative protein folding [35]. Excessive accumulation of folding protein results in consumption of reduced glutathione owing to unstable disulphide bonds[36].Leakage of calcium from ER in response to oxidative injury and stress, resulting in aggregation of calcium in mitochondria stimulating mitochondrial ROS [37]. Notably, several pathological and physiological conditions can suppress normal protein folding leading to disturbances of ER homeostasis such as redox disturbances, glucose starvation, energy depletion, alterations of ER luminal calcium stores, viruses, xenobiotics, lipid accumulation as well as ethanol intoxication [38-40] ER suppress results in stimulation of unfolded protein response (UPR), activates autophagy and proteasome-dependent proteolysis[41-43].

2.c. Lysosome

Lysosomeis a membrane-enclosed spherical cytoplasmic vesicles containing acidic (pH 4-5)hydrolytic lysosomal enzymes, theirmain roles arerecycling and disposal of worn-out and damaged cytoplasmic macromolecules and organelles (degradation of polysaccharides, proteins, and complex lipids into their corresponding structural units respectively: monosaccharides, amino acids (AAs), and free fatty acids [44, 45]. The byproducts of lysosomal degradation, via specialized exporters [46] or via vesicular membrane diffusion for new biosynthesis pathway or energy metabolism [47]. In addition to the digestion of extracellular and foreign materials transmitted to them by phagocytosis, endocytosis, or autophagy[48, 49], found in all animal cells except red blood cells[50].

Over 60 hydrolytic luminal lysosomal enzymes (proteases, sulfatases, phosphatases, glycosidases, lipases, and nucleases) perform the digestion of the load and extra lysosomal roles such as apoptosis, repair of cell membrane, and tissue remodeling [51, 52, 53, 54]. Iron accumulation is the main source of oxidative stress in the hepatocytes, since hepatocytes and Kupffer cells are the main cell types exposed to iron storage in the liver. Iron is a main constituent of iron-sulfur containing enzymes, oxygen transport systems, and oxygen sensing proteins. The major site of iron storage are lysosomes, through transferrin mediated endocytosis goes into the hepatocyte, thenacidification of endosome, reduction of iron, and liberation from transferrin escaping into the cytosol.Notably, aggregation of Fe^{2+} is responsible for formation of ROS having a critical role for cellular compartment damage leading to injuries of cells and organs.

Virtually, liver diseases, as human diseases, are related biochemically to ferritin level elevation in serum resulting from hepatocyte damage. Raised ferritin uptake into hepatocytes induces biological disturbances and promotes lipid peroxidation, reduction of GSH/GSSG ratio and depletion of GSH. Ferritin accumulation stimulates macroautophagy which is completed by chelation of Fe [55]. Increased lysosomal membrane permeability results in escaping of Fe^{2+} into the cytosol, in addition, reduction of Fe^{3+} by superoxide, supportsthe synthesis of ROS enhances peroxidation of cell membrane and cellular damage [56].

3. Hepatotoxicants

1- Chemical-induced hepatotoxicity includes carbon tetrachloride, thioacetamide, diethylnitrosamine, aflatoxin B1, bromobenzene, lithocholic acid, acryl amide, acrolein, alpha-naphthylisothiocyanate, and d-galactosamine. The products of the metabolism of these chemicals result in hepatocytes damage by cell shrinkage, nuclear apoptosis or swelling and necrosis due to direct effect on nucleus, mitochondria, cytoskeleton, and ER [57].

2- Drug-induced hepatotoxicity, these drugs establish their toxicity either by direct effect cellular organelles (non- steroidal anti-inflammatory drugs (diclofenac), paracetamol, anti-cancer, Azathioprine, and Adriamycin) or indirect by mediators (ranitidine, rifampicin, isoniazid, pyrazinamide, erythromycin, halothane, and tamoxifen) [58-61].

3- Metal-induced hepatotoxicityencloses members such as Mercury, Cadmium, and Lead [62- 64].

4- Phytotoxin induced hepatotoxicity implicates harmful substances as Phallotoxin, Microcystine, and Pyrrolizidine alkaloids [65, 66].

5- High fat diet induces hepatotoxicity due to stimulation of steatosis [67].

4. Hepatotoxicity biomarkers

A- Hepatocytes injury biomarkers

1-Alanine aminotransferase (ALA)

In fact, ALT is completely cytoplasmic enzyme which catalyzes the reaction of transamination. It is present in various body tissues (kidney, heart, and muscle) andplasma, but is available in large quantities in the cytosol of hepatocytes compared with other tissues[68]. ALT was previously known as serum glutamate (glutamic)-pyruvate (pyruvic) transaminase [69]. ALT is responsible for transferring of an amino group from L-alanine to α-ketoglutarate, in reversible reaction forming pyruvate and L-glutamate.

L-alanine + α-ketoglutarate⇌pyruvate + L-glutamate

Actually, ALT is widely used as a serum biomarker of hepatocellular injury, but can be elevated due to injury of heart tissue or skeletal muscle, or in response to some medications that do not cause liver injury however; ALT is estimated clinically as a part of a diagnosis of hepatocellular injury. Therefore, other serum hepatotoxicity biomarkers are required[70,71]. Clear elevations of ALT levels in blood serum observed usually in persons with toxin-induced liver injury[72].

2-Aspartate aminotransferase (AST)

Regarding AST, it is a mitochondrial and cytoplasmic enzyme mainly present in the liver, heart, skeletal muscles, kidney, pancreas, erythrocytes, lungs and brain tissues. It participates in the transamination (metabolism) of aspartate **[73]**. Levels of serum AST, after cell damage, elevate within eight hours and reach the peak at 24-36 hours, then return to the normal rangewithin three to seven days. Notably, chronic cell injury results in levels of AST stayrose. AST is used with other enzymes for monitoring the causative agent of liver diseases, for example, with ALT for diagnosis of liver cirrhosis and alcoholic hepatitis [74]. Marked elevation of mitochondrial AST observed in severe chronic liver degeneration and necrosis [75]. The ratio between mitochondrial AST and total AST activity has an important diagnostic value in differentiation between alcoholic hepatitis and hepatocytes necrotic type [76].

B-Cholestasis indicators

1- Alkaline phosphatase (ALP)

Essentially, ALP is a membrane-binding enzyme in mucosal epithelial cells of small intestine, liver, proximal convoluted tubules of kidney, placenta, and bone. It is responsible for lipid transportation in the intestine and bone calcification in bone. ALP stimulates certain reactions particularly, phosphate group hydrolysis from an organic molecule at an alkaline media [77, 78]. If ALP is present in high concentration in blood serum, this refers to liver or bone diseases or tumor [79]. Generally, the lining cells of the bile ducts produce the ALP as a primarily enzyme in cholestatic diseases due to granulomatous inflammation, tumor, benign familial hyperphosphatasemia, pancreatitis, abscesses, pregnancy (3rd trimester), osteomalacia, and duodenitis resulting in ALP evading into blood stream [80]. Alcoholic hepatitis plus drug or xenobiotic hepatitis induced increase of ALP levels attain to three times more than the normal range [81].

2- γ-glutamyltranspeptidase (γGT)

Actually, γGT is microsomal, cytoplasmic enzyme found in liver cells, epithelial lining cells of the bile duct, intestine, renal tubules and pancreas as well as bound to membranes. It takes part in transferring of amino acids across the cytoplasmic membrane and in metabolism of glutathione. It splits glutamyl groups and carries them to peptide chains and receptors. It is indicator for cholestasis. It may be used in combination with ALP for diagnosis of hepatobiliary and bone disease. γGT could be related to metabolism of glutathione. It intercedes the absorption of glutathione from extracellular to intracellular portion which is an important component of cells against oxidative stress [82]. It is an early biomarker of oxidative stress while; serum antioxidant carotenoids (β-carotene, α-carotene, β-cryptoxanthin, and lycopene) are correlated with heavy and moderate alcohol drinkers[83]. Levels of serum γGT reach higher than 10 times of the upper values. It is partially regarding to hepatic injuries, induction of liver microsomal enzymes or pancreatic damages due to alcoholism [84].

3 -5′ Nucleotidase [5'-NT]

Indeed, 5′-Nucleotidase (5′-NT) is an essential membrane glycoprotein enzyme in a large variety of body cells. It stimulates the hydrolysis reaction of the phosphate group of 5′-nucleotides, leading to identical nucleosides. In spite of 5′-NT is widely distributed all over the human body, its marked elevations of 5′-NT are practically necessary for identifying biliary disorders, hepatitis, cholestatic liver disease, liver malignancy, biliary cirrhosis and intrinsic liver damage[85].

C-Biomarkers of the ability of the liver to transport organic anions and biotransformation of drugs

1- Serum bilirubin

Bilirubin (BR) is an endogenous anion results from degradation of haemoglobin within the macrophage or reticuloendothelial system, produced in unconjugated form which directs into the hepatocytes altering it by UDP-glucuronyl transferase to conjugated BR[68]. BR in body is a sensitive equilibrium between the pigment synthesis and removal in body. The clinical sign of Jaundice appears when BR be observable in mucous membranes, the skin, and sclera resulting from over production, and declined hepatic uptake or conjugation of RB or both [86, 87]. The elevated levels of the conjugated BR are seen in hepatocellular damage, viral hepatitis, and ischemic or toxic liver injury [75].

2- Urine bilirubin

Bilirubin in urine refers to hepatobiliary disorder. Normally, unconjugated form of bilirubin is strongly tied with albumin protein therefore; it is not excreted by the glomerular filtration resulting in unconjugated bilirubin not found in urine. The detectable quantity of bilirubin in blood serum is specific to hepatobiliarry disease. Notably, the renal threshold for conjugated bilirubin is small, so this, urine conjugated bilirubin form may be detected though the serum bilirubin levels are normal [88, 89].

3- Urobilinogen

Hepatocellular dysfunctions for instance alcoholic liver injury and malignant disease or well compensated cirrhosis of the liver are well detected with an raise in the urobilinogen in urine [90, 91].

D- Biomarkers of the liver's biosynthetic ability

1-Albumin

Noteworthy, albumin is the highest plasma protein synthesized in the liver and is the indicator for the hepatic efficiency as well as it is synthesized lone in the liver [92].Albumin synthesis is stimulated via thyroid hormone and Corticosteroids [93]. In addition, its synthesis is controlled by liver disease, hormonal equilibrium, nutritional status, and osmotic pressure. The serum albumin readings are decreased in cirrhotic and ascetic patient [94].

2-Prealbumin

Measurement of prealbumin (PA) protein particularly has benefits in diagnosis of drug-induced liver injuries [75]. PA is a form of plasma protein formed by the liver cells. Its biological half-life is short. The basic role of PA is transportation of vitamin A and thyroxine hormone for maturation of lymphocytes in order to stimulate resistance of the body [95]. The levels of PA are indicator and correlated with hepatocytes harm [96, 97].

Noteworthy, in biology system, oxygen based radicals and nitrogen based radicals are two types of free radicals. Free radicals are atoms or molecules that have unpaired electrons, usually unstable and very reactive [98]. Oxidative stress, termed as an inequality between production and elimination of ROS leading to plural oxidative modifications of basic and regulatory methods, can be caused in dissimilar ways. ROS is a combined term used for a group of oxidants, which are either free radicals or molecular species talented of generating free radicals Table (1)[99]. An oxygen molecule (O_2) undergoes four-electron decrease when it is metabolized *in vivo*. During this process, reactive oxygen metabolites are created by the excitation of electrons secondary to addition of energy or interaction with transition elements. The reactive oxygen metabolites thus produced are more highly reactive than the original oxygen molecule and are called active oxygen species. In fact, superoxide ($O_2^{\cdot-}$), hydrogen peroxide (H_2O_2), hydroxyl radicals ($OH^{\cdot}$), and singlet oxygen (O_2) are the main examples of active oxygen species [100]. Indeed, ROS cause protein unfolding, collagen disintegration, lipid breakdown, DNA damage and fragmentation [101].

Table (1): Reactive Oxygen Species (ROS); List of radicals and non-radicals molecules.

Radicals	Non radicals
OH·(Hydroxyl radical)	O_2 (Singlet oxygen)
O_2·-(Superoxide radical)	H_2O_2 (Hydrogen peroxide)
RO_2·(Peroxyl radical)	HOCl (Hypochlorous acid)
RS·(Thyl radical)	O_3 (Ozone)
RO·(Alkoxyl radical)	LOOH (Lipid peroxide)
LOO·(Lipid peroxyl)	

Oxidative Stress in Liver Diseases

1. Oxidative Stress Caused by Alcohol

It was reported that alcohol consumption accounting for an estimated 3.8% of global mortality. Alcoholic liver disease (ALD) is one of the most important causes of liver-related death, which is related with increased dose and time of alcohol ingestion. ALD may progress from steatosis to further severe liver diseases form, such as hepatitis, fibrosis, and cirrhosis [102, 103]. Although pathogenesis of ALD has not been entirely elaborated, the direct effect of ethanol metabolism seems to be related to ROS production, mitochondrial damage and steatosis, which are the common features of acute and chronic alcohol exposure [104, 105, 106]. Studies have demonstrated that enzymatic as well as non-enzymatic systems which preserving cellular homeostasis is remarkably affected by alcohol in various models.

2. Oxidative Stress Caused by Drugs

The liver is the most frequently targeted organ in terms of drug toxicity. The production of radical species, specifically ROS and RNS, has been planned as an early occasion of drugs hepatotoxicity and as an indicator of hepatotoxic potential [107].It has been discovered that a lot of drugs could stimulate oxidative stress including augment of cellular oxidants and lipid peroxidation, diminution of antioxidants in the liver, such as anti- inflammation drugs, anti-analgesic drugs, anti-cancer drugs and antidepressants. For instance, sulfasalazine, a drug to treat inflammatory bowel diseases, has been found to induce hepatic oxidative damage [108]. Oral sulfasalazine administration could diminish superoxide dismutase (SOD) activity significantly. In addition, zoledronic acid is used to treat the cancer associated hypercalcemia. It has been revealed that zoledronic acid significantly elevated MDA and nitric oxide levels, while reduced GSH levels, which indicated that zoledronic acid could induce oxidative stress and decrease antioxidant level in liver [109]. Furthermore, paracetamol which is an broadly used analgesic compound in mice was evaluated by it induced a notable increase of MDA and nitrite as well as nitrate in the liver, with potent decrease of total SOD and Cu/Zn-SOD activity [110].

3. Oxidative Stress Caused by Environmental Pollutants

Environmental pollutants such as heavy metals and microcystin have been shown to cause oxidative damage in liver of animal models. Antioxidant defense system in rat liver was damaged after mercury chloride treatment [111]. The results showed that low dose of mercury could incur oxidative stress and hepatic damage. Besides mercury, lead was also found to exacerbate liver lipid peroxidation in protein-undernutrited rats, in which the study also suggested that free radical is a pathological mechanism for hepatotoxicity of lead[112].

4. Oxidative Stress Caused by Other Factors

Other factors such as radiation and temperature may also induce hepatic oxidative stress. The oxidative stress induced through exposure of mobile phone-like radiation has been inspected in the liver of guinea pigs [113]. The results showed that after radiation exposure, the levels of MDA and total nitric oxide were significantly increased and the activities of SOD and glutathione peroxidase (GPx) were reduced in the liver of guinea pigs. Additionally, the severity of oxidative damage was increased along with the period of radiation exposure. Furthermore, study observed that cold stress could lead to decrease in CAT, SOD and GSH-Px activities in rat liver when the rats were kept at 10° C for a week, which indicated that cold stress may cause hepatic damage which is associated with oxidative stress [114].

Moreover, the effect of ZnO_2 nanoparticles, a common cosmetic constituent, on cellular oxidative stress in mouse liver was investigated [115]. After exposure to ZnO_2 nanoparticles, capability of hepatic cells was decreased in concentration-dependent manner, and decrease in antioxidant enzyme levels as well as increase in DNA adduct. Actually, studies have suggested that maternal high-fat diet feeding could raise the incidence of metabolism-related diseases in offspring, including chronic liver disease. Zhang et al. found that maternal high-fat diet increased the level of plasma triglyceride and hepatic TBARS significantly [116]. The size of lipid droplets in the liver of rat offspring was also increased. In another study, the effect of high dietary salt on hepatic antioxidant defensing enzyme of fructose-fed rats was investigated [117]. Feeding fructose-fed rats with high-salt diet could trigger hyperinsulinemia and insulin resistance resulting in membrane perturbation. This potentially enhanced hepatic lipid peroxidation in the presence of steatosis, and led to decrease in antioxidant defenses, as observed by reduction of GSH, SOD and CAT activities. These results indicated that consumption of salt-rich diet by insulin-resistant subjects could lead to sodium reabsorption, which may aggravate hepatic lipid peroxidation related to damage antioxidant defenses.

Antioxidants and natural defense from ROS induced damages

Uncontrolled production of ROS can lead to their gathering causing oxidative stress in the cells. Therefore, cells have evolved defense mechanisms for protection against ROS mediated oxidative damage. These comprise antioxidant defenses to maintain a check on the generation of ROS. An antioxidant is a substance that is present at small concentrations and significantly setbacks or prevents oxidation of the oxidizable substrate. Antioxidants are effective as they can donate their own electrons to ROS and thus neutralizing the adverse effects of the latter. In general, an antioxidant in the body may work at three different levels: (a) prevention; keeping formation of reactive species to a minimum e.g. desferrioxamine (b) interception; scavenging reactive species either via using catalytic and non-catalytic molecules e.g. ascorbic acid, alpha-tocopherol and (c) repair; repairing damaged target molecules e.g. glutathione[118]. The antioxidant systems are classified into two major groups, enzymatic anti-oxidants and non-enzymatic antioxidants. Enzymatic antioxidants present in the body include SOD, catalase and GPx that act as body's first line of defense against ROS by catalyzing their conversion to less reactive or inert species (Figure 2) [119].

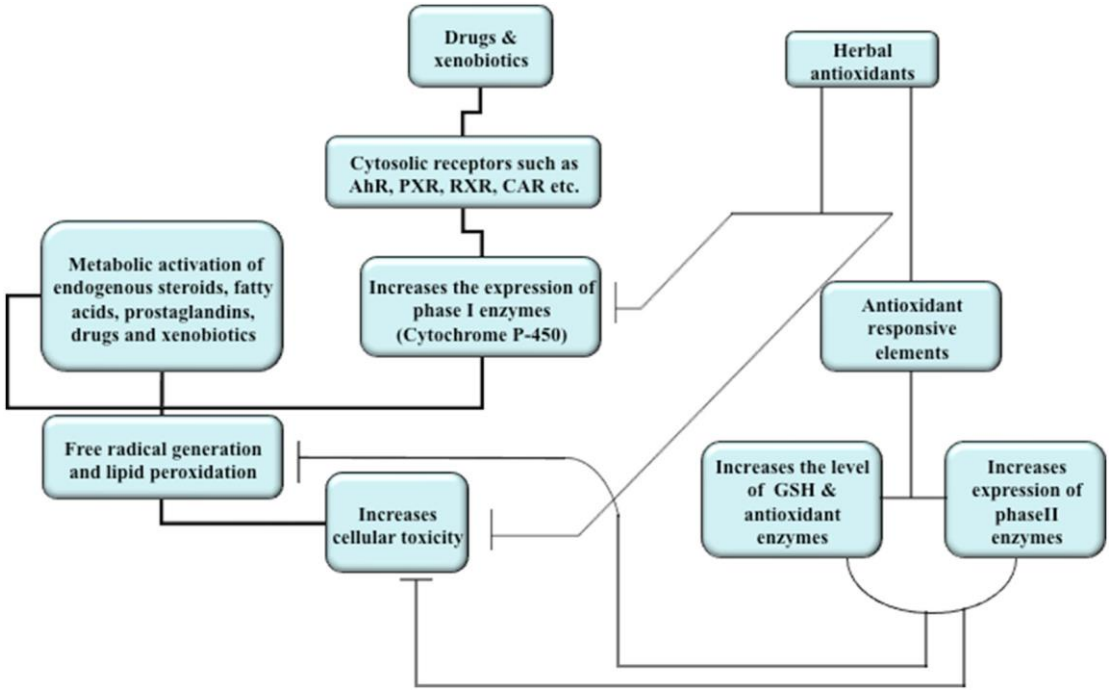

Figure 2. Overview of mechanism of drug and xenobiotic metabolism and effects of herbal antioxidants.

Several low molecular weight molecules present inside the cell provide secondary defense against free radicals and act as non-enzymatic antioxidants. A few examples of such molecules include GSH, α-tocopherol, ascorbate, bilirubin [118]. These agents either scavenge the ROS directly or prevent the production of ROS through sequestration of redox active metals like iron and copper [99].

Natural products as antioxidants

Natural products have shown great promise in combating against the toxicity of several commonly used drugs, including acetaminophen. Additionally, some of these natural products, such as resveratrol and curcumin, are now widely accepted chemopreventive agents. Due to easy availability and dietary nature, it is time to promote the natural products as supplementary medication with drugs that also cause toxicity to cells. Use of herbs in treatment of various liver disorders is common in China. Now these medicines are being gradually accepted worldwide, particularly in Asia, Europe and North America. Nevertheless, the application strategy may differ in East and West due to variety of reasons, such as, philosophical viewpoint, concept of diseases, and treatment approaches [14]. A variety of dietary plants including grams, legumes, fruits, vegetables, tea as well as wine contain antioxidants. The prophylactic properties of dietary plants have been attributed to the antioxidants / polyphenols present in them. Polyphenols with over 8000 structural variants are secondary metabolites of plants and represent a huge extent of substances having aromatic ring(s) bearing one or more hydroxyl moieties [99]. Polyphenols are effective ROS scavengers and metal chelators because of the presence of multiple hydroxyl groups. Examples of polyphenolic natural antioxidants derived from plant sources include vitamin E, flavonoids, cinnamic acid derivatives, curcumin, caffeine, catechins, gallic acid derivatives, salicylic acid derivatives, chlorogenic acid, resveratrol, folate, anthocyanins and tannins [120]. Apart from polyphenols there are also some plant derived non-phenolic secondary metabolites such as melatonin, carotenoids, retinal, thiols, jasmonic acid, eicosapentaenoic acid, ascopyrones and allicin that show excellent antioxidant activity [121,122]. Vitamin C, the water soluble natural vitamin, plays a crucial role in regenerating lipid soluble antioxidants like vitamin E. Both vitamin E and C are used as standards for evaluating the antioxidant capacity of new molecules [118]. Noteworthy, scientists suggested a crude classification of herbal drugs in two groups; firstly, the main ingredients, for example silybin, osthole, cumarin, glycorrhizin, flavonoids and so on; and secondly, the supporting substances like sugars, amino acids, resins, tannins, and volatile oil. To declare the subject, figure (2) illustrates mechanism of drug and xenobiotic metabolism and effects of herbal antioxidants [14].

Limitations of antioxidant supplementation

The primary concern regarding antioxidant supplementation is their potentially deleterious effects on ROS production (pro-oxidant action) especially when precise modulation of ROS levels are needed to allow normal cell function [123]. In fact, some negative effects of antioxidants when used in dietary supplements (flavanoids, carotenoids, vitamin C and synthetic compounds) have emerged in the last few decades [124]. Mechanistic investigation has revealed that antioxidants may exhibit pro-oxidant activity depending on the specific set of conditions. Of particular importance are their dosage, redox conditions and also the presence of free transition metals in cellular environment [125]. For example, ascorbate, a well-known antioxidant in the presence of high concentration of ferric iron is a potent mediator of lipid peroxidation. Recent studies recommend that ascorbate sometimes increases DNA damage in humans. Similarly β-carotene also can behave as a prooxidant in the lungs of smokers. Of note, natural antioxidant compounds have relatively poor bioavailability. It is therefore necessary to take into cognizance the bioavailability and differential activities of natural and synthetic antioxidant compounds before considering them as therapeutic or pharmacological agents [99].

CONCLUSION

Several mechanisms are involved in the initiation of liver cell damage and aggravate ongoing injury processes. Dysfunction

of these vital cell organelles results in impairment of dynamic equilibrium in homeostatic condition, thus resulting in intracellular oxidative stress with excessive formation of ROS. Hepatotoxicity remains a major cause of drug withdrawal from the market. Natural products have shown great promise in combating against the toxicity of several commonly used drugs, including acetaminophen. Additionally, some of these natural products, such as resveratrol and curcumin, are now widely accepted chemopreventive agents. Although a majority of natural products investigated to date are non-toxic, some studies have shown liver toxicity by certain natural products. Therefore, the proper selection of the natural products is also necessary.

REFERENCES

[1] Kmieć Z. "Cooperation of liver cells in health and disease". AdvAnatEmbryol Cell Biol. 2001., 161:1-151.

[2] Ogu C, Maxa J. Drug interactions due to cytochrome P_{450}. ProcBaylUniv Med Cent, 2000; 13:421-423.

[3] Naruse K, Tang W, Makuuchi M. Artificial and bioartificial liver support: A review of perfusion treatment for hepatic failure patients. World J Gastroentero. 2007., l13: 1516-1521.

[4] Singh R, Kumar S, Rana AC, Sharma N.Different models of hepatotoxicity and related liver diseases: A review, IRJP, 2012., 3 (7):86-95.

[5] Ahsan R, Islam M, Bulbul JI, Musadik A, Haque E.Hepatoprotective activity of methanol extract of some medicinal plants against carbon tetrachloride- induced hepatotoxicity in rats Eur j sci res., 2009., 37(2):302-310.

[6] Reichen J. The Role of the Sinusoidal Endothelium in Liver Function . News Physiol. Sci. 1999., 14:117-121.

[7] Friedman SE, Grendell JH, McQuaid K, Kenneth R.Current diagnosis & treatment in gastroenterology. New York: lang medical Book / mcgraw-hill. 2008., 664-679.

[8] Saukkonen JJ, Cohn DL, Jasmer RM, Schenker S, Jereb JA, Nolan CM, Peloquin CA, Gordin FM, Nunes D, Strader DB, Bernardo J. venkataramanan R, Sterling TR. An Official ATS Statement: Hepatotoxicity of antituberculosis therapy. Am J RespirCrit Care Med.2006., 174: 935-952.

[9] Ostapowicz G, Fontan R J, Schiødt F V, Larson A, Davern TJ, Han S HB, McCashland TM, Shakil AO, Hay JE, Hynan L, Crippin JS, Blei AT, Samuel G, Reisch J, Lee WM.Results of a Prospective Study of Acute Liver Failure at 17 Tertiary Care Centers in the United States, Ann Intern Med.2002., 137:947-954.

[10] Jaeschke H, Gores GJ, Cederbaum AI, Hinson JA, PessayreD,Lemasters J. Mechanisms of hepatotoxicity. J Toxicol Sci.2002., 65(2):166-76.

[11] Subramaniam S, Han HBH, Elumalai N, Lakshmi SYS.Hepatoprotective effect of ethanolic extract of whole plant of Andrographispaniculata against CCL_3-induced hepatotoxicity in rats. Comp Clin Path.2015., 24: 1-7.

[12] Pandit A, Sachdeva T, Bafna P. Drug-induced hepatotoxicity: A review. Journal of Applied Pharmaceutical Science 2012., 2 (5):233-243.

[13] Giordano, C., Rivas, J., &Zervos, X. An update on treatment of drug-induced liver injury. J ClinTransl Hepatol 2014., 2(2): 74.

[14] Singh D, Cho W C, Upadhyay G. Drug-induced liver toxicity and prevention by herbal antioxidants: an overview. Frontiers in physiology 2016; 6: 363.

[15] Subramaniam A, Pushpangadan P. Development of Phytomedicines for liver diseases. Indian J Pharmacology 1999; 31: 166-175.

[16] Ghori SS, Khan M, Rahman SA. Amelioration of carbon tetrachloride-and paracetamol-induced hepatotoxicity in rats by Ficusdalhousiae. Bangladesh J Pharmacol 2014;9(4):588-594.

[17] Rizzardini M, Zappone M, Villa P, Gnocchi P, Sironi M, Diomede L et al.Kupffer cell depletion partially prevents hepatic heme oxygenase1 messenger RNA accumulation in systemic inflammation in mice: role of interleukin 1beta. Hepatology 1998; 27:703–710.

[18] Muzafar AS, Manju T. Asian Liver Toxicity and its Amelioration by Natural Antioxidants - AReview J. Exp. Sci. 2018; 32(1):35-43.

[19] Kedderis GL. Biochemical basis of hepatocellular injury. Toxicol Pathol.1996; 24(1):77-83.

[20]Lynch T, Price A. The effect of cytochrome P450 metabolism on drug response, interactions, and adverse effects. Am Fam Physician 2007; 76(3):391-396.

[21] Dienhart M, Pfeiffer K, Schagger H, Stuart RA.Formation of the yeast F1F0-ATP synthase dimeric complex does not require the ATPase inhibitor protein, Inh1. J. Biol. Chem. 2002; 277: 39289–39295.

[22] RavagnanL,RoumierT, KroemerG. Mitochondria the killer organelles and their weapons. J. Cell. Physiology 2002; 192:131-137.

[23] Murphy MP. Modulating mitochondrial intracellular location as a redox signal. Sci. Signal.2012; 5(242):39.

[24] Richter C.Do mitochondrial DNA fragments promote cancer and aging? FEBS Lett. 1988; 241: 1-5.

[25] Yakes FM, Van Houten B.Mitochondrial DNA damage is more extensive and persists longer than nuclear DNA damage in human cells following oxidative stress. Proc. Natl. Acad. Sci. USA 1997; 94(2): 514–519.

[26] Teschke R, Schulze J. Suspected herbal hepatotoxicity: requirements for appropriate causality assessment by the US Pharmacopeia. Drug Saf. 2012; 35(12):1091-7.

[27] Pessayre D,Fromenty B, Berson A, Robin MA, Letteron P, Moreau M et al.Central role of mitochondria in drug-induced liver injury. Drug Metab Rev. 2012; 44(1):34-87.

[28] Shayiq RM, Roberts DW, Rothstein K, Snawder JE, Benson W, Ma XM.Repeat exposure to incremental doses of acetaminophen provides protection against acetaminophen-induced lethality in mice: an explanation for high acetaminophen dosage in humans without hepatic injury. Hepatology, 1999; 29(2):451-463.

[29] Roberg K, Kagedal K, O¨llinger K. Microinjection of cathepsinDinducescaspase-dependent apoptosis in fibroblasts. Am. J. Pathol. 2002; 161:96-98.

[30] Robin MA, Le Roy M, Descatoire V, Pessayer D.Plasma membrane cytochromes P450 as neoantigens and autoimmune targets in drug-induced hepatitis. J Hepatol 1997; 26: 23–30.

[31] Uetrecht J.New concepts in immunology relevant to idiosyncratic drug reactions: the 'danger hypothesis' and innate immune system. Chem Res Toxicol 1999; 12:387–95.

[32] Park BK, Kitteringham NR, Powell H, Pirmohamed M. Advances in molecular toxicology-towards understanding idiosyncratic drug toxicity. Toxicology 2000; 153: 39–60.

[33] Borgese N, Francolini M, Snapp E. "Endoplasmic reticulum architecture: structures in flux," Current Opinion in Cell Biology 2006;18(4):358–364.

[34] Reddy JK. "Peroxisome proliferators and peroxisome proliferator-activated receptor-α : biotic and xenobiotic sensing," Am J Pathol 2004;164(6):2305–232.

[35] Csala M, Margittai E, Banhegyi G. "Redox control of endoplasmic reticulum function," Antioxidants & Redox Signaling, 2010; 13(1):77–108.

[36] Cuozzo JW, Kaiser CA. "Competition between glutathione and protein thiols for disulphide-bond formation," Nat Cell Biol 1999; 1(3):130–135.

[37] Zhang K, Kaufman RJ. "From endoplasmic-reticulum stress to the inflammatory response," Nature 2008; 454(7203): 455–462.

[38] Malhi H, Kaufman RJ. Endoplasmic reticulum stress in liver disease. J Hepatol 2011; 54: 795–809.

[39] Cnop M, Foufelle F, Velloso LA. Endoplasmic reticulum stress, obesity and diabetes. Trends Mol Med 2012; 18: 59–68.

[40] Cheng B, Gong H, Xiao H, Petersen RB, Zheng L, Huang K. Inhibiting toxic aggregation of amyloidogenic proteins: a therapeutic strategy for protein misfolding diseases. BiochimBiophysActa 2013; 1830: 4860–4871.

[42] Hoyer-Hansen M, Jäättelä M. Connecting endoplasmic reticulum stress to autophagy by unfolded protein response and calcium. Cell Death Differ 2007; 14: 1576–1582.

[43] Digaleh H, Kiaei M, Khodagholi F. Nrf2 and Nrf1signaling and ER stress crosstalk: implication for proteasomal degradation and autophagy. Cell Mol Life Sci 2013; 70: 4681–4694.

[44] Huotari J, Helenius A. Endosome maturation. EMBO J. 2011; 30:3481–3500.

[45] Luzio JP, Pryor PR, Bright NA. Lysosomes: fusion and function. Nat. Rev. Mol. Cell Biol. 2006; 8:622–32.

[46] Ruivo R, Anne C, Sagne C, Gasnier B. Molecular and cellular basis of lysosomaltransmembraneprotein dysfunction. Biochim. Biophys. Acta. 2009; 1793:636–49.

[47] Saftig P, Klumperman J. Lysosome biogenesis and lysosomal membrane proteins: Trafficking meets function. Nat. Rev. Mol. Cell Biol. 2000; 10:623–35.

[48] Kolter T, Sandhoff K.Lysosomal degradation of membrane lipids.FEBS Letters, 2010; 584:1700-1712.

[49] Pryor JPR, Luzio P.Delivery of endocytosed membrane proteins to the lysosome. Biochimica et BiophysicaActa (BBA)-Molecular Cell Research, 2009: 1793 (4):615-624.

[50] Mindell JA. "Lysosomal Acidification Mechanisms". Annual Review of Physiology.2012; 74(1): 69–86.

[51] Gerasimenko JV, Gerasimenko OV, Petersen OH. Membrane repair: Ca(2þ)-elicited lysosomalexocytosis.Current Biology, 2001; 11:971-974.

[52] Kroemer G, Jaattela M. Lysosomes and autophagy in cell death control.Nature Reviews Cancer, 2005; 5:886-897.

[53] Vasiljeva O, Turk B. Dual contrasting roles of cysteine cathepsins in cancer progression: apoptosis versus tumour invasion. Biochimie 2008; 90: 380-386.

[54] Aits S, Jaattela M.Lysosomal Cell Death at a glance.J Cell Sci, 2013; 126:1905-1912.

[55] Krenn MA, Schürz M, Teufl B, Uchida K, Eckl PM, Bresgen N. "Ferritin-stimulated lipid peroxidation, lysosomal leak, and macroautophagy promote lysosomal 'metastability' in primary hepatocytes determining in vitro cell survival,"Free Radical Biology and Medicine 2015; 80:48-58.

[56] Lill R, Hoffmann B, Molik S, Antonio JP, Nicole RC, Oliver S et al. "The role of mitochondria in cellular iron-sulfur protein biogenesis and iron metabolism," Biochimica et BiophysicaActa—Molecular Cell Research 2015; 1823(9):1491–1508.

[57] Ashif I, Mohammad KI, Syed EH. Experimental hepatotoxicity Inducing agents: A Review, Int J Pharm Sci Res 2016; 6(11):325-335.

[58] Meek IL, Van MAFJ, Vonkeman HE. Non-steroidal anti-inflammatory drugs: an overview of cardiovascular risks. Pharmaceuticals. 2010; 3:2146–2162.

[59] Parmar SR, Vashrambhai PH, Kalia K. Hepatoprotective activity of some plants extract against paracetamol induced hepatotoxicity in rats. J Herbal Med Toxicol 2010; 4:101–6.

[60] Sakr SA, Abo-El-Yazid SM. Effect of fenugreek seed extract on adriamycin-induced hepatotoxicity and oxidative stress in albino rats. ToxicolInd Health 2012; 28(10):876-885.

[61] Talpur M, Shaikh S, Shah SA, Memon AR, Khohazo HK. Zinc sulphate in azathioprine induced hepatotoxicity -An experimental study. WJBMS 2014; 1(4):72-80.

[62] Singh N, Rani P, Gupta M, Goel N, Tandan N. Effects of aqueous extract of camellia sinensis on liver markers of cadmium treated rats. E3 J Biotechnol Pharm Res 2013; 4(5):89-93.

[63] Suganthi V, Gowri S, Gurusamy K. Hepatoprotective activity of Cayratiacarnosaon liver damage caused by lead acetate in rats. Sch Res Lib 2013; 3(2):76-79.

[64] Ekambaram M, Ramalingam K, Balasubramanian A. Effect of Portulacaquadrifida Linn on Mercury-Induced Hepatotoxicity in Swiss Albino Mice.Res Rev J PharmacolToxicol Stud. 2014;2:18–21.

[65] Clark SP, Davis MA, Ryan TP, Searfoss GH, Hooser SB. Hepatic gene expression changes in mice associated with prolonged sublethalmicrocystin exposure. ToxicolPathol 2007;35(4):594-598.

[66] Lanternier F, Roulot D, Bentata M, Pol S, Viard JP, Gordien E, Jeantils Vet al.GastroenterolClin Biol. 2007;31(10):822-827.

[67] Sanyal AJ. AGA technical review on nonalcoholic fatty liver disease. Gastroenterology 2002;123:1705–1725.

[68] Mauro P, Renze B, Wouter W. Enzymes. In: Tietz text book of clinical chemistry and molecular diagnostics. Carl AB, Edward R, David EB. 4th edition, Elsevier 2006; 604-616.

[69] Karmen A, Wroblewski F, Ladue JS."Transaminase activity in human blood". J Clin Invest 1955; 34 (1): 126–131.

[70] Lindblom P, Rafter I, Copley C,Andersson U, Hedberg JJ, Berg AL, et al. Isoforms of alanine aminotransferases in human tissues and serum – differential tissue expression using novel antibodies. Arch BiochemBiophys 2007; 466(1):66–77.

[71] Ghouri N, Preiss D, Sattar N. "Liver enzymes, nonalcoholic fatty liver disease, and incident cardiovascular disease: a narrative review and clinical perspective of prospective data". Hepatology. 2010;52 (3): 1156–1161.

[72] Kallei L, Hahn A, Roder VZ. Correlation between histological findings and serum transaminase values in chronic diseases of the liver. ActaMedicaScandinavika 1964;175:49-56.

[73] Voet D, Voet JG. Biochemistry, Biomolecules, mechanisms of enzyme action and metabolism. Electron transport and oxidative phosphorylation; 3rd edition; John Wiley and Sons, Inc. United States of America, 2004; 742-797.

[74] Pincus MR, Tierno PM, Fenelus M, Bowne WB, Bluth MH. Evaluation of liver function. In: McPherson RA, Pincus MR, eds. Henry's Clinical Diagnosis and Management by Laboratory Methods. 22nd ed. Philadelphia, PA: Elsevier Saunders: Chap 21, 2011.

[75] Thapa BR, Anuj W. Liver Function Tests and their Interpretation. Indian J Pediatr 2007;74: 663-671.

[76] Panteghini M, Falsetti F, Chiari E,Malchiodi A.Determinaton of Aspartate aminotransferase isoenzymes in hepatic disease. Lab J Res Lab Med 1983;10: 515-519.

[77] Rankin SA, Chrisitiansen A, Lee W, Banavara DS, Lopez-Hernandez A. Invited review: The application of alkaline phosphatase assays for the validation of milk product pasteurization. J Dairy Sci 2010;93(12):5538-5551.

[78] Arika WM, Nyamai DW, Osano KO, Ngugi MP, and Njagi ENM. Biochemical Markers of *In Vivo*Hepatotoxicity.JClinToxicol 2016; 6(2):297-305.

[79] Blanchard KT, Clay RJ, Morris JB. Pulmonary activation and toxicity of cyclopentadienyl manganese tricarbonyl. ToxicolApplPharmacol. 1996;136(2):280-288.

[80] Beaussier M., Wendum D., Schiffer E., Dumont S., Rey C., Lienhart A.et al. Prominent contribution of portal mesenchymal cells to liver fibrosis in ischemic and obstructive cholestatic injuries. Lab Invest 2007; 87(3):292-303.

[81] Wolf PL. Biochemical diagnosis of liver disease. Indian J ClinBiochem1999; 14(1):59-90.

[82] Nurbanu G, Eren G, Zeliha AB. Gamma-GlutamylTransferase Levels in Patients with Acute Ischemic Stroke, Cardiovascular Psychiatry and Neurology 2014; 1-4

[83] Sugiura M, Nakamura M , Ikoma Y, Yano M, Ogawa K, Matsumoto H et al. High serum carotenoids are inversely associated with serum gamma-glutamyltransferase in alcohol drinkers within normal liver function. Am J Epidemiol 2005;15:180-186.

[84] Wu A, Slavin G, Levi AJ. Elevated serum gamma-glutamyl-transferase (transpeptidase) and histological liver damage in alcoholism. Am J Gastroenterol1976; 65: 318-323.

[85] Hamed Khalili, Barham Abu Dayyeh, Lawrence SF. Assessment of Liver Function in Clinical Practice. Chronic Liver Failure 2010; 47-76

[86] Thomsen HF, Hardt F, Juhl E. Diagnosis of Gilbert's syndrome. Scand J Gastroenterol 1981;16:699-703.

[87] Beckingham IJ, Ryder SD. Clinical review ABC of diseases of liver, pancreas, and biliary system Investigation of liver and biliary disease. BMJ 2001; 322: 33-36.

[88] Daniel SP, Marshall MK. Evaluation of the liver: laboratory tests. Schiff's diseases of the liver,8th edn. USA; JB Lippincott publications 1999; 205-239.

[89] American Gastroenterological association. American gastroenterological association medical position statement: Evaluation of liver chemistry tests. Gastroenterology 2002; 123:1364-1366.

[90] Sherlock S. Assessment of liver function Disease of liver and biliary system: Sheila Sherlock, 10thed, London; Blackwell science ltd, 1997; 17-32.

[91] Rosalki SB, Mcintyre N. Biochemical investigations in the management of liver disease. Oxford textbook of clinical hepatology, 2nd ed. New York; Oxford university press, 1999; 503-521.

[92] Killingsworth, L.M.. Plasma protein patterns in health and disease. Crit. Rev. Chn. Lab. Sci 1979;11,1-30.

[93] Bradbeer, JN, Zanelli, JM, Lindsay, PC, Pearson, J Reeve, J. Relationship between the location of osteoblastic alkaline phosphatase activity and bone formation in human iliac crest bone. J. Bone Min. I Res 1992;7, 905-912.

[94] Rosalki SB, Foo AY, Burlina A, Prellwitz W, Stieber P, Neumeier D et al.Multicentric evaluation of Iso-ALP test kit for measurement of bone alkaline phosphatase in serum and plasma. Clin.Chem 1993; 39,648-652.

[95] Aliyazicioglu Y, Deger O, Karahan C, Yildirmis S, Kucukoduk S. Reference values of cord blood transferrin, ceruloplasmin, alpha-1 antitrypsin, prealbumin, and alpha-2 macroglobulin concentrations in healthy term newborns. Turk J Pediatr 2007; 49: 52–54.

[96] Saito M, Seo Y, Yano Y, Miki A, Yoshida M, Azuma T. Short-term reductions in non-protein respiratory quotient and prealbumin can be associated with the long-term deterioration of liver function after transcatheter arterial chemoembolization in patients with hepatocellular carcinoma. J Gastroenterol 2012;47: 704–714.

[97] Liu F, Cai LY, Zhong L, Chen C, Xu F, Zhao ZX. Model for end-stage liver disease combined with serum prealbumin to predict the prognosis of patients with decompensated liver cirrhosis. J Dig Dis 2010;11: 352–357.

[98] Li S, Tan HY, Wang N, Zhang ZJ, Lao L, Wong CW. The role of oxidative stress and antioxidants in liver diseases. Int J MolSci 2015; 16(11): 26087-26124.

[99] Kunwar A, Priyadarsini KI. Free radicals, oxidative stress and importance of antioxidants in human health. J Med Allied Sci 2011; 1(2): 53-60.

[100] Betteridge DJ. What is oxidative stress?. Metabolism 2000;49(2): 3-8.

[101] Said MA, Aiman IA. Oxidative stress versus antioxidants. American Journal of Bioscience and Bioengineering 2014; 2(5): 60-71.

[102] Banerjee P, Jana S,Chakraborty S,Swarnakar S. Inflammation and MMPs in alcohol-induced liverdiseases and protective action of antioxidants. Indian J.Biochem. Biol 2013; 50:377–386.

[103] Gao B,Bataller R. Alcoholic liver disease: Pathogenesis and new therapeutic targets. Gastroenterology 2011;141:1572–1585.

[104] Masalkar PD, Abhang SA. Oxidative stress and antioxidant status in patients with alcoholic liver disease.Clin. Chim. Acta2005;355: 61–65.

[105] Beier JI, McClain CJ. Mechanisms and cell signaling in alcoholic liver disease. Biol. Chem 2010; 391:1249–1264.

[106] Diehl AM.Recent events in alcoholic liver disease V. Effects of ethanol on liver regeneration. Am. J.Physiol.-Gastrointest. Liver Physiol 2005;288:1–6.

[107] Videla LA. Oxidative stress signaling underlying liver disease and hepatoprotective mechanisms. World J.Hepatol 2009; 1: 72–78.

[108] Linares V, Alonso V,Albina ML, Belles M,Sirvent JJ, Domingo JL et al. Lipid peroxidationand antioxidant status in kidney and liver of rats treated with sulfasalazine. Toxicology 2009; 256: 152–156.

[109] Karabulut AB,Gui M,Karabulut E,Kiran TR,Ocak SG,Otlu O. Oxidant and antioxidant activity in rabbit livers treated with zoledronic acid. Transplant. Proc 2010; 42:3820–3822.

[110] Mladenovic D,Radosavljevic T,Ninkovic M,Vucevic D,Jesic-Vukicevic R,Todorovic V. Liverantioxidant capacity in the early phase of acuteparacetamol-induced liver injury in mice. Food Chem.Toxicol 2009;47:866–870.

[111] Bando I, Reus MI, Andres D,Cascales M. Endogenous antioxidant defence system in rat liver followingmercury chloride oral intoxication. J. Biochem. Mol. Toxicol 2005;19:154–161.

[112] Adegbesan BO,Adenuga GA. Effect of lead exposure on liver lipid peroxidative and antioxidant defensesystems of protein-undernourished rats. Biol. Trace Element Res 2007;116:219–225.

[113] Ozgur E, Guler G,Seyhan N. Mobile phone radiation-induced free radical damage in the liver is inhibitedby the antioxidants N-acetyl cysteine and epigallocatechin-gallate. Int. J. Radiat. Biol 2010;86: 935–945.

[114] Yildirim NC,Yurekli M,Yildirim N.Investigation of some antioxidant enzymes activities depending onadrenomedullintreatment and cold stress in rat liver tissue. Turk. J. Biochem 2010;35: 138–142.

[115] Syama S,Reshma SC,Sreekanth PJ,Varma HK,Mohanan PV. Effect of zinc oxide nanoparticles on cellular oxidative stress and antioxidant defense mechanisms in mouse liver. Environ. Toxicol. Pharmacol 2013;95:495–503.

[116] Zhang X,Strakovsky R, Zhou D, Zhang Y, Pan YX. A maternal high-fat diet represses the expressionof antioxidant defense genes and induces the cellular senescence pathway in the liver of male offspringrats. J. Nutr 2011;141:1254–1259.

[117] Dornas WC, Lima WG, Santos RC, Guerra JF, Souza MO, Silva M. et al.High dietary salt decreases antioxidant defenses in the

liver of fructose-fedinsulin-resistant rats. J. Nutr. Biochem 2013;. 24: 2016–2022.

[118] Kohen R, Nyska A.Oxidation of biological systems: oxida-tive stress phenomena, antioxidants, redox reactions, and methods for their quantification. ToxicolPathol 2002;30:620-630.

[119] Mates JM.. Effects of antioxidant enzymes in the molecular control of reactive oxygen species toxicology. Toxicology2000;153:83-104.

[120] Bors W, Heller W, Michel C, Stettmaier K. "Flavonoids and Polyphenols: chemistry and biology" In Handbook of Antioxidants. New York, 1996;409.

[121] Bendich A, Olson JA.. Biological actions of carotenoids. FASEB J 1989;3:1927-1932.

[122] Goldman A. Melatonin, a review. Brit J ClinPharma 1995;19:258-260.

[123] Seifried HE, Anderson DE, Milner JA, Greenwald P. New developments in antioxidant research, Nova Science Publishers Inc., Hauppauge (NY)2006..

[124] Herbert V.The antioxidant supplement myth. Am J ClinNutr 1994; 60:157–168.

[125] Galati G, Sabzevari O, Wilson JX, Brien P J.Prooxidant activity and cellular effects of the phenoxyl radicals of dietary flavonoids and other polyphenolics. Toxicology 2002; 177:91–104.