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Drugs and Steatohepatitis

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ABSTRACT

In addition to the usual associations with insulin resistance, type 2 diabetes, central obesity, and hypertriglyceridemia, nonalcoholic steatohepatitis (NASH) has been associated with several drugs and toxins. However, drug-induced liver disease is a relatively uncommon cause of steatohepatitis. The term drug-induced steatohepatitis is preferred when the association appears to result from a direct toxic effect of the drug on the liver. For some agents implicated as causing cirrhosis or fatty liver disorders, the association may be coincidental because NASH is a common component of the insulin resistance (or metabolic) syndrome. In other instances, corticosteroids, tamoxifen, and estrogens may precipitate NASH in predisposed persons by exacerbating insulin resistance, central obesity, diabetes, and hypertriglyceridemia, and methotrexate may worsen hepatic fibrosis in NASH. Drug-induced steatohepatitis is associated with prolonged therapy (more than 6 months) and possibly drug accumulation, which in the case of perhexiline maleate is favored by a genetic polymorphism of *CYP2D6* that leads to slow perhexiline oxidation. The toxic mechanism appears to involve mitochondrial injury, which causes steatosis because of impaired β-oxidation of fatty acids, and leads to generation of reactive oxygen species and ATP depletion. Thus, drug-induced steatohepatitis may provide clues to injurious events in the more common metabolic forms of NASH. A clinical feature of some types of drug-induced steatohepatitis is progression after discontinuation of the causative agent. It follows that early recognition of hepatotoxicity is crucial to prevent the development of severer forms of liver disease and improve the clinical outcome.

KEYWORDS: Nonalcoholic steatohepatitis, insulin resistance syndrome, drugs, perhexiline maleate, amiodarone, tamoxifen, estrogens, corticosteroids, methotrexate, calcium channel blockers, industrial hepatotoxicity, hepatic fibrosis

Objectives: Upon completion of this article, the reader should (1) be familiar with the drugs associated with steatohepatitis; (2) understand the pathophysiologic basis for those associations, including possible etiopathogenic mechanisms; and (3) appreciate the standard of care required for early recognition of drug-induced steatohepatitis to avert poor clinical outcomes.

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DEFINITION OF NASH AND IMPORTANCE OF DRUG-INDUCED STEATOHEPATITIS

Ludwig introduced the term nonalcoholic steatohepatitis (NASH) to describe a form of liver disease that is histologically indistinguishable from alcoholic hepatitis but occurs in people who do not consume excess ethanol.[1] It is now clear that the spectrum of nonalcoholic fatty liver disorders (NAFL) extends from bland steatosis, which is nonprogressive, through steatohepatitis (NASH) to forms of "cryptogenic" cirrhosis in which steatosis may be inconspicuous.[2-7] To reflect this, NASH is now conceptualized as encompassing at least three components among the tetrad of steatosis, hepatocellular injury, focal mixed cell–type inflammation, and fibrosis.[8] However, there is not yet complete agreement on the semantics (NAFL versus NASH) or definition of what constitutes steatohepatitis in the broad spectrum of fatty liver disorders. The recommended criteria of Brunt will be adopted here,[9] as recently reviewed in *Seminars*.[8] This pathological definition of NASH requires steatosis, significant hepatocellular injury (most often indicated by ballooning degeneration), diffuse mixed lobular inflammation, and perisinusoidal and perivenular fibrosis.

Drug-induced liver diseases (DILDs) are clinicopathologic patterns of liver injury caused by drugs or other foreign compounds.[10-12] Steatohepatitis is a rare form of DILD,[11,13] and drugs account for fewer than 2% of cases of NASH. In order to implicate medications or toxins as causes of steatohepatitis, there should be a close relationship with drug ingestion, including absence of liver test abnormalities before drug ingestion and their resolution after discontinuing the agent. Ideally, recurrence of test abnormalities after reintroduction of the drug (rechallenge) will have been demonstrated, and there should be a biological basis for hepatotoxicity. It is noteworthy that drugs that cause steatohepatitis are often associated with other types of liver injury, particularly acute hepatitis with a shorter latent period to onset. On the other hand, many of the reported associations between drugs and NASH are problematic because of the tenuous temporal relationships, lack of rechallenge data, and high prevalence of NASH in the population for which the drug was prescribed. In particular, some agents implicated as causing cirrhosis or other forms of NAFL in obese middle-aged diabetic women (methyldopa, calcium channel blockers, estrogens, methotrexate) may be fortuitous coassociations with "primary" NASH, which most often occurs among persons with the insulin resistance or metabolic syndrome.[7,8,14-19]

Another possibility, as with tamoxifen and methotrexate, is that drugs can precipitate or exacerbate underlying NASH by accentuating the predisposing metabolic factors, including insulin resistance, lipid disorders, or activation of stellate cells and other profibrogenic mechanisms. This proposal is still conjectural, but it is worthy of investigation for both its practical clinical significance and what it may reveal about the pathogenesis of the much more common "metabolic" forms of NASH.

PATHOLOGY OF DRUG-INDUCED STEATOHEPATITIS

The association between drugs and a form of chronic liver disease that resembled alcoholic steatohepatitis was first recognized in Japan in 1973 for the vasodilator and antianginal agent Coralgil (4,4'-diethylaminoethoxyhexestrol).[12,19,20] Reports from Europe then observed that perhexiline maleate was implicated in causing a spectrum of liver disease that included alcoholic hepatitis–like lesions with Mallory bodies and cirrhosis.[21-23] This experience extended the conceptual base of steatohepatitis beyond alcoholic liver disease (note that Ludwig's seminal paper on metabolic NASH appeared in 1979).

In general, drug-induced steatohepatitis resembles alcoholic liver disease more closely than NASH associated with diabetes and the insulin resistance syndrome. Thus, cirrhosis and hepatic failure are common, and the liver pathology shows impressive polymorphonuclear leukocyte (polymorph) infiltration, ballooning degeneration of hepatocytes, and Mallory bodies (Fig. 1). Progression of fibrosis to cirrhosis can occur over weeks or months in drug-induced steatohepatitis, whereas it may take decades and most often never occurs in patients with NASH.

Myeloid bodies are a characteristic ultrastructural feature of drug-induced steatohepatitis (Fig. 2). These are derived from enlarged lysosomes stuffed with whorled membranous material that resembles the myelin sheath of nerves. Myeloid bodies have been ascribed to phospholipidosis, which animal studies show is a dose-dependent phenomenon attributable to the physicochemical properties of cationic amphiphilic drugs.[24] These properties favor accumulation of the drug in lipid membranes and proton-rich organelles, including lysosomes and mitochondria. Drugs associated with phospholipidosis may inhibit lysosomal phospholipases directly or by binding to phospholipids, thereby inhibiting the turnover of lipid bilayers.[25] The relationship of the storage disorder phospholipidosis to the development of steatohepatitis is not clear.[24] Phospholipidosis does not seem to lead to NASH directly, and many drugs associated with this ultrastructural phenomenon never seem to cause steatohepatitis (Table 1).

In other respects, the hepatic lesions of drug-induced steatohepatitis are similar or identical to those of alcoholic steatohepatitis or NASH. With perhexiline, Coralgil, and amiodarone, focal necrosis, Mallory bodies, mixed cellular inflammation with numerous

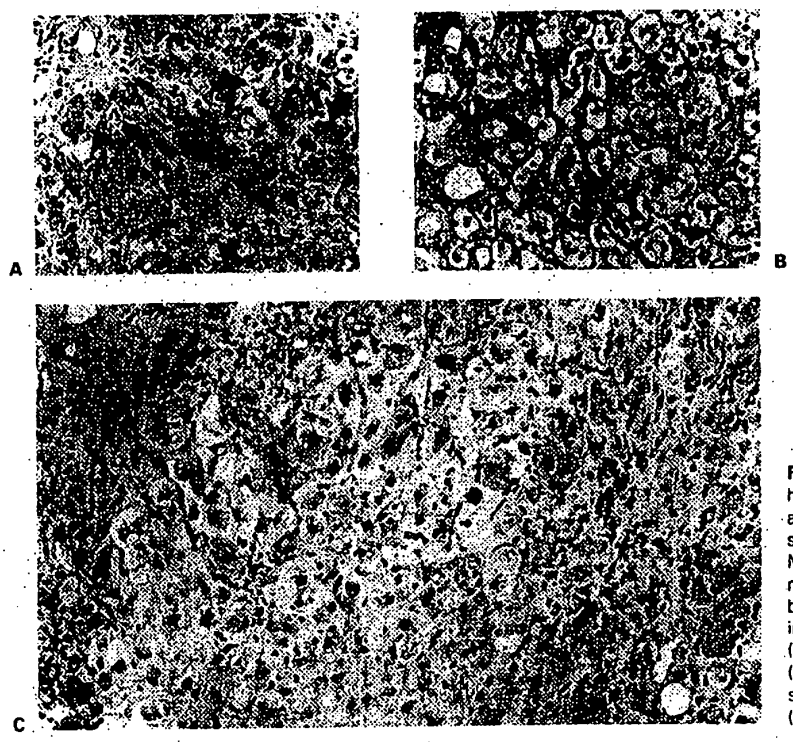

Figure 1 Perhexiline maleate–induced hepatitis. (A) The biopsy shows moderately severe hepatitis with focal necrosis, prominent polymorph infiltrate, and Mallory bodies (arrows) (H & E, original magnification × 600). (B) Pericellular fibrosis. Mallory bodies are clearly evident in several enlarged, hydropic liver cells (Sirius red, original magnification × 600). (C) Low-power view of the same biopsy specimen indicates evolving cirrhosis (H & E, original magnification × 360).

polymorphs, perisinusoidal fibrosis, and cirrhosis may be striking. The pathology of steatohepatitis reported in association with nifedipine, diltiazem, tamoxifen, estrogens, and corticosteroids appears to resemble more closely cases of NASH occurring with the insulin resistance syndrome, but definitive histologic studies are lacking.[20,26–34]

PATHOGENESIS

The several reasons for associations between drugs and steatohepatitis are summarized in Table 2. The first possibility is that the association is spurious; that is, a person with NASH happens to be taking the drug for conditions associated with the insulin resistance syndrome, such as hypertension or other cardiovascular disorders. The second is that the drug may precipitate or accentuate risk factors for NASH, such as central obesity, diabetes, and hypertriglyceridemia. A third alternative is that occult alcoholism may involved; this has been suspected in some reported associations between industrial solvents and hepatotoxicity.[11,35]

The final possibility is that some drugs actually *cause* steatohepatitis by a direct hepatotoxic mechanism. This seems to be the case for perhexiline maleate, Coralgil, amiodarone, and possibly tamoxifen. Perhexiline maleate–induced liver disease occurred principally in persons who had the debrisoquine slow metabolizer phenotype,[23] which results from a genetic polymorphism or mutation of the cytochrome P450 (CYP) gene, *CYP2D6*. CYP2D6 is centrally involved in perhexiline metabolism. Together with the evidence of partial dose dependence,[11] this implicates accumulation of the drug in the pathogenesis of steatohepatitis. French investigators have shown that perhexiline, amiodarone, and tamoxifen can accumulate in mitochondria and inhibit mitochondrial β-oxidation, thereby causing steatosis.[36–38] They also inhibit oxidative phosphorylation,[36–39] and in the longer term this could be relevant to the pathogenesis of liver injury by favoring electron leakage with production of reactive oxygen species (ROS) causing lipid peroxidation as well as decreasing cellular ATP levels. Thus, drug-induced steatohepatitis may be a paradigm for injurious events in the more common metabolic forms of NASH, in which ROS production, oxidative stress, and mitochondrial injury appear to operate as part of the pathogenesis of liver injury in a preexisting fatty liver.[16,18,19,40–42]

Other studies implicate peroxisome proliferator-activated receptors (PPARs) as leading to NASH.[43] PPARα is a transcription factor that governs both microsomal (via CYP4A) and peroxisomal (β-oxidation) pathways of lipid oxidation and ultimately production of ROS. Increased lipid peroxidation is a crucial difference between the livers of rodents with experimental steatohepatitis and those of *ob/ob* genetically obese (leptin-deficient) mice that have uncomplicated steatosis.[42] PPARγ also plays a regulatory role in lipid homeostasis

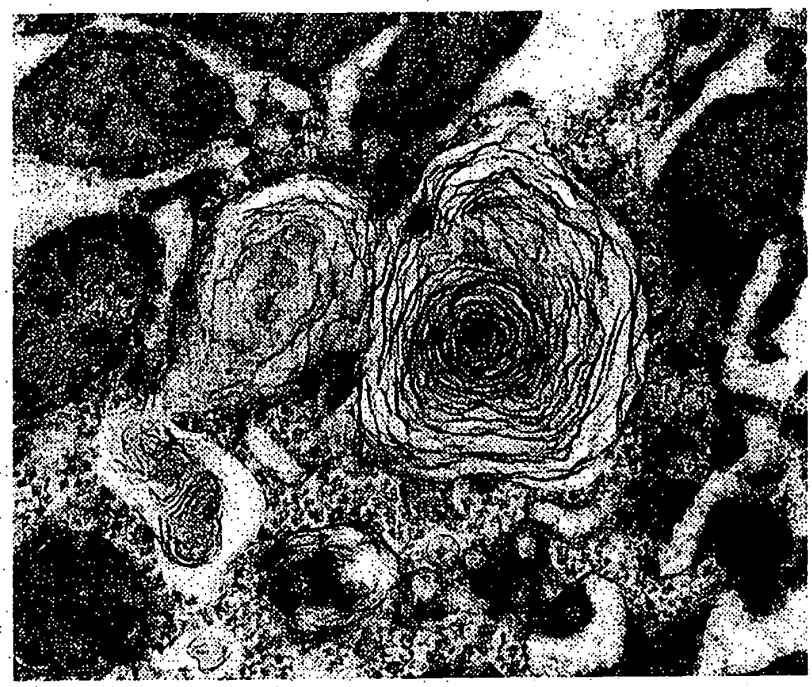

Figure 2 Phospholipidosis in a person taking amiodarone. Electron micrograph of portion of hepatocyte showing "myeloid bodies" as roughly concentric arrays of closely packed membranous whorls (original magnification × 4900).

and is up-regulated in livers of obese and diabetic mice.[44] Thiazolidinediones (troglitazone, rosiglitazone), which reverse insulin resistance and correct hyperglycemia and hyperlipemia in type 2 diabetes, also bind to and activate this receptor. In mice with obesity-associated up-regulation of hepatic PPARγ, thiazolidinediones produced severe periacinar microvesicular steatosis, indicative of mitochondrial injury.[44] It therefore seems possible that type 2 diabetes may sensitize the liver to PPARγ-activating drugs, but further studies are required to establish whether the important hepatotoxicity associated with this group of drugs includes steatohepatitis or overlaps mechanistically with the production of steatohepatitis.

CLINICOPATHOLOGICAL FEATURES AND COURSE WITH SPECIFIC AGENTS

The drugs reported to be associated with steatohepatitis are listed in Table 1.

Perhexiline Maleate

Perhexiline maleate was used extensively in Europe to treat angina pectoris but has now been withdrawn. Hepatic side effects were found in up to one third of cases.[11,12,22] They included phospholipidosis, hepatocellular hyperplasia, granulomatous hepatitis, steatohepatitis, and micronodular cirrhosis. In cases of steatohepatitis, Mallory bodies and pericellular fibrosis were prominent. Perhexiline-associated steatohepatitis was often severe, and complications such as portal hypertension led to death in more than 50% of cases.[11,12,22]

Perhexiline hepatotoxicity was partly related to the total dose of perhexiline. However, people who metabolize perhexiline slowly because of genetic polymorphism in *CYP2D6* may have been particularly susceptible.[23] Further, in cases of steatohepatitis caused by perhexiline maleate, liver injury sometimes progressed after drug withdrawal.[22] This may reflect the long tissue residence times rather than autopropagative mechanisms of steatohepatitis, but it could also be a manifestation of mitochondrial injury; slow recovery or failure to recover has been better documented for nucleoside reverse transcriptase inhibitors (NRTIs), such as fialuridine and drugs used in highly active antiretroviral therapy (HAART).[45–48] Experimental studies of the role of mitochondrial injury in liver disease caused by perhexiline and amiodarone (see the following) have been described by Fromenty, Pessayre, and colleagues.[36–38]

Amiodarone

Amiodarone is an iodinated benzofuran derivative used for therapy-resistant ventricular tachyarrhythmias. However, in 25% of patients therapy must be discontinued because of adverse effects. These include pulmonary in-

Table 1 Drugs Reported to Be Associated with Phospholipidosis

Phospholipidosis and steatohepatitis
Amiodarone
Perhexiline maleate
4,4′-Diethylaminoethoxyhexestrol (Coralgil)
Chloroquine
Nifedipine
Tamoxifen
Diltiazem
Verapamil
Phospholipidosis alone*
Chlorphentermine
Fenfluramine
Triparanol
Azacosterol
Amitriptyline
Imipramine
Chlorpromazine
Thioridazine
Chlorcyclizine
Tripelennamine
Amantadine
Propranolol
Oral contraceptive steroids
Co-trimaxazole

*Observations are in experimental animals, except for chlorpromazine and co-trimoxazole.
Data from references 11, 24, 25, and 55.

filtrates, worsening cardiac failure, hypothyroidism, peripheral neuropathy, nephrotoxicity, and corneal deposits, but liver disease is one of the most serious.[49–52] Abnormal liver tests are noted in 15% to 80% of patients treated with amiodarone, and clinically significant liver disease has been noted in 0.6% to 3%.[11,53] The spectrum of hepatic disorders includes rare cases of acute liver failure due to severe hepatitis or a Reye's syndrome–like illness, cholestatic hepatitis, and granulomas.[49,50,53–59] Several cases of amiodarone-induced liver disease have been proved by rechallenge.[49,50] Amiodarone is the drug most commonly implicated as causing steatohepatitis[11,53–55]; cirrhosis is present in 15% to 50% of such cases.[55,57] A feature that differs from most other types of DILD is that progression of amiodarone steatohepatitis may occur despite discontinuation of the drug.[53,59]

Amiodarone is highly concentrated in the liver. Thus, after a few weeks of treatment, the drug accounts for as much as 1% of the wet weight of the liver, and because the iodine content absorbs radiation, the liver appears opaque on computed tomography (CT).[60] This unusual radiologic appearance is of no clinical significance. However, opacification of the liver is always found in those with amiodarone-induced liver disease, indicating that hepatic storage of the drug is a prerequisite for steatohepatitis. Hepatic storage of amiodarone also produces phopholipidosis.[24] In animals fed amiodarone, development of phospholipidosis is time and dose dependent[24] and may result from the direct inhibition of phospholipase or from the formation of nondegradable drug-phospholipid complexes. It appears to have no relationship to hepatocyte injury and steatohepatitis. However, amiodarone is concentrated in mitochondria by virtue of its physicochemical properties and may interrupt mitochondrial electron transport.[36–38] Thus, in rats and mice, treatment with amiodarone produced microvesicular steatosis, augmented mitochondrial production of ROS, and caused lipid peroxidation.[36–38] In laboratory studies of hepatocytes and mitochondrial subcellular fractions, amiodarone (as well as perhexiline and 4,4′-diethylaminoethoxyhexestrol) inhibited carnitine palmitoyltransferase 1 and acyl-coenzyme A dehydrogenase, decreased β-oxidation, and inhibited mitochondrial respiration.[36–38]

The risk of amiodarone-induced steatohepatitis is related to the duration of exposure; the mean is 21 months, and nearly all affected persons have taken amiodarone for more than 1 year. There is no clear relationship to the incremental dose, but there is a possible relationship to the total dose,[58,61] and the frequency of other toxic effects of amiodarone (most are thought to be dose dependent) is increased in patients with liver disease.[61] Patients' complaints include fatigue, nausea and vomiting, malaise, weight loss, and abdominal swelling as a result of ascites. Features of drug hypersensitivity, such as fever, rash, and eosinophilia, are not conspicuous.[54] Hepatomegaly, jaundice, bruising, and other features of chronic liver disease may be present. The mortality is high, partly because of continued progression of liver disease after discontinuation of amiodarone.[51,56,61]

Liver test abnormalities include raised aminotransferases, most often to at least five times the upper limit of normal, and minor increases in alkaline phosphatase. The ratio of aspartate transaminase (AST) to alanine transaminase (ALT) is close to unity, a change different from that in alcoholic hepatitis. In severe cases, hyperbilirubinemia, low serum albumin levels, and prolongation of the prothrombin time indicate liver failure. The histologic changes include phospholipidosis, steatosis, focal necrosis with Mallory's hyaline, infiltration with polymorphs, pericellular fibrosis, and often cirrhosis.[55,62] In some cases, there is markedly granular cytoplasm,[62] but ultrastructural studies of mitochondrial damage that could explain this finding are lacking.

Although serial liver tests are recommended in those taking amiodarone,[61] it is not known whether this will prevent significant hepatotoxicity. The possibility that DILD may be the cause of abnormal liver tests in a person taking amiodarone poses some dilemmas. First, there are often other potential causes—cardiac failure,

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