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Editorial

Risk factors and pathogenesis of cholesterol gallstones: state of the Art

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Abstract. – **The aim of this article is to present an update of selected aspects of the pathogenesis and risk factors of cholesterol gallstones, a highly prevalent Western disease. The etiology of cholesterol cholelithiasis is considered to be multifactorial, with interaction of genetic and environmental factors. Mechanisms of cholesterol lithogenesis include biliary cholesterol hypersecretion, supersaturation and crystallization, stone formation and growth, and bile stasis within the gallbladder. Each of these various steps could be under genetic control and/or be influenced through intermediate pathogenic steps linked to a variety of environmental factors.**

Key Words:

Gallstones, Etiology, Risk factors, Pathophysiolog

Introduction

The etiology of cholesterol cholelithiasis is considered to be a multifactorial disease, with interaction of genetic and environmental factors[1,2]. Ultrasonographic epidemiological studies have shown very different prevalence of gallstone disease in different populations. The highest prevalence has been found among some North American Indians[3] and Chileans[4], followed by Peruvians[5], Mexican Americans[6] and Europeans[7,8]. The lowest prevalence is observed in Asiatic populations[9]. Gallbladder cancer is endemic in Bolivia[11] and Chile[11] suggesting that genetic lithogenic and, or environmental unknown factors have a high penetrance in South America.

Research in this area has been principally focused on four aspects in the last 20 years. First, the disclosing of the prevalence and risk factors of cholelithiasis in different areas of the world with the pioneering work performed in Italy[6,7]. Secondly, a series of studies have addressed the intrahepatic defect(s) in cholesterol metabolism leading to biliary cholesterol hypersecretion, the key initial pathophysiological event leading to gallstone formation. Third, numerous in vitro and in vivo studies have been oriented to identify biliary proteins as promoter or inhibitor of cholesterol crystal formation. Fourth, altered gallbladder motility is another potentially important pathogenic factor[1]

Risk factors

The multifactorial nature of all chronic diseases, including cholesterol gallstones results from the interaction of genetic and environmental factors. It is thought that these latter are most likely the consequence of westernization of modern societies, including a high intake of refined carbohydrates, and a high prevalence of obesity, non-insulin dependent diabetes, and sedentary life-style[2]. The genetic hypothesis has been supported by family[12-14] as well epidemiological surveys suggesting that Amerindians may have lithogenic genes of high penetrance[4,5]. Furthermore, since biliary cholesterol is mostly derived from performed cholesterol of plasma lipoproteins[1], studies has been focused in genes related

with cholesterol transport like. Human studies have looked for these candidate genes that might favor cholesterol gallstone formation, including expression of apo E[15,16], apo B, apo A-I, and cholesterol ester transfer protein[17]. Similarly, experimental models in mice[18] and strain differences in hamsters[19] have also supported the existence of specific gene loci that favor the formation of cholesterol gallstones.

Figure 1 presents a summary of the accepted risk factors for cholesterol gallstone formation. The principal independent factors associated to gallbladder disease among different population studies are sex, age, low HDL cholesterol, high BMI, percent body fat, a higher than normal serum glucose in women (with, or without NIDDM), parity and hyperinsulinemia[2,4]. A strikingly consistent and frequent finding is the reciprocal relationship between HDL serum cholesterol concentration and gallstone disease, suggesting the hypothesis of a underlying abnormality of HDL cholesterol metabolism in cholesterol gallstone formation[20]. Non insulin – dependent diabetes mellitus (NIDDM), another genetic and environmentally determined chronic disease highly prevalent in Pima Indians[21] and Mexican Americans[22], but not in Chileans[4], has also been considered to favor gallstone formation[2]. Another group of important risk factors refers to the effects of female hormones that might have an important role in cholesterol metabolism and gallbladder motility[23].

Pathogenesis

The key pathogenic steps that are most likely involved in gallstone formation are schematically presented in Figure 2. Mechanisms of cholesterol lithogenesis include biliary cholesterol hypersecretion and supersaturation, cholesterol microcrystal formation, stone growth and gallbladder stasis. Each of these various steps could be under genetic controll

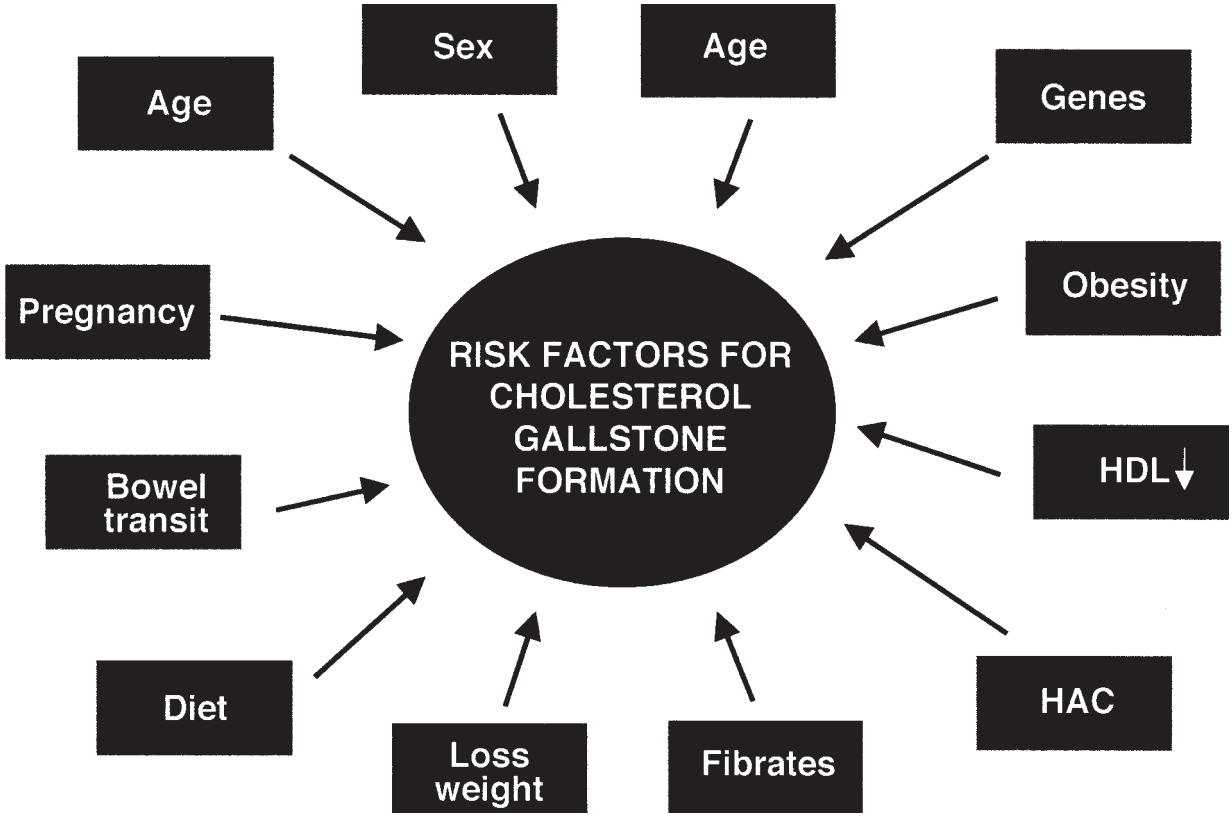

Figure 1. Known risk factors for cholesterol gallstones. The principal are sex (women), age obesity, pregnancy, Amerindian lithogenic genes and hyperinsulinemia with a lower than normal plasma HDL cholesterol concentration.

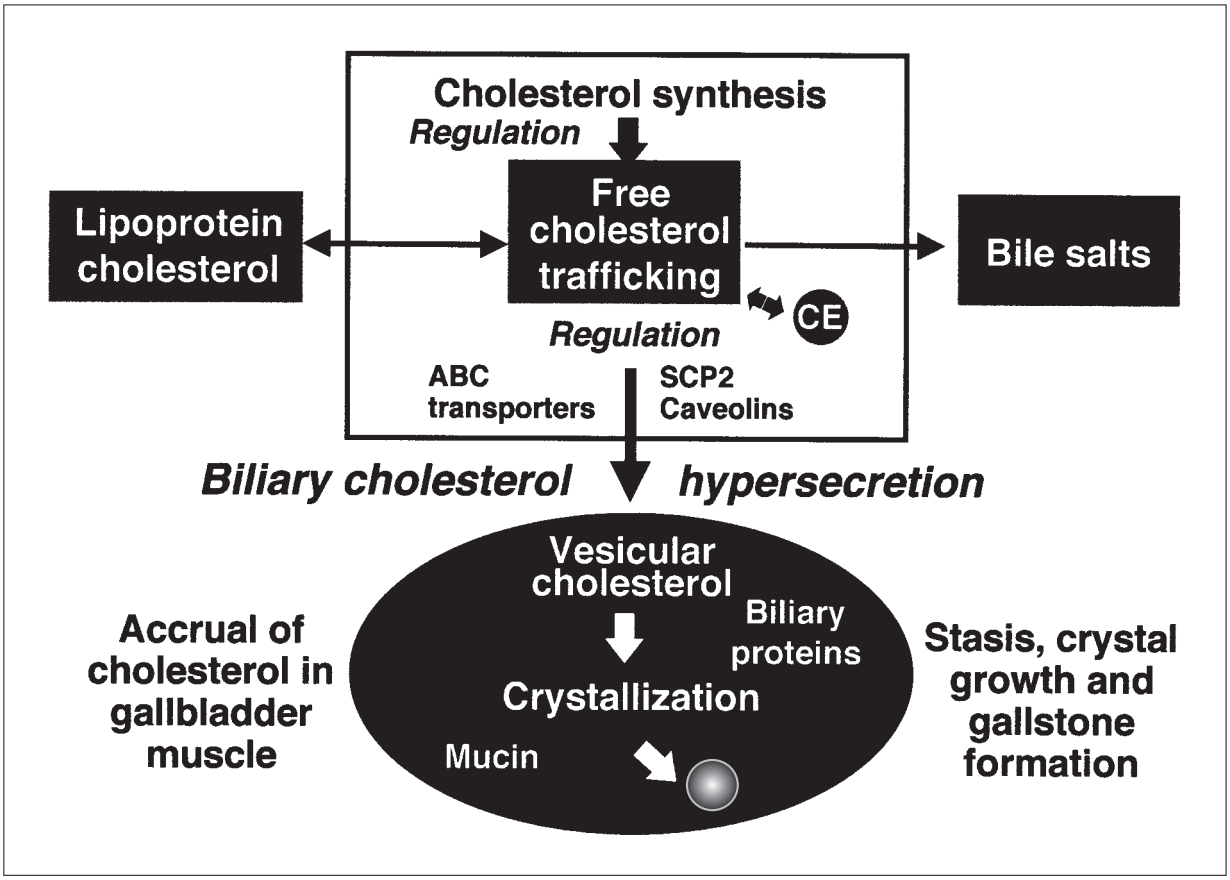

Figure 2. This figure represents the key pathogenic steps that probably interact or have an individual major contribution in the formation of gallstones. The upper rectangle represents the liver. Metabolically active cholesterol is principally located (80%) in the plasma membrane of hepatocytes. The intense trafficking of free cholesterol is a highly complex and regulated function of hepatocytes. Cholesterol is secreted as VLDL and HDL cholesterol through the sinusoidal membrane. It is also delivered to the apical pole of hepatocytes as newly synthesized bile acids and solubilized in unilamellar biliary vesicles. It is thought that biliary cholesterol hypersecretion might result from abnormal regulation of hepatic cholesterogenesis, or increase delivery of cholesterol to the canalicular membrane. A number of proteins that participate in cholesterol in cholesterol transport in the hepatocyte might be altered and responsible for this increase, including SCP-2, sterol carrier protein 2; NPC1, lysosomal Niemman-Pick protein C1; caveolins, ABC 1 canalicular cholesterol transporter.

and/or be influenced through intermediate pathogenic events interrelated to a variety of environmental factors. Biliary cholesterol hypersecretion has been found in Pima Indians[24], normal weight-Chilean Hispanics[25], in Swedish gallstone patients[26] and in obese subjects[27]. Therefore, cholesterol hypersecretion into bile would be a common initiating pathogenic mechanism for cholesterol gallstone formation, as shown in Figure 2. Besides its relationship to obesity, insulin resistance has been found associated with low serum HDL cholesterol concentration, a phenomenon probably dependent of a high catabolic rate of HDL particles[28]. This interpretation could be related to the increased rates of biliary cholesterol secretion found in gallstone patients, since biliary cholesterol mainly derives from HDL cholesterol[29]. Interestingly, recent experiments in mice with genetic manipulation of the HDL receptor SR-BI in the liver were associated to reciprocal changes in in cholesterol secretion into bile and serum HDL cholesterol levels (ultima revisión de Trigatti, Rigotti, Krieger).

It is likely that hepatic cholesterol hypersecretion into bile would be a common final pathogenic mechanism for cholesterol gallstone formation in the majority of cases. Moreover, it has been shown by stepwise logistic regression analysis, that biliary choles-

terol crystallization primarily depends on cholesterol saturation, but not on crystallization promoter biliary proteins[31]. After initial in vitro and correlative evidence supporting the role of these cholesterol precipitation-modulating proteins in gallstone formation, more detailed studies are not consistent with this working hypothesis (Miquel at al Gastro y Carey el at in JLR). However it is generally accepted that mucin proteins are essential for cholesterol crystal aggregation and stone growth[1].

The key molecular abnormalities underlying cholesterol hypersecretion into bile are not yet fully understood. Genes that regulate cholesterol metabolism and trafficking within the hepatocyte are considered potentially important in favoring biliary cholesterol hypersecretion. It has also been postulated that abnormal regulation of hepatic cholesterol synthesis, bile acid synthesis, or esterification, would be the primary metabolic abnormalities in cholesterol gallstone disease[1]. However, studies related to these parameters have given controversial results, particularly those related to hepatic 3-hydroxy-3-methylglutaryl-CoA reductase activity in gallstone patients. Several groups have reported increased enzyme activity, reflecting increased hepatic cholesterogenesis[1]. However, this has not been the case in other studies[1]. More recently, it was found by antisense treatment that modulation of hepatic sterol carrier protein 2 (SCP-2) expression could modify biliary cholesterol secretion and therefore, this carrier protein could play a key role in cholesterol gallstone disease[32]. Indeed, it has been shown that adenovirus-mediated overexpression of SCP2 in mouse liver results in increased biliary cholesterol secretion[40]. Interestingly, preliminary data from Japan has shown that hepatic SCP-2 concentration was increased in gallstone patients[33], suggesting that this protein might be mediating apreferential channeling of hepatic cholesterol into the bile.

Future trends

Research in this area, as occur in all chronic diseases, is principally oriented to find cost – effective measures for secondary and primary prevention. Cholesterol gallstones represent a major health problem in the western world, particularly among Amerindian populations, where gallbladder diseases reach epidemic characteristics[4]. In the last 5 years a number of studies have been oriented to find the underlying abnormalities in the regulation of hepatic cholesterol metabolism and biliary lipid secretion of this disease. Among them, the discovery of the mdr[2] canalicular transporter as a phospholipid flippase that regulates biliary phospholipid secretion has been one of the important discoveries of the last decade[34]. The identification of a number of genes responsible for the expression of proteins that regulate key steps of cholesterol and bile acid synthesis and trafficking, and lipoprotein cholesterol receptor and apolipoprotein expression[35-40] are also emerging as potentially relevant. In addition, high-throughput genome scanning, transcriptional profiling, and proteomic approaches in high risk populations and families might also provides new insights into etiology and pathogenesis of cholelithiasis. These series of new discoveries might help to disclose the molecular abnormalities of hepatic cholesterol metabolism in gallstone patients and in subjects with atherosclerosis, another disturbance of cholesterol solubilization transport that determines deposition of free cholesterol within the arterial wall.

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