無 HFE 基因突變之慢性 C 型肝炎病人肝臟纖維化嚴重度：與肝臟鐵質沉積、血清鐵質指標、C 型肝炎病毒量與基因型之關係

Hepatic Fibrosis Severity in Chronic Hepatitis C Patients without HFE Mutations: Relationships with Hepatic Iron Deposition, Serum Iron Index, Viral Load and Genotype of Hepatitis C Virus

中文摘要

研究目的：

輕度到中度的鐵質過度負擔在慢性 C 型肝炎病人是常見的現象，而鐵質的過度負擔可能扮演一個肝炎進展的角色，且肝臟鐵質的過度負擔可能與造成遺傳性血色素沉著症(hemochromatosis)的隱性遺傳 HFE 基因，包括 C282Y 與 H63D 兩種突變有關，但 HFE 基因突變在東方人並不常見。本篇研究的目的爲，探討在無 HFE 基因突變的慢性 C 型肝炎病人中，肝臟纖維化嚴重度與血清鐵質指標或肝臟鐵質沉積之間的相關性；此外，也要探討其它可能影響這群病人肝臟纖維化嚴重度之潛在因子，包括年齡、性別、肝功能指數、C 型肝炎之病毒量與基因型；在本研究中，我們特別調整會影響肝臟鐵質沉積或肝臟纖維化嚴重度的干擾因子，也就是排除 HFE 基因突變、飲酒過量、肥胖、B 型肝炎與人類免疫不全病毒感染之病人。

研究材料與方法：

我們的研究總共收集了 32 個無 HFE 基因突變的慢性 C 型肝炎病人，我們用超音波指引執行肝臟穿刺取得肝組織，肝臟纖維化嚴重度用 Metavir system 來決定其分期；用 Perls’ stain 評估肝組織鐵質沉積的程度；我們檢驗血清鐵質指標，包括鐵質(iron)與 total iron binding capacity (TIBC)，ferritin；用 reverse transcription-polymerase chain reaction (RT-PCR)的原理檢驗 C 型肝炎之病毒量，並用 type-specific PCR 的方法檢測 C 型肝炎病毒之基因型；每個病人都以 polymerase chain reaction-restriction fragment length polymorphism (PCR-RFLP)的方法分析是否有 HFE 基因突變，以排除其影響。

結果：

收集了 33 個符合條件的病人，其中有一個因爲有 HFE 基因突變(H63D heterozygosity)而被排除，所以總共有 32 個病人。這 32 個病人的平均年齡是 56.47±10.92 歲，14 個病人(43.75%)有血清鐵質指標增加的現象，只有 4 個病人(12.5%)有陽性的肝臟鐵質染色，其中 3 個是第一級，而 1 個病人是第二級的 Perls’ stain。這 32 個病人中，16 個是重度的肝臟纖維化(stage 3 or 4)，而另外 16 個是輕度的肝臟纖維化(stage 0, 1 or 2)。重度肝臟纖維化病人比輕度病人的年齡有顯著意義的大(60.75±6.50 比 52.19±12.85 歲，

P=0.024)；其它變數，包括性別、肝功能指數、血清鐵質指標，肝臟鐵質染色，C 型肝炎病毒量與基因型，在這兩群病人間皆無顯著差異。再以多變量回歸分析，病人年齡仍是一個有意義的肝臟纖維化嚴重度的獨立預測因子(P=0.035, odds ratio=1.312)。

陽性的肝臟鐵質染色與 alanine aminotransferase (ALT) (P=0.017)，三個血清鐵質指標，包括 ferritin (P=0.008)、鐵質(iron) (P=0.019)、transferrin saturation (iron/TIBC) (P=0.003)皆有顯著相關。以 Spearman 相關分析，ferritin 的數值與 ALT (P=0.003)，鐵質(iron) (P=0.011)、transferrin saturation (P=0.002)皆有顯著相關性；然而，ferritin 與肝臟組織發炎活性的等級或纖維化嚴重度的分期之間，並無顯著相關性。

因女性可能比男性有較低的鐵質指標，我們依性別再將病人分爲兩組，結果發現不論男或女組，所有血清鐵質指標與肝臟鐵質染色都與肝臟纖維化的嚴重度無顯著相關。

結論：

慢性 C 型肝炎肝臟纖維化嚴重度與病人之年紀有關；肝臟之鐵質沉積在無 HFE 基因突變之慢性 C 型肝炎病人並不常見，而血清鐵質指標與肝臟鐵質沉積兩者能代表生化上肝臟之慢性發炎狀態，但與 C 型肝炎病毒所造成組織學上肝臟之纖維化分期或發炎等級皆無顯著相關性；C 型肝炎之病毒量或基因型與肝臟組織纖維化或發炎嚴重度亦無顯著相關。

英文摘要

BACKGROUND/AIMS: Mild to moderate iron overload is common in chronic hepatitis C (CHC) and may play a role in the progression of liver disease. That may be related to the recessively inherited HFE mutations, C282Y and H63D; however, they are infrequent in Asia. The aim of this study was to evaluate the association among hepatic fibrosis and serum iron indices, hepatic iron stores in CHC patients without HFE mutations. This study was also performed to assess the other potential factors related to hepatic fibrosis severity in these patients, including age, gender, liver enzyme tests, viral load and genotype of hepatitis C virus (HCV). We had adjusted the other confounders, such as HFE mutations, alcohol abuse, obesity, and concurrent human immunodeficiency virus or hepatitis B virus infection.

PATIENTS AND METHODS: Total 32 CHC patients without HFE mutations were included in our study. The hepatic specimens were obtained with the SURECUT needle by ultrasonography-guided biopsy of liver. The severity of hepatic fibrosis was determined according to the Metavir system. Hepatic iron deposition was assessed by Perls' stain on liver biopsy specimens. We also examined the serum iron markers, including ferritin, iron and total iron binding capacity (TIBC). The method with which the viral load of HCV was checked was based on the principle of reverse

transcription-polymerase chain reaction (RT-PCR). We use the method of type-specific PCR to assess the genotype of HCV and polymerase chain reaction-restriction fragment length polymorphism (PCR-RFLP) to analyze HFE gene mutations.

RESULTS: Thirty-three patients who fulfilled patient criteria were chosen; however, one of these patients was excluded due to the presence of HFE mutation (H63D heterozygous mutation). Thus, 32 patients were studied. The mean age of the 32 patients was 56.47 ± 10.92 year-old. Fourteen patients (43.75%) had increased serum iron stores and only four patients (12.5%) had positive hepatic iron stain. In the four patients, three patients were grade one and one patient was grade two on Perls' stain. Of 32 patients, 16 patients had severe hepatic fibrosis (stage 3 or 4) and 16 patients had mild fibrosis (stage 0, 1 or 2). The CHC patients with severe hepatic fibrosis were significantly older than the CHC patients with mild fibrosis (60.75±6.50 vs 52.19±12.85 year-old, P=0.024). The other variables, including gender, liver enzyme tests, serum iron indices, increased serum iron store, positive hepatic iron stain, viral load and genotype of HCV, were not significantly different between patients with severe and mild hepatic fibrosis. In multivariate logistic regression analysis, the age at biopsy was still an independent predictor of severe hepatic fibrosis (P=0.035, odds ratio= 1.312). The positive hepatic iron stain was significantly associated with the values of alanine aminotransferase (ALT) (P=0.017) and all the three serum iron indices, including ferritin (P=0.008), iron (P=0.019) and tranferrin saturation (iron/TIBC) (P=0.003). The ferritin level had significant correlation with the value of ALT, iron and transferrin saturation in Spearman correlation test (P=0.003, 0.011 and 0.002, respectively). Nonetheless, no significant correlation was found between ferritin and grade of inflammation activity or stage of hepatic fibrosis severity. We stratified our data according to patient sex because women may have lower serum iron markers than men. All the serum iron indices and hepatic iron stain were not associated with severe hepatic fibrosis in men and women, respectively.

CONCLUSIONS: The severity of HCV-related liver injury is associated with patient age at biopsy. Significant iron deposition in the liver is uncommon in CHC patients without HFE mutations. Both serum iron indices and hepatic iron deposition have correlation with biochemically chronic inflammation condition of liver but they are unrelated to the grade and stage of histologically HCV-related liver injury. The viral load and genotype of HCV are also not associated with hepatic fibrosis severity and inflammation activity.