Intrahepatic Expression of C-C Motif ligand 5 in Patients with Chronic Hepatitis B

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ABSTRACT

Background: *C-C motif ligand 5 (CCL5) is reported to play a key role in acute and chronic liver diseases. However, the association between CCL5 and chronic hepatitis B (CHB) remains to be explored. We aimed to investigate the CCL5 expression in the liver tissues of CHB patients and compared the CCL5 expression among CHB patients with different stages of liver inflammation and fibrosis.*

Methods: *Liver tissue specimens from 51 CHB patients who underwent liver biopsy and twelve healthy liver donors were included in the present study. CCL5 expression in the liver tissues was analyzed using immunohistochemistry. The hepatic inflammation grades and fibrotic stages of CHB patients were assessed by the Scheuer classification system.*

Results: *Livers of CHB patients exhibited significantly accumulated CCL5+ cells when compared to those of healthy controls (42.80 ± 4.37 vs. 7.25 ± 0.99/HPF, P < .001). CHB patients with higher hepatic inflammation grades had more CCL5+ cells in their livers than those with lower grades (P < .05). However, the numbers of CCL5+ cells were not correlated with the fibrotic stages in CHB patients (r = .073, P = .61). The number of CCL5+ cells in the liver tissues of CHB patients was positively correlated with alanine transaminase levels (r = .278, P = .041) and aspartate aminotransferase levels (r = .328, P = .009).*

Conclusions: *CHB patients have a significant accumulation of CCL5+ cells in the liver, and CCL5 may play a pathological role in hepatic inflammation of CHB.*

Keywords: *CCL5, CHB, hepatic fibrosis, hepatic inflammation*

INTRODUCTION

Chronic hepatitis B (CHB) is a major health problem worldwide, especially in Asia.[1,2] Around 2 billion people worldwide are infected with hepatitis B virus (HBV), of whom 240 million are CHB patients.[1] Chronic HBV infection can cause persistent hepatic inflammation, leading to the apoptosis of parenchymal cells and replacement by connective tissue and extracellular matrix (ECM) proteins, inducing hepatic fibrosis and cirrhosis.[3,4] It is estimated that about 30% of cirrhosis cases were caused by HBV infection, and around 650 000 people die each year due to liver failure and hepatocellular carcinoma caused by HBV.[5,6]

In the pathophysiological process of CHB, chemokines, such as monocyte chemotactic protein-1 (MCP-1), inducible protein-10 (IP-10), and C-C motif ligand 5 (CCL5) were secreted in large quantities, leading to infiltration of various leukocytes, and pro-inflammatory and pro-fibrotic cytokine release, which aggravates hepatic inflammation and fibrosis.[7-10] Inhibition of chemokine secretion may alleviate hepatic inflammation and fibrosis.[11-14] Recent studies have found that serum CCL5 levels are positively correlated with alanine aminotransferase (ALT) levels in patients with HBV infection, suggesting that CCL5 may be associated with hepatic inflammation.[15]

CCL5, also known as regulated upon activation normal T cell expressed and secreted factor (RANTES), mainly come from macrophages and T cells.[16,17] CCL5 is able to recruit a variety of leukocytes by binding cellular surface

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receptors-C-C chemokine receptor (CCR)1, CCR3, and CCR5, which means that CCL5 can aggravate the inflammation of insult.[18] Besides, recent studies have found that hepatic stellate cells (HSCs) highly express CCR5, which has a positive effect on promoting liver fibrosis, suggesting that CCL5 is able to directly activate HSCs and promote liver fibrosis.[19-21] These findings indicate that CCL5 may play a role in the inflammation and fibrosis of the liver.

Furthermore, CCL5 has been demonstrated to have proinflammatory and profibrotic effects in acute liver injury and nonalcoholic fatty liver disease (NAFLD).[22-24] With regard to viral hepatitis, hepatitis C virus (HCV)-induced CCL5 secretion from macrophages can activate HSCs, promote hepatic fibrosis, and aggravate hepatocellular carcinoma (HCC) development.[13] In patients with CHB, the serum CCL5 level increased significantly with aggravated liver injury.[25] Wu et al. also evaluated the serum CCL5 profile in acute exacerbation of CHB patients.[10] However, few studies have determined the intrahepatic CCL5 expression in CHB patients. In the present study, we analyzed the CCL5 expression in the liver tissues of CHB patients and explored the association of the CCL5 expression with hepatic inflammation and fibrosis in CHB patients.

METHODS

Patients and Specimens

Paraffin-embedded liver specimens were selected from the Department of Pathology of the Nanjing Drum Tower Hospital and were further used according to the decision of the Ethics Committee of the Nanjing Drum Tower Hospital (Date: February 15, 2017; 2017022). Hepatic specimens from 51 CHB patients were included in the present study. CHB patients with other chronic liver diseases including HCC, nonalcoholic fatty liver disease (NAFLD), and autoimmune liver diseases were excluded. The characteristics of the CHB patients are presented in Table 1. Twelve healthy hepatic tissue samples (HC) were collected from donors whose livers were used for transplantation. Every patient of this study had received informed consent and agreed to the study.

Histology

Hepatic tissues embedded in paraffin were cut into 2 μm sections and stained with hematoxylin-eosin or Masson according to standard protocols. The degree of hepatic inflammation and fibrosis was graded according to the Scheuer classification system.[26] Inflammatory activity was evaluated according to a 5-grade scale (G) and hepatic fibrosis was assessed by a 5-stage scale (S).[26]

Table 1. Clinical Features of the Patients with Chronic HBV Infection

	CHB (n = 51)
Age (year)	49.87 ± 11.33
Gender (M/F)	28/23
ALT (U/L)	51.25 ± 41.28
AST (U/L)	60.42 ± 62.22
ALP (U/L)	82.86 ± 40.16
GGT (U/L)	52.89 ± 48.545
Tbil (μmol/L)	39.24 ± 28.19
Alb (g/L)	36.15 ± 5.21
HBeAg positive (%)	16 (31.37)
HBV DNA (Log10 IU/mL)	4.33 ± 1.54
Inflammation grades	
G1/G2/G3/G4	16/17/13/5
Fibrotic stages	
S1/S2/S3/S4	10/11/13/17

Data are expressed as the mean ± standard error (SEM).
Alb, albumin; ALP, alkaline phosphatase; ALT, alanine aminotransferase; AST, aspartate aminotransferase; GGT, gamma-glutamyl transferase; HBV, hepatitis B virus; Tbil, total bilirubin.

Immunohistochemistry

CCL5 immunohistochemistry was performed as described previously.[27] Briefly, the prepared sections of the liver were dewaxed, hyalinized, and antigen repaired in order. Then, the sections were incubated with primary antibody rabbit anti-CCL5 (ab9679, Abcam) overnight at 4°C. The sections were then stained with goat anti-rabbit immunoglobulin G (IgG) (GV800, Dako) at 37°C for 30 minutes. At last, the sections were slightly stained with hematoxylin and fixed with neutral balata.

CCL5+ cell counts per high-power field (HPF) were performed as described previously.[27] Briefly, every section was randomly captured to 5 × 400 magnification images. Positive CCL5 staining cells per HPF of each image were manually assessed by 2 experienced pathologists.

Statistical Analysis

All data are expressed as the mean ± standard error (SEM). Statistical comparisons between 2 groups were performed using the Student's t test or Mann-Whitney U test. Multiple comparisons were performed using the ANOVA

test and then the Student-Newman-Keuls (SNK) test for multiple comparisons. Correlation analysis was performed using the Spearman rank correlation test. A two-sided value of $P < .05$ was considered statistically significant.

RESULTS

Patient Characteristics

Liver specimens from 51 CHB patients and 12 healthy liver tissue specimens for liver donors were included in the present study. The mean age of CHB patients was 49.87 ± 11.33 years. Twenty-three subjects (45.01%) were female and 16 subjects (31.37%) were HBeAg positive. The distribution of each inflammation grade in the subjects was as follows: G1, 16 patients; G2, 17 patients; G3, 13 patients; and G4, 5 patients. The distribution of each fibrotic stage in the subjects was as follows: S1, 10 patients; S2, 11 patients; S3, 13 patients; and S4, 17 patients. Other detailed demographic and laboratory parameters of the subjects are shown in Table 1.

Intrahepatic Accumulation of CCL5 in CHB Patients

As shown in Figure 1A and B, there was a small amount of positive CCL5 staining cells in the liver of healthy controls. The livers of CHB patients exhibited significantly increased positive CCL5 staining cells (42.80 ± 4.37/HPF) as compared to those of normal tissues (7.25 ± 0.99/HPF, $P < .001$). Besides, CCL5 staining cells were mainly accumulated in the periportal sinusoidal domain of liver tissues in CHB patients.

Association of Intrahepatic Accumulation of CCL5+ Cells with Hepatic Inflammation Grades and Fibrotic Stages in CHB Patients

Compared with those patients who had lower hepatic inflammation grades (G), patients with higher grades had more CCL5+ cells in their livers (Figure 2A). Remarkable increased CCL5+ cells were found in the liver tissues of patients with different fibrotic stages (S) compared with those of healthy controls. However, the CCL5+ cells were not significantly different among CHB patients with different fibrotic stages (Figure 2B). The inflammation distributions were not significantly different among CHB patients with different fibrotic stages ($P = .169$, Supplementary Figure 1). The correlation analysis showed that the inflammation grades ($r = .255$, $P = .040$), but not fibrotic stages ($r = .073$, $P = .610$), were positively correlated with the density of CCL5+ cells (Figure 3A and B) in CHB patients.

The Clinical Relevance of CCL5 in CHB Patients

We further analyzed the association between the number of CCL5+ cells and the clinical parameters. As shown

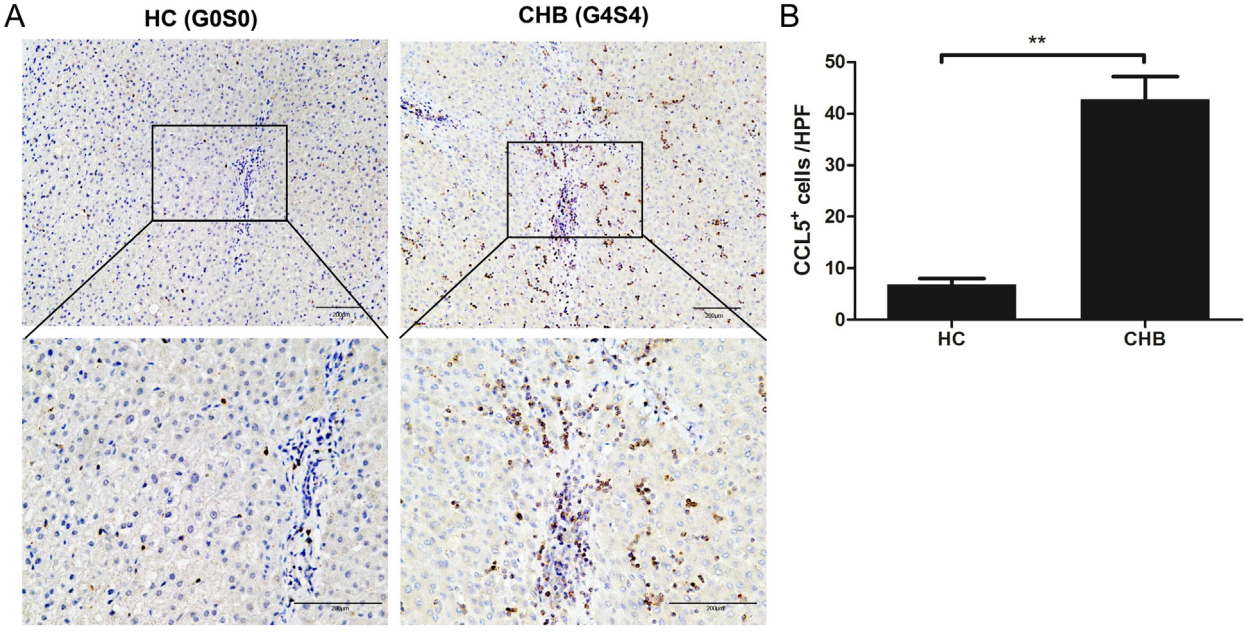

Figure 1. Immunohistochemical analysis of CCL5+ cells in the liver tissues of CHB patients and healthy control. (A) Representative figures of CCL5 immunohistochemistry of healthy controls and CHB patients. (B) Comparison of CCL5+ cells quantity between healthy controls and CHB patients. CHB, chronic hepatitis B; G, inflammation grades; HC, healthy control; HPF, high-power field; S, fibrotic stages. $^{**}P < .01$. Bar = 200 μm.

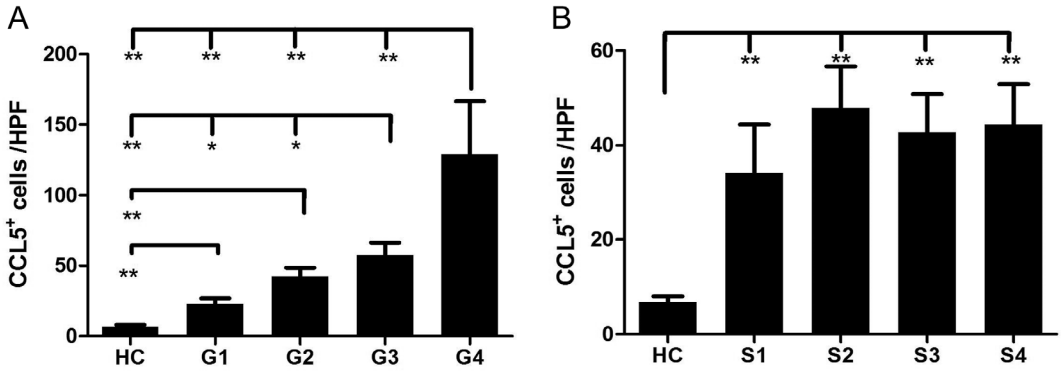

Figure 2. CCL5+ cells in CHB patients with different inflammation grades (A) and fibrotic stages (B). G, inflammation grades; S, fibrotic stages. *P < .05, **P < .01.

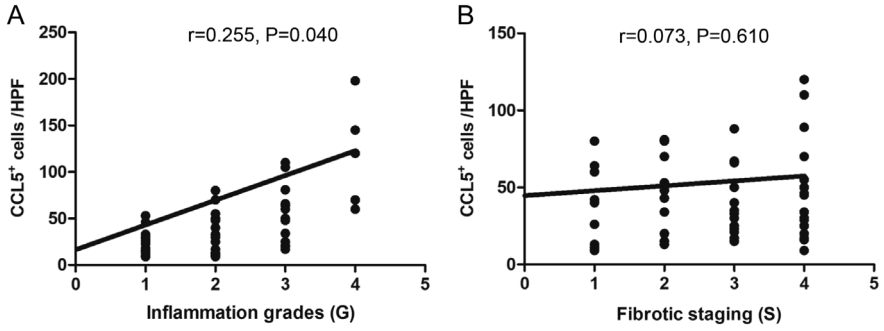

Figure 3. Correlation analyses between inflammation grades (A), fibrotic stages (B), and CCL5+ cells quantity in CHB patients.

Table 2. The Clinical Relevance of CCL5+ Cells in Patients with Chronic HBV Infection

	R	*P* value
ALT	0.278	.041
AST	0.328	.009
ALP	−0.043	.678
GGT	0.013	.545
Tbil	0.224	.061
Alb	−0.034	.901
HBV DNA	−0.137	.945
FIB-4	0.237	.284
APRI	0.164	.185

Alb, albumin; ALP, alkaline phosphatase; ALT, alanine aminotransferase; APRI, aspartate aminotransferase-to-platelet ratio index; AST, aspartate aminotransferase; FIB-4, Fibrosis-4 score; GGT, gamma-glutamyl transferase; HBV, hepatitis B virus; Tbil, total bilirubin.

in Table 2, both the serum alanine transaminase (ALT) (r = .278, P = .041) and aspartate aminotransferase (AST) (r = .328, P = .009) of CHB patients were positively correlated with the numbers of CCL5+ cells. However, there were no correlations between CCL5+ cells' numbers and other clinical indicators, including albumin (Alb) (r = −.034, P = .901), alkaline phosphatase (ALP) (r = −.043, P = .678), gamma-glutamyl transferase (GGT) (r = .013, P = .545), total bilirubin (Tbil) (r = .224, P = .061), HBV DNA (r = −.137, P = .945), aspartate aminotransferase-to-platelet ratio index (APRI) (r = .164, P = .185), and Fibrosis-4 scores (FIB-4) (r = .237, P = .284).

DISCUSSION

During the pathophysiological process of CHB, a large number of leukocytes, such as T cells and macrophages, are mobilized and recruited into the liver.[3,28-30] CCL5, mainly secreted by macrophages and T cells, is able to recruit a variety of leukocytes to the inflammatory sites.[18]

The elevated expression of CCL5 was observed in several types of diseases, such as asthma, autoimmune diseases, cancer, and atherosclerosis.[31-35] In recent years, more and more studies have focused on CCL5 in acute and chronic liver diseases, and its role in promoting inflammation and fibrosis has also been recognized.[23,36] For example,

Kim et al. revealed that CCL5 contributes to hepatic steatosis and inflammation in mice fed with choline-deficient, L-amino acid-defined, high-fat diet.[24] Li et al. supported that during NAFLD, CCAAT/enhancer-binding protein β (C/EBPβ) induced unregulated CCL5 expression, which plays a pivotal regulatory role in hepatic fibrosis.[37] The role of CCL5 in liver fibrosis is further strengthened by a study from Berres et al., in which, compared to wild type mice, $CCl5^{-/-}$ mice intraperitoneally injected with carbon tetrachloride (CCl_4) or fed on a methionine and choline-deficient (MCD) diet exhibited decreased hepatic fibrosis and reduced stellate cell activation and immune cell infiltration.[14] CCL5 secretion from macrophages induced by HCV infection can promote hepatic fibrosis and aggregate HCC development via HSCs activation.[13] Hu et al. reported that the serum CCL5 level increased in CHB patients and significantly decreased in cirrhosis patients.[25]

In the present study, we investigated CCL5 expression in the liver tissues of CHB patients. Firstly, we observed that the CCL5 expression was significantly elevated in the liver tissues of CHB patients and positively with hepatic inflammation grades. Besides, there was a positive correlation between CCL5 and liver enzymes such as ALT and AST. These results suggest that CCL5 plays a potential pathological role in hepatic inflammation of CHB patients.

In our present study, intrahepatic CCL5 expression is not correlated with the stage of fibrosis in chronic CHB patients. However, many researches have confirmed the role of CCL5 in liver fibrosis in vitro and in vivo.[13,14,36] It should be acknowledged that the sample size of our study was relatively small. Thus, the association of intrahepatic CCL5 expression and hepatic fibrosis deserves further investigation.

In conclusion, our study demonstrated that CHB patients have a significant accumulation of CCL5 in the liver. The study provides additional evidence on the important role of CCL5 in the pathogenesis of CHB. Future efforts shall focus on the biological functions of CCL5 in CHB.

Ethics Committee Approval: The study has been approved by the Ethics Committee of Nanjing Drum Tower Hospital.

Informed Consent: The patients had received informed consent.

Peer-review: Externally peer-reviewed.

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REFERENCES

1. Zampino R, Boemio A, Sagnelli C, et al. Hepatitis B virus burden in developing countries. World J Gastroenterol. 2015;21(42):11941-11953. [CrossRef]
2. Kwon SY, Lee CH. Epidemiology and prevention of hepatitis B virus infection. Korean J Hepatol. 2011;17(2):87-95. [CrossRef]
3. Bertoletti A, Le Bert N. Immunotherapy for chronic hepatitis B virus infection. Gut Liver. 2018;12(5):497-507. [CrossRef]
4. Seeger C, Mason WS. Molecular biology of hepatitis B virus infection. Virology. 2015;479-480:672-686. [CrossRef]
5. Lozano R, Naghavi M, Foreman K, et al. Global and regional mortality from 235 causes of death for 20 age groups in 1990 and 2010: a systematic analysis for the Global Burden of Disease Study 2010. Lancet. 2012;380(9859):2095-2128. [CrossRef]
6. Goldstein ST, Zhou F, Hadler SC, Bell BP, Mast EE, Margolis HS. A mathematical model to estimate global hepatitis B disease burden and vaccination impact. Int J Epidemiol. 2005;34(6):1329-1339. [CrossRef]
7. Chen XJ, Ye HH, Xing LX, et al. Association of mRNA expression level of IP-10 in peripheral blood mononuclear cells with HBV-associated acute-on-chronic liver failure and its prognosis. Hua Zhong Keji Da Xue Xuebao Yixue Yingdewen Ban. 2017;37:755-760.
8. Pellicoro A, Ramachandran P, Iredale JP, Fallowfield JA. Liver fibrosis and repair: immune regulation of wound healing in a solid organ. Nat Rev Immunol. 2014;14(3):181-194. [CrossRef]
9. Wynn TA, Ramalingam TR. Mechanisms of fibrosis: therapeutic translation for fibrotic disease. Nat Med. 2012;18(7):1028-1040. [CrossRef]
10. Wu HL, Kao JH, Chen TC, et al. Serum cytokine/chemokine profiles in acute exacerbation of chronic hepatitis B: clinical and mechanistic implications. J Gastroenterol Hepatol. 2014;29(8):1629-1636. [CrossRef]
11. Zaldivar MM, Pauels K, von Hundelshausen P, et al. CXC chemokine ligand 4 (CXCL4) is a platelet-derived mediator of experimental liver fibrosis. Hepatology. 2010;51(4):1345-1353. [CrossRef]
12. Baeck C, Wei X, Bartneck M, et al. Pharmacological inhibition of the chemokine C-C motif chemokine ligand 2 (monocyte chemoattractant protein 1) accelerates liver fibrosis regression by suppressing Ly-6C(+) macrophage infiltration in mice. Hepatology. 2014;59(3):1060-1072. [CrossRef]

13. Sasaki R, Devhare PB, Steele R, Ray R, Ray RB. Hepatitis C virus-induced CCL5 secretion from macrophages activates hepatic stellate cells. Hepatology. 2017;66(3):746-757. [CrossRef]
14. Berres ML, Koenen RR, Rueland A, et al. Antagonism of the chemokine Ccl5 ameliorates experimental liver fibrosis in mice. J Clin Invest. 2010;120(11):4129-4140. [CrossRef]
15. Wang JW, Wang LY, Zhao ZH, et al. Correlationship between chemokines and oxidative stress in chronic hepatitis B. Zhonghua Shi Yan He Lin Chuang Bing Du Xue Za Zhi. 2012;26(4):246-249.
16. Appay V, Rowland-Jones SL. RANTES: a versatile and controversial chemokine. Trends Immunol. 2001;22(2):83-87. [CrossRef]
17. Chenoweth MJ, Mian MF, Barra NG, et al. IL-15 can signal via IL-15Rα, JNK, and NF-κB to drive RANTES production by myeloid cells. J Immunol. 2012;188(9):4149-4157. [CrossRef]
18. Pakianathan DR, Kuta EG, Artis DR, Skelton NJ, Hébert CA. Distinct but overlapping epitopes for the interaction of a CC-chemokine with CCR1, CCR3 and CCR5. Biochemistry. 1997;36(32):9642-9648. [CrossRef]
19. Puche JE, Saiman Y, Friedman SL. Hepatic stellate cells and liver fibrosis. Compr Physiol. 2013;3(4):1473-1492. [CrossRef]
20. Gupta G, Khadem F, Uzonna JE. Role of hepatic stellate cell (HSC)-derived cytokines in hepatic inflammation and immunity. Cytokine. 2018:234-243.
21. Seki E, De Minicis S, Gwak GY, et al. CCR1 and CCR5 promote hepatic fibrosis in mice. J Clin Invest. 2009;119(7):1858-1870. [CrossRef]
22. Roh YS, Seki E. Chemokines and chemokine receptors in the development of NAFLD. Adv Exp Med Biol. 2018;1061:45-53. [CrossRef]
23. Pérez-Martínez L, Pérez-Matute P, Aguilera-Lizarraga J, et al. Maraviroc, a CCR5 antagonist, ameliorates the development of hepatic steatosis in a mouse model of non-alcoholic fatty liver disease (NAFLD). J Antimicrob Chemother. 2014;69(7):1903-1910. [CrossRef]
24. Kim BM, Abdelfattah AM, Vasan R, Fuchs BC, Choi MY. Hepatic stellate cells secrete Ccl5 to induce hepatocyte steatosis. Sci Rep. 2018;8(1):7499. [CrossRef]
25. Hu L, Zhu Y, Zhang J, et al. Potential circulating biomarkers of circulating chemokines CCL5, MIP-1β and HA as for early detection of cirrhosis related to chronic HBV (hepatitis B virus) infection.. BMC Infect Dis. 2019;19(1):523.
26. Scheuer PJ. Classification of chronic viral hepatitis: a need for reassessment. J Hepatol. 1991;13(3):372-374. [CrossRef]
27. Zhao XA, Chen G, Liu Y, et al. Curcumin reduces Ly6C(hi) monocyte infiltration to protect against liver fibrosis by inhibiting kupffer cells activation to reduce chemokines secretion. Biomed Pharma. 2018:106;868-878.
28. Kapoor R, Kottilil S. Strategies to eliminate HBV infection. Future Virol. 2014;9(6):565-585. [CrossRef]
29. Sandhu P, Haque M, Humphries BT, et al. Hepatitis B virus immunopathology, model systems, and current therapies. Front Immunol. 2017;8:436.
30. Peeridogaheh H, Meshkat Z, Habibzadeh S, et al. Current concepts on immunopathogenesis of hepatitis B virus infection. Virus Res. 2018;245:29-43. [CrossRef]
31. Ansari AW, Heiken H, Moenkemeyer M, Schmidt RE. Dichotomous effects of C-C chemokines in HIV-1 pathogenesis. Immunol Lett. 2007;110(1):1-5. [CrossRef]
32. Gao D, Fish EN. Chemokines in breast cancer: regulating metabolism. Cytokine. 2018;109:57-64. [CrossRef]
33. Donatella A, Naike C. Inhibition of the CCL5/CCR5 axis against the progression of gastric cancer. Int J Mol Sci. 2018;19:1477.
34. Suffee N, Richard B, Hlawaty H, Oudar O, Charnaux N, Sutton A. Angiogenic properties of the chemokine RANTES/CCL5. Biochem Soc Trans. 2011;39(6):1649-1653. [CrossRef]
35. Marques RE, Guabiraba R, Russo RC, Teixeira MM. Targeting CCL5 in inflammation. Expert Opin Ther Tar. 2013;17(12):1439-1460. [CrossRef]
36. Stock MK, Hammerich L, do O NT, et al. Met-CCL5 modifies monocyte subpopulations during liver fibrosis regression. Int J Clin Exp Pathol. 2013;6(4):678-685.
37. Li BH, He FP, Yang X, Chen YW, Fan JG. Steatosis induced CCL5 contributes to early-stage liver fibrosis in nonalcoholic fatty liver disease progress. Transl Res. 2017;180:103.e4-117.e4. [CrossRef]