JGH Open An open access journal of gastroenterology and hepatology Open Access

JGHF Journal of Gastroenterology and Hepatology Foundation

doi:10.1002/jgh3.12707

BRIEF REPORT

Elevated blood monocyte counts in alcohol-associated hepatitis

Jordan K. Voss,*[,1] Zhuan Li[†] and Steven A. Weinman[†]

*School of Medicine, University of Kansas Medical Center, [†]Department of Internal Medicine, University of Kansas Medical Center, Kansas City, Kansas, USA

Key words
alcoholic liver disease, hepatic inflammation, liver immunobiology.

Accepted for publication 5 January 2022.

Correspondence
Steven A. Weinman, Department of Internal Medicine, University of Kansas Medical Center, 3901 Rainbow Blvd., Kansas City, KS 66160, USA.
Email: sweinman@kumc.edu

Present address
[1]Department of Internal Medicine, Mayo Clinic, Rochester, Minnesota, USA

Declaration of conflict of interest: None.

Funding support: National Center for Advancing Translational Sciences TR002368-02UL1TR002366

Funding support: National Institute on Alcohol Abuse and Alcoholism AA026025

Funding support: National Natural Science Foundation of China 81974458

Introduction

Alcohol-associated hepatitis (AH) is a severe inflammatory condition characterized by acutely worsening liver function in the setting of prolonged heavy drinking. AH is a distinct form of alcohol-associated liver disease that may occur in patients without prior features of alcohol-associated liver disease or may occur in the setting of underlying fibrosis or cirrhosis. Hepatic macrophages play a key role in the pathogenesis and resolution of inflammation in AH.[1] In addition, studies in animal models of AH have demonstrated that circulating monocytes infiltrate the liver and contribute to the pathogenesis of AH.[2] Current treatment for AH is limited to glucocorticoids, which have shown modest benefit in patients with severe AH, likely through broad anti-inflammatory effects.[3] Newer experimental therapies in AH include reduction of circulating granulocytes and monocytes through apheresis.[4,5]

Despite the evidence that monocytes play a key role in the development and progression of alcohol-associated liver diseases and their potential therapeutic relevance, there are few studies that evaluate the monocyte count in patients with AH. In a brief report describing a cohort of 12 patients, McKeever *et al.* demonstrated that patients with AH have elevated absolute monocyte count (AMC) compared to healthy controls.[6] Verghis *et al.* showed that patients with severe AH have elevated monocyte counts with impaired monocyte oxidative burst in a cohort of 42 patients.[7] To our knowledge, these represent the only studies of AMC in AH. Given the small sample size contained in these previous studies, we sought to confirm these findings in a larger retrospective cohort. Additionally, we sought to evaluate the relationship between AMC in patients with AH and disease severity and prognosis.

Methods

Study population. Patients aged 18–70 years admitted to a tertiary care academic medical center hospital with an ICD-9 or ICD-10 diagnosis code associated with alcoholic hepatitis were identified retrospectively. Patients with ICD codes for infection

(sepsis, spontaneous bacterial peritonitis, urinary tract infection, pneumonia), immunodeficiency, HIV, or hematologic malignancy were excluded. The full listing of ICD codes used for inclusion and exclusion can be found in Supplementary Material. Based on the recommendations from the National Institute on Alcohol Abuse and Alcoholism (NIAAA) Alcoholic Hepatitis Consortia, the following laboratory inclusion criteria, which are consistent with a probable diagnosis of AH, were applied: AST >50; AST/ALT ratio >1.5; AST and ALT <400, and total bilirubin >3 mg/dL.[8] Patients with an ICD-9 or ICD-10 diagnosis code associated with alcohol-associated liver disease other than AH who also met the laboratory inclusion criteria were included if they had a liver biopsy within 14 days prior to or 30 days after admission showing histologic features consistent with AH. A histologic diagnosis of AH was defined as macrovesicular steatosis plus one or more of the following: neutrophil infiltration, hepatocyte ballooning, or Mallory–Denk bodies. This histologic definition is also consistent with recommendations from the NIAAA Alcoholic Hepatitis Consortia.[8]

Only the patient's first recorded admission for AH was included in the study. Subsequent readmissions for AH were excluded. Initial and follow-up AMC, absolute lymphocyte count (ALC), absolute neutrophil count (ANC), and white blood cell count (WBC) were determined. Follow-up AMC on hospital day 10 (±2 days) was recorded. Clinical outcomes including the MELD score, Maddrey discriminant function (MDF), and 30-, 90-, 180-day, and 1-year mortality were assessed. MELD and MDF scores were calculated using the component values from a patient's first lab report obtained after admission. MDF score was estimated from the international normalized ratio (INR) value assuming a prothrombin control time of 12.0 s and an international sensitivity index of 1.0. Data were automatically extracted through the center's I2B2-based electronic health record research data repository, the design of which has been previously described.[9,10] Data cleaning was conducting using SQLiteStudio version 3.2.1. Pathology reports were manually reviewed for patients receiving a liver biopsy.

Statistical analysis. Continuous variables were evaluated for normality using the Shapiro–Wilk test and quantile–quantile plots. One-sample Student's *t*-test and the Wilcoxon signed-rank tests were used to compare the absolute blood counts with the clinical laboratory's normal reference ranges. Spearman's correlation coefficient was used to evaluate relationships between MELD, MDF, and AMC. AMC was compared between cases stratified as mild (MDF ≤32) or severe (MDF >32) using the two-sample *t*-test and the nonparametric Wilcoxon Mann–Whitney test. Logistic regression was used to evaluate the effect of AMC on 30-, 90-, 180-day, and 1-year mortality. All statistical analyses were conducted using SAS version 9.4 (Cary, NC, USA).

Table 1 Demographics, comorbidities, and clinical laboratory data ($N = 164$)

Demographics		
Age, median (IQR)	48	(38–54)
Female, *n* (%)	60	(37)
Race, *n* (%)		
White	126	(77)
Black	14	(9)
Other	22	(13)
Unknown	2	(1)
Ethnicity, *n* (%)		
Non-Hispanic, Latino, or Spanish Origin	144	(78)
Hispanic, Latino, or Spanish	18	(11)
Not recorded	1	(1)
Patient characteristics		
HCV infection, *n* (%)	33	(20)
HBV infection, *n* (%)	5	(3)
MELD, mean ± SD	26.2	± 6.8
MDF, median (IQR)	53	(32–84)
Total bilirubin (mg/dL), median (IQR)	10.3	(5.9–18.2)
AST (U/L), median (IQR)	139	(96–200)
ALT (U/L), median (IQR)	56	(32–68)

Cell counts	Mean	95% CI	Reference range[‡]	*P* value[§]
White cell count ($\times 10^9$/L)	11.3	(10.3–12.4)	4.5–11.0	0.3350
Monocyte count ($\times 10^9$/L)	0.95	(0.87–1.02)	0.0–0.8	**0.0015**
Neutrophil count ($\times 10^9$/L)	8.3	(7.5–9.0)	1.8–7.0	0.0903
Lymphocyte count ($\times 10^9$/L)	1.3	(1.2–1.5)	1.0–4.8	n/a[¶]
Monocyte count Day 10 ($\times 10^9$/L)[†]	1.1	(0.91–1.31)	0.0–0.8	**0.0039**

[†] $n = 37$ for day 10 (±2 days) monocyte count.

[‡]Reference range refers to the clinical laboratory's normal reference range for the specified cell count.

[§]*P*-values are in comparison to the upper limit of the normal reference range.

[¶]*P*-value is omitted for ALC, as the mean is within the normal reference ranges.

AH, alcohol-associated hepatitis; ALT, alanine aminotransferase; AST, aspartate aminotransferase; HBV, hepatitis B virus; HCV, hepatitis C virus; MDF, Maddrey discriminant function; MELD, model for end-stage liver disease.

Results

The patient and clinical characteristics are summarized in Table 1. The mean WBC (11.33×10^9 cells/L), ALC (1.3×10^9 cells/L), and ANC (8.26×10^9 cells/L) were not significantly higher than the normal reference ranges. The mean AMC was 0.95×10^9 cells/L (Fig. 1), which is significantly higher than the upper limit of normal ($P = 0.0015$). The follow-up AMC on day 10 (±2 days), which was available for 37 of the 164 patients, was also significantly elevated (mean = 1.11×10^9 cells/L, $P = 0.0039$).

AMC was found to be positively correlated with the MELD score with a Spearman's correlation coefficient (*R*) of

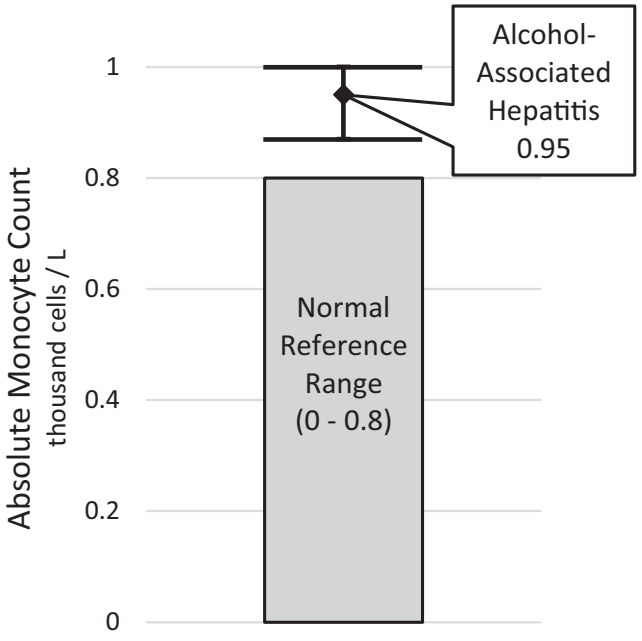

Figure 1 Mean absolute monocyte count in patients with alcohol-associated hepatitis, 950 cells/L, which is significantly higher than the upper limit of normal (800 cells/μL) ($P < 0.0001$).

0.400, $P < 0.0001$. AMC was similarly correlated with the Maddrey discriminant function score ($R = 0.330$, $P < 0.0001$). The mean monocyte count in patients with severe AH was 1010 cells/μL, which is significantly higher than in patients with mild AH (MDF <32) who had a mean monocyte count of 750 ($P = 0.0011$). Similarly, patients with an MELD score of >20 have a higher mean AMC (1020 cells/μL) compared to patients with an MELD score ≤20 (mean AMC 710 cells/μL) ($P = 0.0002$).

Mortality at 30, 90, 180 days, and 1 year was 9, 14, 17, and 24%, respectively. Initial AMC following admission for AH was not associated with increased odds of 30-, 90-, 180-day, or 1-year mortality. Follow-up AMC on day 10 (±2 days) was not associated with increased odds of mortality at 30 days (OR 5.30, 95% CI 1.00–28.19) or 90 days (OR 2.55, 95% CI 0.69–9.50). However, day 10 (±2 days) AMC was weakly associated with increased odds of mortality at 180 days (OR 3.92, 95% CI 1.01–15.31), and 1 year (OR 4.33, 95% CI 1.10–17.05), although the 95% confidence intervals approach 1 and are wide, indicating a high level of uncertainty in the point estimates.

Discussion

This retrospective cohort study supports the findings from McKeever *et al*. and Vergis *et al*. of an elevated AMC in AH, although in a much larger cohort of patients than has previously been studied.[6,7] We also find that the AMC is positively correlated with disease severity as determined by MELD and MDF scores. Monocyte count at the time of admission was not associated with increased odds of mortality. To our knowledge, this represents the first study to assess the relationship between monocyte count and severity of AH.

This study has several limitations. First, the study relied on retrospective data, which were originally collected for clinical and administrative purposes rather than for research purposes. As such, we were unable to account for the effect of potential confounders such as ongoing alcohol use or relapse on mortality. However, we have no reason to believe that the baseline or day-10 monocyte count would be higher (or lower) in those who continue to drink or relapse subsequent to the time of the index hospitalization. Second, the day-10 monocyte count was available only for 37 out of 164 patients in the AH cohort, thus limiting the statistical power of this portion of the analysis. In this smaller subset of 37 patients, the follow-up monocyte count on day 10 (±2 days) of hospital admission was also not associated with a significant increase in mortality at 30 or 60 days. However, the association between the day-10 monocyte count and mortality did exceed the threshold of statistical significance at 6 months and 1 year, but the 95% confidence intervals for the odds ratio were wide and approached 1. In addition, the availability of the day-10 monocyte count indicates that these patients were hospitalized for at least 10 (±2 days) days and were still receiving regular blood counts. As such, these 37 patients likely represent a more severely ill group and the association of AMC with increased odds of mortality may not generalize to all patients with AH. As such, the results of our analysis of monocyte count and prognosis warrant further study, preferably in a prospective manner.

The findings of elevated AMC in patients with AH and the association of AMC with disease severity are important in that they provide evidence that circulating monocytes play a role in the pathogenesis of AH. In the context of previous studies demonstrating hepatic infiltration of circulating monocytes in animal models of AH, the findings presented here suggest that peripheral monocytes play a clinically important role in the course of AH and should be further studied in human populations to provide insight into the pathogenesis and explore possible monocyte-directed therapies for AH.

Acknowledgments

This work was supported by grants AA02605 from the National Institutes of Alcohol Abuse and Alcoholism. JV was supported by TL1 grant TR002368-02 through a CTSA grant from National Center for Advancing Translational Sciences awarded to the University of Kansas Clinical and Translational Science Institute. The research data repository utilized is funded by CTSA Award UL1TR002366. ZL is supported by Grant 81974458 from the National Natural Science Foundation of China and Grants 2019RS1042, 2019TP1035, and 2018RS3072 from the China Hunan Provincial Science and Technology Department.

References

1 Li Z, Weinman SA. Regulation of hepatic inflammation via macrophage cell death. *Semin. Liver Dis.* 2018; **38**: 340–50.
2 Li Z, Zhao J, Zhang S, Weinman SA. FOXO3-dependent apoptosis limits alcohol-induced liver inflammation by promoting infiltrating macrophage differentiation. *Cell Death Discov.* 2018; **4**: 1–14.
3 Thursz MR, Forrest EH, Ryder S, STOPAH investigators. Prednisolone or pentoxifylline for alcoholic hepatitis. *N. Engl. J. Med.* 2015; **373**: 282–3.

4 Morabito V, Novelli S, Poli L *et al*. Adacolumn granulocyte-apheresis for alcoholic hepatitis: preliminary study. *Transplant. Proc.* 2016; **48**: 352–8.
5 Watanabe K, Uchida Y, Sugawara K *et al*. Sequential therapy consisting of glucocorticoid infusions followed by granulocyte-monocyte absorptive apheresis in patients with severe alcoholic hepatitis. *J. Gastroenterol.* 2017; **52**: 830–7.
6 Mckeever U, O'Mahoney C, Lawlor E, Kinsella A, Weir D, Feighery C. Monocytosis: a feature of alcoholic liver disease. *Lancet.* 1983; **322**: 1492.
7 Vergis N, Khamri W, Beale K *et al*. Defective monocyte oxidative burst predicts infection in alcoholic hepatitis and is associated with reduced expression of NADPH oxidase. *Gut*. 2017; **66**: 519–29.
8 Crabb DW, Bataller R, Chalasani NP *et al*. Standard definitions and common data elements for clinical trials in patients with alcoholic hepatitis: recommendation from the NIAAA Alcoholic Hepatitis Consortia. *Gastroenterology*. 2016; **150**: 785–90.
9 Waitman LR, Warren JJ, Manos EL, Connolly DW. Expressing observations from electronic medical record flowsheets in an i2b2 based clinical data repository to support research and quality improvement. *AMIA Annu. Symp. Proc*. 2011; **2011**: 1454–63.
10 Murphy SN, Weber G, Mendis M *et al*. Serving the enterprise and beyond with informatics for integrating biology and the bedside (i2b2). *J Am Med Inform Assoc*. 2010; **17**: 124–30.