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Effect of hepatitis C infection on anemia in hemodialysis patients

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Abstract
Hepatitis C (HCV) infection is commonly seen in dialysis patients, but its long-term deleterious effects in these patients are unknown. We evaluated the effect of HCV infection on anemia in our hemodialysis population. This retrospective case control study was carried out from January 1999 to February 2007. The HCV positive patients were assessed for a 12-month period by quarterly lab results for the prevalence of anemia, iron stores, dialysis adequacy, and alanine aminotranferase levels. Their requirements of erythropoietin (EPO) and intravenous (IV) iron were assessed during these months of clinical stability. A control group of age-matched, race-matched, and gender-matched hemodialysis patients with no history of HCV was similarly assessed for anemia, iron stores, and EPO and IV-iron requirements. Twenty-two HCV-positive patients were included for comparison analysis with 44 control patients for 1:2 matching. The mean EPO requirement for the hepatitis group was 17,307 $\pm$ 14,708 U/month in comparison with the control group, which required 49,134 $\pm$ 49,375 U/month (p value $<$0.01). The mean dose of IV-iron was 120 $\pm$ 143 mg/month for hepatitis patients and 163 $\pm$ 112 mg/month in the control group (p=0.07). The patients with HCV have lower requirement of exogenous EPO replacement compared with their age-matched, gender-matched, and race-matched dialysis counterparts. The IV-iron requirement was not significantly different between the 2 groups but had a suggestive lower trend in the hepatitis group. This needs to be further studied in larger trials.

Key words: Hepatitis C, anemia, EPO, IV-iron

INTRODUCTION

Since its first diagnosis in 1989, hepatitis C (HCV) has been a significant viral infection among hemodialysis patients.[1] The figures on its estimated prevalence have changed as more dialysis centers across the country have adopted routine HCV testing. During 1992 to 1999, national surveillance data indicated that the proportion of dialysis centers that tested patients for anti-HCV increased from 22% to 56% (CDC Web site, unpublished data).[2] Anti-HCV testing has now become routine in most dialysis units across the country. In 1999, nationwide prevalence of anti-HCV was 8.9%, with some centers reporting a prevalence of $>$40% (CDC, unpublished data).[2] Other studies of hemodialysis patients in the United States have reported anti-HCV prevalence of 10% to 36% among adults.[2] As of 2002, the prevalence of HCV in hemodialysis units across the country was estimated at 7.8%.[3] This decline is related mainly to increased awareness, better testing modalities for blood products, and better infection control measures practiced by the dialysis units. The introduction of recombinant erythropoietin (EPO) for management of anemia in the dialysis population minimized the need for frequent blood transfusions, which has also helped in reducing the risk of exposure to HCV. However, there continues to be a significant risk of transmission based on exposure to blood products, needle sticks, and through dialysis

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personnel (if proper precautions are not adhered to). Patients with end-stage renal disease (ESRD) on dialysis have depressed alanine aminotranferase (ALT) levels, making the antibody testing the only reliable modality to diagnose this infection.[4,5] Hepatitis C is a chronic infection with long-term deleterious effects on the dialysis patients that have often been undiagnosed because of the high mortality and shortened life span of these patients.

Sahin et al. have previously studied hemodialysis patients with HCV and suggested a lower EPO and iron requirement in these patients.[6] We have also observed a few patients in our dialysis units with HCV who had minimal or no requirement for EPO. This study was undertaken to assess the effect of HCV infection on anemia in the hemodialysis population in our outpatient unit.

MATERIALS AND METHODS

All hemodialysis patients in our institution whose records were available from January 1999 to February 2007 were reviewed for HCV serologies. For the purpose of the study, patients were labeled as HCV-positive if they had positive anti-HCV antibodies on 2 separate occasions or on 1 occasion along with a confirmatory HCV polymerase chain reaction testing or evidence of cirrhosis on liver imaging and/or biopsy. Patients with borderline positive tests or with a history of HCV with negative serologic tests were excluded from the study. Other exclusion criteria included polycystic kidney disease, cryoglobulinemia, hepatitis B, active malignancy, hematopoetic disorders (including multiple myeloma), chronic infections (including osteomyelitis), active gastrointestinal bleeding, active treatment with interferon and/or ribavirin, and the presence of mass lesions or multiple cysts on renal ultrasound.

The patients had to be stable on hemodialysis for at least 6 months with a urea reduction ratio (URR) $>65\%$ and demonstrate compliance with fewer than 3 missed dialysis sessions per month. Quarterly patient lab data (including hemoglobin, iron, total iron binding capacity, ferritin, ALT) and EPO and intravenous (IV)-iron dosage were obtained from retrospective chart review for a 12-month period. This period was selected for each dialysis patient where they had no hospitalizations, major surgeries, episodes of GI bleeding, access clotting or bacteremia, or any other infections in a 4-week period before the blood draws. Parenteral iron supplementation consisted of either InFeD® (iron dextran) or Ferrlecit (sodium ferric gluconate) as IV-iron formulations and were considered as dose equivalent, milligram for milligram. Erythropoietin was usually administered through the subcutaneous route in our dialysis unit. For patients who received it through the IV route, a conversion factor of 0.68 was applied to adjust for equivalent subcutaneous dose.[7]

An age-matched, gender-matched, and race-matched control group of hemodialysis patients was selected with 2 control patients for each hepatitis patient in the study. If matched control patients were not available, the patient was then excluded from the analysis. The above lab and medication parameters were recorded in a similar fashion in the control patients except for ALT levels. The same exclusion criteria were also applied in selecting the control group.

The EPO doses were reported as average monthly doses and also as average monthly dose per kilogram of body weight, with the assumption that all patients in the study and control groups received all of their dialysis sessions each month. The body weight used was the reported dry weight on dialysis at the beginning of the 12-month period of lab draws. Intravenous-iron dose was also reported as the average monthly dose. Average dose for EPO and IV-iron were reported as mean $\pm$ standard deviation. Serum ferritin levels were averaged for all the four lab values. Transferrin saturation (TSAT) $>20\%$ and ferritin levels >100 ng/mL were taken as an indication of adequate iron stores in accordance with Kidney Disease Outcomes Quality Initiative (KDOQI) guidelines. Urea reduction ratio was used as a marker for dialysis adequacy in patients to account for differences in dialysis prescriptions in these patients. Average URR was used to compare dialysis adequacy among the 2 groups.

For statistical analysis, Kruskal-Wallis test was used to compare EPO, IV-iron doses and ferritin levels, and URR of patients in hepatitis and the control groups. Fisher's exact test was used to compare group characteristics as a whole in terms of EPO freedom (no use of EPO throughout the study period) and EPO "free" months (months where no EPO was administered). A p value of <0.05 was considered to be statistically significant. Correlation coefficient R was used to analyze any correlation between ALT levels and the use of EPO, IV-iron, and ferritin levels.

RESULTS

Forty-two patients had a confirmed diagnosis of HCV. Of these, 22 patients were included for analysis based on the exclusion criteria and the availability of lab results for the 12-month period with the criteria outlined above (Table 1). The patient demographics are outlined in Table 2. There was a distinct African American preponderance in the group with 16 patients. Four Caucasians and 2 His-

Table 1 List of patients with hepatitis C and their exclusion

Hepatitis C patients	n=42
Hepatitis C patients included in the study	22
Coexisting autosomal dominant polycystic kidney disease	1
Mass lesions on ultrasound	2
Lack of appropriate controls	2
Lack of appropriate lab results	13
Noncompliance with dialysis sessions	2

Table 2 Demographic data

Characteristic	Hepatitis C group (n=22)	Control group (n=44)
Age		
<50	7 (32%)	10 (23%)
50–59	9 (41%)	24 (54%)
60–69	6 (27%)	10 (23%)
Gender		
Male	16 (73%)	32 (73%)
Female	6 (27%)	12 (27%)
Race		
Caucasian	4 (18%)	8 (18%)
African American	16 (73%)	32 (73%)
Hispanic	2 (10%)	4 (10%)

panic patients completed the group. Sixteen patients were male and, hence, comprised about two-thirds of the patient group. The control group had similar age, gender, and race distribution as mentioned in the selection criteria above. Age varied from 39 to 66 years (mean 53 years) in the hepatitis group and 35 to 67 years (mean 53 years) in the control group. The patients in the hepatitis group had an average body weight of 73 ± 15 kg, while the average body weight in the control group was 88 ± 24 kg.

The results are outlined in Table 3. Three patients in the hepatitis group did not require any EPO at any point of time in the study period compared with none in the control group (p=0.03). Based on the hemoglobin results, hepatitis patients did not require EPO in 18 out of 88 quarters in comparison with 7 out of 176 quarters in the control group (p value <0.0001).

The mean EPO dose for the patients in the hepatitis group was 17,307 ± 14,708 U/month (range 0–194,500 U/month) in comparison with the control group which required 49,134 ± 49,375 U/month (range 6900–867,200 U/month; p=0.001). In terms of EPO dose per body weight, the hepatitis group had an average dose of 253 ± 218 U/kg/month (range 0–720.37 U/kg/month) in comparison with control group dose of 560 ± 519 U/kg/month (range 17.25–2466.34 U/kg/month; p=0.02) (Figure 1).

The mean dose of IV-iron was 120 ± 143 mg/month for hepatitis patients and 163 ± 112 mg/month in the control group. There was suggestion of lower iron requirements in the hepatitis group; however, it was not statistically significant (p=0.07) (Figure 2).

The average serum ferritin levels were higher in the control group; 727 ± 272 ng/mL in comparison with 632 ± 260 ng/mL in the hepatitis group. However, this was not statistically significant (p=0.4) (Figure 3). The patients in the control group were able to achieve ferritin levels >100 ng/mL on a more consistent basis; 174 out of 176 lab draws compared with 83 out of 87 lab draws in the hepatitis group.

Patients in both the groups were able to achieve adequate TSAT>20% on most lab results. Out of 87 lab values in the hepatitis group, TSAT >20% was seen in 80 instances. In the control group, TSAT >20% was seen in 162 out of 176 lab draws (p=1).

The adequacy of dialysis was assessed in all patients using the URR. All quarterly URR values were averaged

Table 3 EPO and iron dose and ferritin levels for the hepatitis and the control groups

Parameter	Hepatitis C	Control	p value
Patients not requiring EPO	3/22	0/44	0.03[a]
Months without EPO use	18/88	7/176	<0.001[a]
EPO dose (U/month)	17,307 ± 14,708	49,134 ± 49,375	0.001[b]
EPO dose (U/kg/month)	253 ± 218	560 ± 519	0.02[b]
IV-iron (mg/month)	120 ± 143	163 ± 112	0.07[b]
Ferritin (ng/mL)	632 ± 260	727 ± 272	0.42[b]
URR (%)	76 ± 3.5	74.5 ± 5	0.76[b]

[a]Fisher's exact test was used.
[b]Kruskal-Wallis test was used.
EPO=erythropoietin; IV=intravenous; URR=urea reduction ratio.

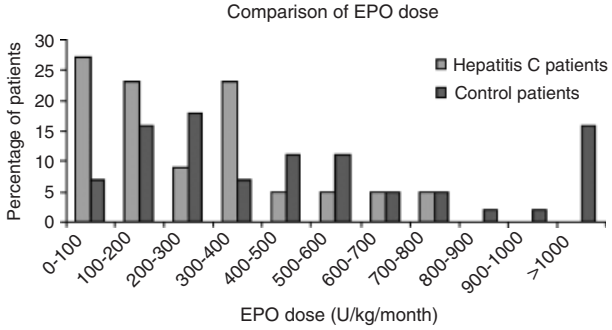

Figure 1 Comparison of erythropoietin dose between the hepatitis C and the control groups.

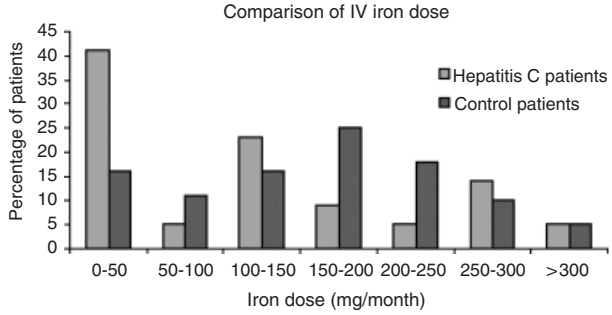

Figure 3 Comparison of serum ferritin levels between the hepatitis C and the control groups.

out for each patient and then compared among the two groups. The average URR for the hepatitis group was 76 ± 3.5%, while the average URR for the control population was 74.5 ± 5% (p value = 0.7).

The average ALT level in the hepatitis group was 29 ± 16 U/L. In terms of patients that required no EPO replacement (n = 3) compared with those who required exogenous replacement in the hepatitis group (n = 20), no significant difference was seen in the ALT levels among the patients. There was no statistically significant correlation between average ALT levels and the EPO requirement, IV-iron requirement, or ferritin levels in the hepatitis group.

DISCUSSION

Patients with ESRD on dialytic support are usually anemic due to lack of EPO secretion from the kidney. A few exceptions include polycystic kidney disease where kidneys are able to maintain EPO secretion despite loss of other functions. In our study, HCV has emerged as another potential cause of EPO freedom in some dialysis patients as evidenced by 3 patients who had no need for exogenous

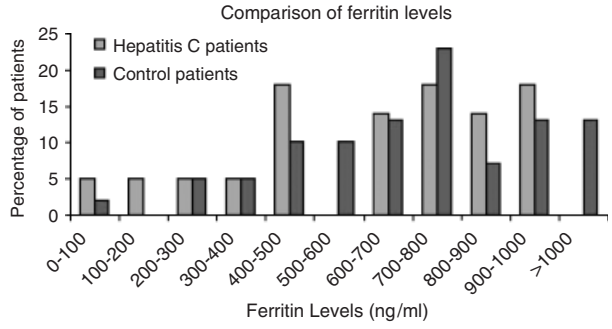

Figure 2 Comparison of intravenous-iron dose between the hepatitis C and the control groups.

EPO replacement throughout the study period compared with none in the matched control group. Also as a group, these patients required less EPO therapy in comparison with their age-matched, race-matched, and gender-matched non-HCV patients on dialysis during the period of clinical stability with adequate dialysis.

Red cell production in the body requires the presence of EPO and adequate iron stores. Erythropoietin is the major hormone required for red cell precursor proliferation in the bone marrow. It stimulates the blast forming units-erythroid in the bone marrow to mature into proerythroblasts, which later form the circulating red cells. Its major site of production is the peritubular fibroblasts in the kidney. Some degree of endogenous EPO production also comes from the liver.[8] This has been found to be significant especially in the fetus and in nephrectomized animals in previous studies. The exact site of EPO production in the liver is not clear. Some studies have pointed it out to be located in the Kupffer cells[9] while others believe it to be within hepatocytes surrounding the central veins, along with contribution from the Ito cells in the space of Disse.[10] In either case, hepatonecrosis from inflammation from hepatitis or a regenerating liver postinjury could potentially result in release of EPO from hepatocytes into the circulation.

The other major requirement for erythropoiesis is adequate iron stores. This is assessed mainly by calculating serum TSAT (serum iron/serum total iron binding capacity × 100) and measuring circulating ferritin levels (because bone marrow biopsy to estimate iron stores is rarely performed). After dialysis therapy is initiated, almost all patients require periodic IV-iron supplementation to keep iron saturation in the range of 20% to 50% and ferritin levels between 100 and 800 ng/mL based on KDOQI guidelines. We obtained our data at least 6 months after dialysis begun to ensure clinical stability of these patients, to allow resolution of the deleterious

effects of uremia on hematopoietic function, and to confirm achievement of adequate iron stores.

Patients with HCV tend to have higher ferritin compared with non-HCV patients, as ferritin is an acute phase reactant that is released from the liver with hepatic inflammation.[11,12] Although this may be the case in the nondialysis population, the infusion of IV-iron on dialysis elevates the ferritin levels, even in non-HCV patients. Exogenous iron replacement has been known to cause release of free iron and oxyradical formation, which can damage cellular lipids and nucleic acids[13] and, thus, less iron requirement may be beneficial. Sahin et al. had found a lower requirement of IV-iron in dialysis patients with HCV.[6] Our study did not show a lower iron requirement in these patients although there was a suggestive trend. This lack of difference may be due to the small number of patients in our study or may be related to the diverse patient population consisting of mainly African American patients and, thus, merits further evaluation.

The exogenous EPO requirement of patients with HCV was overall significantly lower even when adjusted for body weight. We hypothesize that the chronic inflammation as a result of HCV infection or the increased production from the regenerating liver cells causes increased circulating EPO—causing improved hematocrit in these patients. Quantifying the role of hepatic inflammation, however, is a difficult task as the commonly used markers like ALT are unreliable in dialysis patients.

Patients on dialysis in general tend to have lower ALT levels.[12,14] Hepatitis C, unlike hepatitis B infection, has a greater tendency toward intermittent exacerbations and remissions, with a very variable and fluctuating ALT profile. Thus patients with HCV on dialysis may have normal ALT levels despite significant histological liver damage.[15] It is estimated that 20% to 25% of HCV patients with normal ALT levels will have histological damage on liver biopsy.[16] Because ALT elevation is not a reliable feature of dialysis patients with HCV, it cannot be used as a surrogate marker to quantify inflammation in the liver. There have been reports of using the AST/ALT ratio as a predictor of hepatic inflammation both in the nondialysis and dialysis population with HCV; however, this warrants further study.[17,18]

Simple testing for EPO levels to confirm increased hepatic EPO production would be inaccurate in most patients unless they were not receiving any exogenous EPO replacement. In patients receiving exogenous replacement, measurement of their endogenous levels would not be possible because current EPO assays do not differentiate endogenous from exogenous EPO.[19] Normal EPO levels vary from 15 to 19 mIU/mL. However, in response to anemia or hypoxia, these levels can increase 100-fold on an exponential curve. Whether the hepatic production of EPO responds to anemia and hypoxia in a similar fashion to renal EPO production is not known. An invasive way of proving our hypothesis would be to quantify EPO mRNA levels in the hepatocytes obtained on liver biopsy from our patients and/or measuring serum EPO levels in blood sampled from the hepatic veins during periods of clinical stability on dialysis, which could be a daunting task in itself.

As in the previous study done by Sahin et al.[6] we confirmed the need for lower EPO requirement in our subset of patients with HCV. In pure financial terms, this may appear to translate to cost savings on EPO at the dialysis center; however, the overall health care expenditure from HCV may far outweigh this benefit.

Freedom from EPO or very low requirements usually prompts investigation for acquired renal cystic disease, renal cell carcinoma, or polycythemia vera in dialysis patients. We suggest that it should be a reason to look for occult HCV infection, given the higher risk of acquiring this infection on hemodialysis and having normal ALT levels.

CONCLUSION

Hepatitis C patients tend to have higher baseline hemoglobin and decreased need for EPO therapy on dialysis. The possible explanation for these findings may be the release of hepatic EPO because of chronic hepatic inflammation secondary to HCV. This needs to be further studied.

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