Decreased microRNA-122 Levels with HCV Clearance in HIV-HCV Co-Infection

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ABSTRACT

Background and Aims: Micro RNA-122 (miR-122) is under investigation as a target for direct antiviral agents against the hepatitis C virus (HCV), and as a biomarker for both cancer and acute liver injury. Previous data suggest HCV mono-infection is associated with increased serum miR-122 levels. This study sought to determine outcomes in regard to miR-122 levels following clearance of HCV in human immunodeficiency virus (HIV) co-infected patients.

Methods: Nine HCV-HIV co-infected patients undergoing antiviral therapy were treated with interferon and ribavirin for 48 weeks between January 2009 and March 2011, and had serial miR-122 levels measured in triplicate from serum with mirVana™ PARIS™ kit according to the instructions from the manufacturer (Ambion, AM1556). Values were measured at baseline, 1 week, 4 weeks, end of treatment (EOT; 48 weeks), and at either 12 or 24 weeks after treatment completion (SVR). SAS V9.3 was used to analyze these data. Change from baseline (copies/µL) was calculated as Log10 (Baseline)-Log 10(time), where time was 1 week, 4weeks, EOT, and SVR; a repeated measures ANOVA was used to compare the results over time for the patients. If the ANOVA was found significant, post hoc, pairwise comparisons were used to examine change from baseline across the four time points.

Results: Six of nine achieved SVR, 1 was undetectable at EOT but relapsed, and 2 patients were non-responders. Among the 6 patients achieving SVR, all showed a decrease in miR-122 levels between 0.16 and 1.46 logs, between baseline and SVR. The ANOVA confirmed a significant decrease in miR-122 levels from 1 week to SVR24 (p=0.0225). Significant pairwise comparisons for change from baseline were found at 1 week versus SVR (p=0.0063), 4 weeks versus SVR (p=0.0086), and EOT versus SVR (p=0.0458).

Conclusion: Clearance of chronic HCV is associated with decreased miR-122 levels in HIV co-infected patients and was not improved in patients with continued infection who failed to respond to treatment.

INTRODUCTION

Micro RNA-122 (miR-122) is under investigation as a target for direct antiviral agents against the hepatitis C virus (HCV) [1], and as biomarkers for cancer and acute liver injury [2-6]. miRs are highly conserved, small, single stranded RNA molecules, of either human or viral origin [7]. miR expression can be signaled by external stimuli, leading to cell response, and function to inhibit translation of protein products [2]. They are responsible for cell functions as diverse as proliferation, apoptosis, development, differentiation, metabolism and immunity.

Evidence suggests that the presence of miR-122 is necessary for HCV replication [7, 8]. Though no association between HCV viral load and miR-122 levels have been observed [9], lower miR-122 levels at baseline may be predictive of non-response because of decreased cellular response to interferon (IFN) and HCV infection is associated with increased serum miR-122 levels [8].

MATERIALS AND METHODS

Patients: Nine HIV/HCV co-infected patients attending clinic at UTSW who planned to undergo double therapy with interferon and ribavirin were enrolled in this study. Six patients achieved a sustained viral response (SVR; measured at 12 or 24 weeks after treatment; 66%), two patients did not respond to treatment (22%), and one patient relapsed after treatment was completed (11%). Serial miR-122 values were taken at baseline, week 1, week 4, week 48 (end of treatment [EOT]), and SVR.

Data extraction: The miR-122 was extracted from serum with mirVana™ PARIS™ Kit, and levels were measured with a Taqman MicroRNA assay. Values were measured in triplicate with the median value used in the analysis. Values are presented in Table 1.

Statistics: Change from baseline (copies/µL) was calculated as Log10 (Baseline)-Log 10(time), where time was 1 week, 4weeks, EOT, and SVR; a repeated measures ANOVA was used to compare the results over time for the patients. If the ANOVA was found significant, post hoc, pairwise comparisons were used to examine change from baseline across the four time points. SAS V9.3 was used to analyze these data.

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RESULTS

Table 1: Serial miR-122 Values Across Treatment Timepoints

Patient	Baseline	Week 1	Week 4	EOT	SVR
A	19,888.11	7,684.84	8,877.93	4,290.44	7,949.37
B	2,971.24	6,218.71	26,170.32	3,306.59	412.56
C	43,108.46	9,778.02	10,755.26	54,833.16	2810.1
D	4,348.36	17,330.23	4,128.28	2,114.11	3,002.93
E	23,073.12	8,691.02	5,578.08	5,430.45	1,886.39
F	12,191.61	9,389.43	6,941.15	(Not Done)	425.71
Median	16,039.86	9,040.23	7,909.54	4,290.44	2,348.25

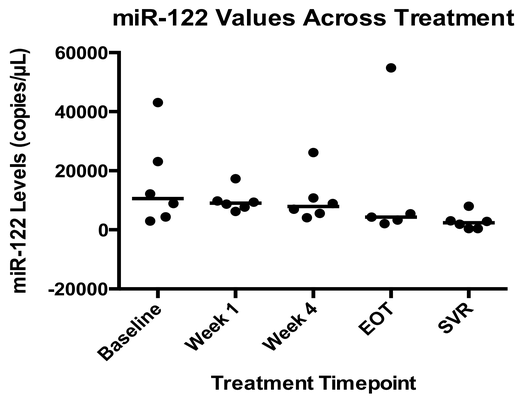
Median values are presented for all 6 SVR patients. Outlier seen at EOT for patient C. This outlier patient had a mild AST/ALT elevation when miR-122 value was measured. Patient F did not have miR-122 values taken at EOT.

RESULTS

- In the sustained viral response (SVR) group (Figure 1), baseline miR-122 levels ranged from 2,971 to 43,108 copies/µL (Average: 18,678, Median: 16,040).
- Median miR-122 values decreased at all time points measured. All subjects had lower miR-122 values at SVR as compared to baseline, with a Log10 decrease ranging from 0.16 to 1.46.
- The ANOVA confirmed a significant decrease in miR-122 levels from 1 week to SVR (p=0.0225).
- Significant pairwise comparisons for change from baseline were found at 1 week versus SVR (p=0.0063), 4 weeks versus SVR (p=0.0086), and EOT versus SVR (p=0.0458).
- No decrease was observed in patients not achieving a SVR.
- We identified one outlier at the EOT. This patient had a corresponding AST and ALT elevation.

RESULTS

Figure 1: Serial miR-122 Values Across Treatment Timepoints



Median values are represented as a line at each timepoint. Outlier seen at EOT from patient C. This outlier patient had a mild AST/ALT elevation when miR-122 value was measured. SVR patients only..

SUMMARY/CONCLUSIONS

- Clearance of HCV is associated with decreased values of miR-122 in HIV co-infected subjects.
- These data provide further evidence that hepatitis C virus infected hepatocytes have augmented levels of miR-122, that resolve with viral clearance.

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