Sa1449

DDW Digestive Disease Week®

June 2, 2018

Inflammation Biomarkers Decrease During ELAD Treatment In Alcoholic Hepatitis Subjects

Lee K. Landeen[1], Jason Lapetoda[1], Samantha C. Anderson[1], Zheng Li[1], William O. Frank[1], Ali Al-Khafaji[2], Amay Parikh[3], Andres Duarte-Rojo[4], Geoff McCaughan[5], Lance Stein[6], Michael Allison[7], Nikunj Shah[8], Ram Subramanian[9], Raza Malik[10], Robert Brown[11], Ross MacNicholas[12], Sumeet Asrani[13], Talal Adhami[14], David Reich[15], Julie Thompson[16], Kristina Chacko[17], Brian Borg[18], Lewis Teperman[19], Tarek Hassanein[20], and Patricia W. Bedard[1]

[1]Vital Therapies, Inc., San Diego, CA; [2]University of Pittsburgh Medical Center, Pittsburgh, PA; [3]Rutgers University, Newark, NJ; [4]University of Arkansas for Medical Sciences, Little Rock, AR; [5]Royal Prince Alfred Hospital, Sydney; [6]Piedmont Atlanta Hospital, Atlanta, GA; [7]Cambridge University Hospital Addenbrookes, Cambridge; [8]Rush University Medical Center, Chicago, IL; [9]Emory University, Atlanta, GA; [10]Beth Israel Deaconess Medical Center, Boston, MA; [11]Columbia University Medical Center, New York, NY; [12]Sir Charles Gairdner Hospital, Perth; [13]Baylor University Medical Center, Dallas, TX; [14]Cleveland Clinic Foundation, Cleveland, OH; [15]Drexel University, Philadelphia, PA; [16]University of Minnesota Medical Center, Minneapolis, MN; [17]Montefiore Medical Center, Bronx, NY; [18]University of Mississippi Medical Center, Jackson, MS; [19]North Shore University Hospital, Manhasset, NY; [20]Southern California Research Center, Coronado, CA

VITAL THERAPIES™ TARGETING LIVER DISEASE

INTRODUCTION

Chronic alcohol consumption increases gut translocation of lipo-polysaccharide (LPS) into the portal vein and may initiate systemic inflammatory response syndrome (SIRS), a major predictor of multi-organ failure (MOF) and mortality in severe alcoholic hepatitis (sAH) patients. Therapies that reduce inflammation (e.g. interleukin-1 receptor antagonist [IL-1Ra]) should provide clinical benefit. VTL C3A cells respond to pro-inflammatory cytokine stimulation by increasing IL-1Ra secretion.[1]

AIM

This study reports IL-1Ra, procalcitonin (PCT) and ferritin (FRTN) plasma levels in sAH subjects undergoing ELAD treatment or standard of care (Control) in the VTI-208 Phase 3 trial, and relates them to the presence of SIRS, antibiotic use, and mortality up to Day 91.

MATERIALS & METHODS

Vital Therapies' (VTL) ELAD® System. The ELAD System is an investigational human hepatoma cell-based liver treatment comprised of four metabolically active ELAD cartridges with ancillary device components and support circuitry intended to continuously treat subjects with sAH for up to 5 days.

sAH Subjects' Demographics in Subsets A. Sixty subjects (30 Control, 30 ELAD) enrolled in the VTI-208 clinical trial (NCT01471028) met the following subset criteria: <50 years, ≤2.5 INR, <1.3 mg/dL creatinine (CR), ≥16 mg/dL total bilirubin [tBili], and <30 MELD score.

Clinical Study. Performed in accordance with 21 CFR Parts 50, 54, 56 and 312 Subpart D; ICH E6; the ethical principles in the Declaration of Helsinki; and/or the EU Directive 2001/20/EC; Directive 2005/28; and CPMP/ICH/135/95 with annotation by the TGA, Australia.

Sample Collection for Subset B. Twenty-five (11 Control, 14 ELAD) of the sixty VTI-208 subjects consented to blood draws and had samples collected at screening (baseline), during the 5-day ELAD treatment, and at post-treatment follow-up on days 14 and/or 28. Venous whole blood was drawn into Li-heparin vacutainer tubes, centrifuged within 30 minutes, and the separated plasma was stored at -70°C until testing.

Immunoassays. Concentrations of IL-1Ra, PCT and FRTN were determined in previously frozen plasma samples using ProteinSimple Ella custom SimplePlex assays (Bio-Techne, Minneapolis, MN USA).

Statistical Analysis. Immunoassay data and tBili were longitudinally analyzed by ANOVA. Demographics and screening criteria were analyzed using Student's t test or Fisher's exact test, with p ≤0.05 being deemed statistically significant.

RESULTS

Selection Criteria for the Subsets of VTI-208 Subjects Evaluated

The subsets of the subjects enrolled in the VTI-208 trial were selected based on the criteria for the current VTL-308 trial (NCT02612428). Subjects were younger, had lower INR, lower CR, higher tBili, and lower MELD than the total population (Table 1).

	Age (Years)		CR (mg/dL)	tBILI (mg/dL)	MELD
Available	≥18	≤3.5	NA	≥8	18-35
Subsets A and B	**<50**	**≤2.5**	**<1.3**	**≥16**	**<30**

Table 1. Selection Criteria for the Subsets of VTI-208 Subjects Evaluated. The subsets of subjects selected for these analyses conformed to the population selected for the current VTL-308 trial.

Demographics of Subjects in Subset A

There were no differences in age, gender, MELD score, number of organ failures as measured by ACLF score[2], or SIRS (per the American College of Chest Physicians [ACCP]) at screening between ELAD and Control subjects in subset A (Table 2).

Group (ELAD/Control	Subjects (No./Group)	Age (Avg±SEM)	Gender (M/F)	MELD (Avg±SEM)	No ACLF (1 Organ Failure) (No./Group)	ACLF Grade 2 (No./Group)	SIRS=Yes (No./Group)
(A) ELAD	30	40 ± 0.9	13/17	25 ± 0.4	28	2	13
Control	30	41 ± 1.1	16/14	26 ± 0.4	28	2	9
P value	N/A	0.305	0.606	0.184	1.00		0.366
(B) ELAD	14	39 ± 2	4/10	25 ± 0.6	13	1	6
Control	11	39 ± 1	7/4	26 ± 0.5	10	1	3
P value	N/A	0.90	0.12	0.677	1.00		0.68

Table 2. ELAD and Control Subjects in this Subset A were not Significantly Different for Demographics at Screen. There were no statistically significant differences in age, gender, MELD, ACLF criteria[2], or SIRS at screen (see Methods section for specific statistical analyses used).

SIRS and Survival

The SIRS criteria used were per the ACCP. While ELAD subjects were more likely to have SIRS during treatment, subjects in the ELAD group with SIRS at screening had increased overall survival up to 91 days compared with Control (100% vs 56%, p=0.017 [Table 3]).

Group (ELAD/Control)	SIRS=Yes (No./Group)	91 d Survival (No./Total) (% of Total)
ELAD	13/22 (59%)	13/13 (100%)
Control	9/22 (41%)	5/9 (56%)
P value	0.366	0.017

Table 3. SIRS and Survival by Treatment Group. The ELAD group with SIRS at screen had increased overall survival up to 91 days compared with the Control group with SIRS at screen (100% vs 56%, p=0.017).

Increase in Anti-Inflammatory Biomarker IL-1Ra in Subset B

Subset B consisted of twenty-five of the sixty subjects in Table 2 who consented to have blood drawn for later testing. IL-1Ra levels in ELAD subjects in this group were significantly increased vs Control subjects and vs screen levels during study days 3, 5, 7 (i.e. ELAD treatment period), but not at days 14 or 28 (Fig 1).

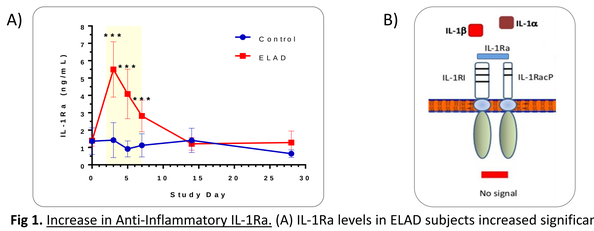

Fig 1. Increase in Anti-Inflammatory IL-1Ra. (A) IL-1Ra levels in ELAD subjects increased significantly vs. screen on days 3, 5, and 7 (i.e. during ELAD treatment period, indicated by the yellow shading; p<0.0001, D3; p<0.0001, D5; and p=0.001, D7). IL-1Ra levels in ELAD subjects were also significantly higher than Control levels during study days 3, 5, and 7 (***p<0.0001, D3; p<0.0002, D5; p=0.004, D7). (B) IL-1Ra is known to exert anti-inflammatory responses by blocking IL-1β signaling.

Decrease in Inflammatory Biomarkers PCT and FRTN in Subset B

FRTN levels were significantly decreased from screen levels in ELAD subjects on study days 5, 7, 14, and 28. PCT levels were significantly decreased from screen levels in ELAD subjects on study days 5, 7, and 28. Neither factor changed significantly in Control subjects. PCT levels were also significantly lower in ELAD subjects vs Control subjects on study day 7 (Fig 2). Antibiotic use was similar between ELAD and Control subjects at all time points.

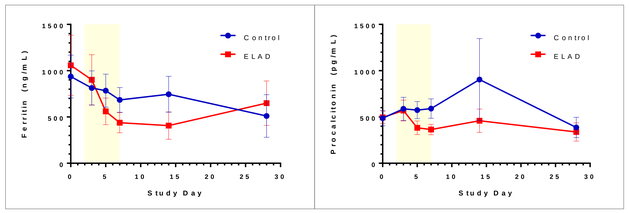

Fig 2. Decreased Inflammatory Biomarkers. (A) Ferritin levels decreased significantly in ELAD-treated subjects during ELAD treatment and thereafter (p=0.0025 ELAD D5 vs. Screen; and p<0.0001 ELAD D7, 14 and 28 vs. Screen). (B) Procalcitonin levels were significantly lower than control p=0.049 at D7, and decreased significantly from screen p=0.034 at D5 , p=0.025 at D7, and p=0.0003 at D28.

Proposed Model of Anti-Inflammatory Effects of ELAD Treatment

IL-1β and IL-6 from sAH subjects should induce an increase in IL-1Ra production by VTL C3A cells. IL-1Ra produced by VTL C3A cells and delivered to AH subjects during ELAD treatment, should block the proinflammatory effects of IL-1β in the sAH subjects, lowering biomarkers of inflammation, PCT and FRTN (Fig 3).

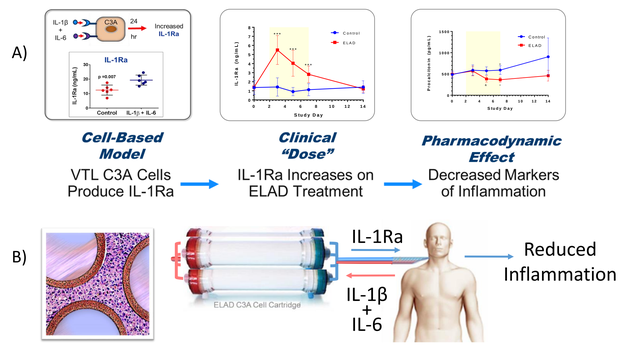

Fig 3. Model of Anti-Inflammatory Effects of ELAD Treatment. (A) Cell-based models have demonstrated that IL-1β and IL-6 in combination increase the production of IL-1Ra by VTL C3A cells.[1] An increase in IL-1Ra was measured in VTI-208 subjects during treatment, suggesting a dose of the protein. Pro-inflammatory proteins were seen to decrease on ELAD treatment. (B) These data suggest a model in which ELAD C3A cells respond to IL-1 and IL-6 from sAH subjects by delivering IL-1Ra to the subjects during ELAD treatment. The resulting increase in anti-inflammatory IL-1Ra could thereby block IL-1β signaling and thereby reducing inflammation.

DISCUSSION

Increased understanding of the mechanisms underlying sAH has shown inflammation to be a key driver of disease progression and associated high mortality.[3] Alcohol consumption leads to increased gut permeability and translocation of bacterial endotoxin into the portal circulation. The resulting increase in liver cell death leads to activation of damage-associated pathogen receptors, such as Toll-like receptors, on immune cells in the liver. These immune cells respond with increased secretion of IL-1β and other pro-inflammatory cytokines. Left unchecked, this increasing escalation of inflammation and cell death often leads to systemic inflammation in the form of SIRS, as defined by the ACCP.[4]

SIRS at hospital admission, with or without infection, leads to MOF and is associated with increased mortality in acute liver failure.[5]

Despite this increased risk of progression to MOF and poorer prognosis, ELAD-treated subjects with SIRS at screen in the VTI-208 trial subset had increased overall survival up to 91 days compared with Control subjects.

To evaluate a potential relationship between increased survival up to 91 days and an anti-inflammatory effect of proteins produced by the VTL C3A cells during ELAD treatment, IL-1Ra, FRTN, and PCT were measured in plasma collected from twenty-five subjects (n=14 ELAD subjects, n=11 Control subjects). Significantly higher levels of IL-1Ra were measured in the ELAD subjects during treatment compared with Control subjects. IL-1Ra is known to block the inflammatory effects of IL-1β in many human inflammatory diseases,[6] in murine models of alcoholic steatohepatitis,[7] and is currently being evaluated in a Phase 2/3 clinical trial for AH (NCT01809132).

Both FRTN and PCT are recognized biomarkers of systemic inflammation. PCT is purported to be an early and more sensitive marker of sepsis than C-reactive protein, and is associated with infection and poorer prognosis.[8] Serum FRTN is an independent factor predicting increased 180-day and 1-year transplant waiting list mortality.[9] Both PCT and FRTN plasma levels were significantly reduced in ELAD subjects during treatment, and were lower than Control subject levels. Incidence of antibiotic use in both groups were not statistically different.

The decrease in systemic markers of inflammation and increased survival in the ELAD-treated group relative to the Control group suggests that VTL C3A cell-released products, such as IL-1Ra, may be dampening inflammation and improving clinical outcomes.

The subset of subjects selected for these analyses was younger with less severe disease progression with respect to coagulopathies and kidney function than the original VTI-208 population. However, this is consistent with the evolution in current clinical thinking towards earlier intervention – i.e. intervention before disease progresses too far and during what one prospective study has termed "the golden window".[10]

CONCLUSIONS

Although these findings have yet to be generated in the current clinical trial VTL-308, they strongly suggest that ELAD plus standard of care may be more effective than standard of care alone in treating sAH subjects with SIRS meeting the Subset A criteria. Subjects in the ELAD group with SIRS at screen had increased overall survival up to 91 days compared with Control subjects, despite similar incidence of baseline infection and antibiotic use in both groups.

REFERENCES

1. Landeen LK, Van Allen J, et al. International Liver Transplantation Society. 2015.
2. Moreau R, Jalan R, Gines P, et al. Gastroenterology 2013.
3. Chirapongsathorn S, Kamath PS, Shah V. Hepatology 2015;62:671-673.
4. ACCP & SCCM. American College of Chest Physicians. Crit Care Med 1992;20:864-874.
5. Rolando N, Williams R. et al. . Hepatology 2000;32:734-739.
6. Dinarello CA, van der Meer JW. Semin Immunol 2013;25:469-484.
7. Petrasek J, Bala S, Barrieau M, et al. J Clin Invest 2012;122:3476-3489.
8. Arkader R, Troster EJ, Okay TS, et al.. Arch Disease in Child 2006;91:117-120.
9. Walker NM, Fletcher LM, et al. Hepatology 2010;51:1683-1691.
10. Choudhury A., APASL ACLF working party. J Gastroenterol Hepatol. 2017;32(12):1989-19
11. https://www.slideshare.net/systemicjia/immune-mechanisms-of-inflamation-in-sjia

CONTACT INFORMATION
Patricia W Bedard, Vital Therapies, Inc., 15010 Ave. of Science, Suite 200, San Diego, CA 92128. pbedard@vitaltherapies.com