

# Biomarker Assessment For Neurotrauma Diagnosis and Improved Triage System (BANDITS): Interim Analysis of Pilot Study of Biomarkers of Mild and Moderate TBI



Kevin K Wang, PhD<sup>1</sup>; Stefania Mondello, MD, PhD<sup>1,2</sup>; Larry Lewis, MD<sup>3</sup>; Gillian Robinson, PhD<sup>4</sup>; Mo Jixiang, PhD<sup>1</sup>; Uwe Muller, PhD<sup>1</sup>; Linda Papa, MD<sup>5</sup>; Frank Tortella, PhD<sup>6</sup>; and Ronald L. Hayes, PhD<sup>1</sup>  
<sup>1</sup>Banyan Biomarkers Inc., <sup>2</sup>University of Florida, Anesthesia, <sup>3</sup>Washington University, St. Louis, <sup>4</sup>Neuren Pharmaceutical, <sup>5</sup>Orlando Regional Medical Center, <sup>6</sup>Walter Reed Army Institute of Research

## Abstract

Biomarker Assessment for Neurotrauma Diagnosis and Improved Triage System (BANDITS) seeks to develop blood-based biomarkers useful for diagnosis, management and prognosis of traumatic brain injury (TBI) experienced by military personnel and ultimately useful in far-forward combat environments. Researchers estimate that more than 300,000 U.S. veterans of wars in Iraq and Afghanistan (20% of the 1.6M) have sustained a mild TBI, also known as concussion, with the majority going untreated (Hogue et al, NJM, 2009). 34 mild and moderate TBI patients were recruited from Orlando Regional Medical Center (ORMC) and Washington University, St Louis (Wash U). Injury magnitude was inferred by the Glasgow Coma Scale (GCS), 29 patients had a mild TBI (GCS 13-15) and 5 had a moderate TBI (GCS 9-12). Blood samples were taken at enrollment (T-E: mean = 3 hrs 35 min) as well as 6 hours after injury. Inclusion criteria captured patients aged 18 or older with an initial GCS of 9-15 and requiring a CT scan as part of their clinical evaluation. The control group consisted of uninjured healthy volunteers aged 18 or older without severe pre-existing chronic conditions or a previous history of TBI or stroke. Blood samples were stored at -70°C and shipped to Banyan Biomarkers, Inc. for analysis. UCH-L1 was analyzed employing a sandwich ELISA developed by Banyan. GFAP was analyzed using a commercial assay (BioVendor USA). Separate pilot studies conducted at 2 sites have indicated that serum levels of UCH-L1 and GFAP are sensitive and specific predictors of mild TBI when assayed approximately 3-6 hrs after injury. Although limited by a small subject population, data suggests that higher levels of UCH-L1 and GFAP are potentially associated with the presence of lesions detectable on CT.

## Introduction

Biomarker Assessment for Neurotrauma Diagnosis and Improved Triage System (BANDITS) seeks to develop blood-based biomarkers useful for diagnosis, management and prognosis of traumatic brain injury (TBI) experienced by military personnel and ultimately useful in far-forward combat environments. Researchers estimate that more than 300,000 U.S. veterans of wars in Iraq and Afghanistan (20% of the 1.6M) have sustained a mild TBI, also known as concussion, with the majority going untreated (Hogue et al, NJM, 2009). Mild and moderate TBI is difficult to diagnose. Current methods to assess severity of TBI are based on CT scans and neurological examinations. These measures are not very sensitive or reliable and most are not available on the battlefield. There are no rapid, definitive and cost-effective blood-based diagnostic tests for TBI. This study measured levels of ubiquitin C-terminal hydrolase-L1 (UCH-L1) and GFAP in blood samples from MMTBI patients. UCH-L1 is present in almost all neurons and averages 1-5% of total soluble brain protein. GFAP is an intermediate filament protein found in astrocytes within the CNS. Although highly enriched in CNS, GFAP has also been located in rat peripheral organs including kidney and testes. Previous studies in our own and other laboratories, however, have suggested potential utility for UCH-L1 and GFAP as biomarkers for acute brain injury.

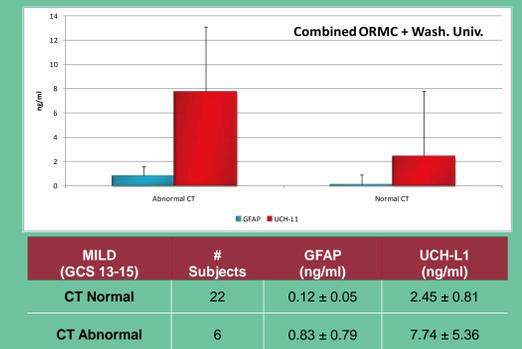
## Methods

A total of 34 mild and moderate TBI patients were recruited from Orlando Regional Medical Center (ORMC) and Washington University, St Louis (Wash U). Injury magnitude was inferred by the Glasgow Coma Scale (GCS), 29 patients had a mild TBI (GCS 13-15) and 5 had a moderate TBI (GCS 9-12). Blood samples were taken at enrollment (T-E: mean = 3 hrs 35 min) as well as 6 hours after injury (T-6). Inclusion criteria captured patients aged 18 or older with an initial GCS of 9-15 and requiring a CT scan as part of their clinical evaluation. Patients also manifested either loss of consciousness or loss of memory for events immediately after the accident. Patients with a previous history of TBI or stroke or with known dementia or psychotic illnesses were excluded. The control group consisted of uninjured healthy volunteers aged 18 or older without severe pre-existing chronic conditions or a previous history of TBI or stroke.

Blood samples were stored at -70°C and shipped to Banyan Biomarkers, Inc. for analysis. UCH-L1 was analyzed employing a sandwich ELISA developed by Banyan. GFAP was analyzed using a commercial assay (BioVendor USA).

Mild and Moderate TBI Population		Control Groups	
ORMC & Wash Univ Combined		Normal Controls (Serum)	
n	34	n	167
Age, years Mean (SD)	42.66 ± 17.08	Age, years Mean (SD)	36.91 ± 14.06
Range	19-87	Min-Max	18-74
F/M, n	13/21	F/M, n	72/95
F/M, %	38.23/ 61.77 %	F/M, %	43.1/ 56.9 %
Ethnicity		Ethnicity	
Not Hispanic or Latino	11	Not Hispanic or Latino	146
Hispanic or Latino	2	Hispanic or Latino	13
Other	1	N/A	8
Race		Race	
Caucasian	24	Caucasian	53
Asian		Asian	7
Black or African American	9	Black or African American	4
Other	1	American Middle Eastern	1
GCS		Alcohol (Y/N)	114/47
13-15	29		N/A 6
9-12	5	Drug	19/ 147
Mechanism of Injury:			N/A 1
Motor Vehicle Accident	9	Smoke (Y/N)	54/112
Moto Cycle Accident	4		N/A 1
Fall	8	Health	Excellent 78
Assault	3		Good 74
Other	10		Average 13
CT			N/A 2
Normal	24		
Abnormal	9		

## Biomarkers vs. CT scan in Mild TBI



## Conclusions

- Separate pilot studies conducted at 2 sites have indicated that serum levels of UCH-L1 and GFAP are sensitive and specific predictors of mild TBI when assayed approximately 3-6 hrs after injury.
- Although limited by a small subject population, data suggests that higher levels of UCH-L1 and GFAP are potentially associated with the presence of lesions detectable on CT.

## Future Studies

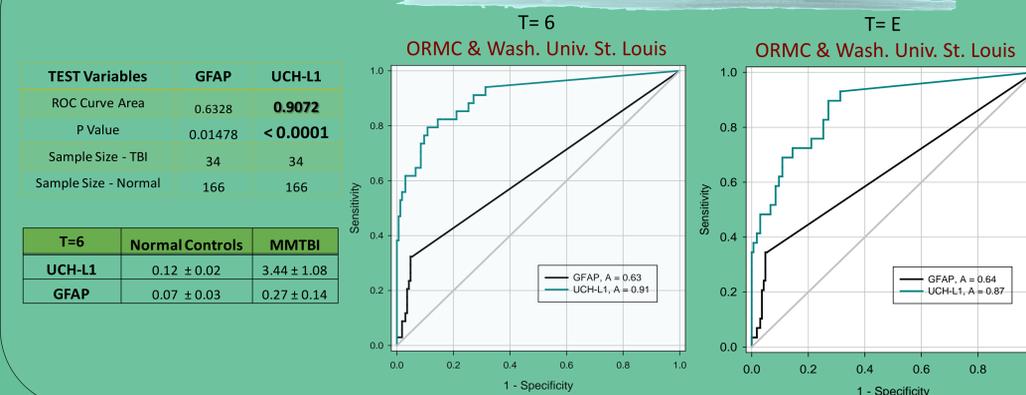
- Implement multicenter feasibility study by Oct 1, 2009 further addressing sensitivity and specificity of biomarkers to detect mild and moderate TBI, biomarker kinetics, relationship of biomarkers to lesions detected on CT and persistent deficits assessed by neuropsychological tests.
- Conduct studies examining relationships between biomarkers and brain pathology in mild TBI patients assessed by magnetic resonance imaging protocols including diffusion tensor and susceptibility weighed imaging.

### For additional information, please contact:

Kevin Wang, PhD  
 Banyan Biomarkers Inc.  
 kwang@banyanbio.com

Ronald L. Hayes, PhD  
 Banyan Biomarkers Inc.  
 rhayes@banyanbio.com

## ROC Curves: Biomarkers in Mild (GCS 13-15) TBI



Variables	GFAP	UCH-L1
ROC Curve Area	0.6428	<b>0.8735</b>
P Value	0.01421	<b>&lt; 0.0001</b>
Sample Size - TBI	29	29
Sample Size - Normal	166	166

T=E	Normal Controls	MMTBI
UCH-L1	0.12 ± 0.02	3.48 ± 1.27
GFAP	0.07 ± 0.03	0.28 ± 0.17

T=E  
**Mean 3h35', Range 15'-14h35'**  
 ORMC: Mean 1h43', Range 15'-4h  
 Wash.Univ.: Mean 4h59', Range 1h20'-14h35'

## UCH-L1: Sensitivity and Specificity

