



**TRACK-TBI**  
Precision Medicine  
Transforming Research and Clinical Knowledge  
in Traumatic Brain Injury Precision Medicine

**Appendix 1: TRACK-TBI Precision Medicine Option 1 Phase II  
Neuroimaging Manual of Procedures (MOP)**

**March 10, 2021**

**Version 1.1**

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## 1. Study Sites and Study Site IDs

- 03 UCSF
- 07 Pittsburgh
- 12 Pennsylvania
- 14 MCW
- 15 Utah

## 2. TRACK-TBI Precision Medicine Option 1 Phase II SPECIFIC AIM 2

**To validate early and ultra-early blood-based and novel imaging biomarkers of DAI, MVI, and neuroinflammation that may serve as predictive and pharmacodynamic biomarkers in a new cohort of moderate-severe subjects.**

In the TRACK-TBI U01 study, MRIs were first obtained at 2-weeks after injury, and the initial blood sample was obtained within 24 hours after injury. Since it is likely that many effective TBI therapies will have to be started within hours or days post-injury, to obtain the necessary precision to design future clinical trials, we must have more granular information about the early evolution of the imaging and molecular biomarkers identified in Phase 1. In Aim 2 we will use the TRACK-TBI clinical recruitment procedures (see section 5-TRACK-TBI Precision Medicine Clinical Protocol) to recruit a new cohort of subjects (n=50) with moderate to severe TBI defined according to VA/DoD criteria<sup>1</sup> through four of the existing TRACK-TBI Network sites. In this cohort, we will validate blood-based biomarkers at early (<24h) and ultra-early time periods (~6h), and neuroimaging biomarkers at early (2w) and ultra-early (within 48h) time periods.

### I) Specific Aim 2.A.

**Validate early and ultra-early predictive blood biomarkers as well as pharmacodynamic molecular biomarkers in the acute phase after injury.**

### II) Specific Aim 2.B.

**Validate early and ultra-early novel imaging biomarkers in the acute phase after injury.**

In this cohort we will obtain MRIs within 2 week of injury (early), and for a subset of 20% of patients, an additional ultra-early MRI within 48 hours of injury. All patients will also receive a follow-up MRI at 3m postinjury. In addition to volumetrics, DTI and rs-fMRI, the new MRI protocol will incorporate novel imaging measures of axonal density, cerebral blood flow, and neuroinflammation, which will all be standardized across sites and MRI vendors prior to participant enrollment.

***Hypothesis 2.B-1:*** MR imaging biomarkers of DAI at ultra-early (within 48h) and early (2w) time points will correlate with elevated plasma biomarkers of acute DAI and predict progressive atrophy (on 3D T1 structural MRI), white matter degeneration and loss of functional connectivity in the period from 1 week to 3 months post-injury.

***Hypothesis 2.B-2:*** MR imaging biomarkers of MVI at ultra-early (within 48h) and early (2w) time points will correlate with elevated plasma biomarkers of acute MVI and predict progressive atrophy (using volumetric analysis of the serial 3D T1-weighted MRI scans) from 1 week to 3 months post-injury.

***Hypothesis 2.B-3:*** We further hypothesize that acutely elevated plasma biomarkers of MVI will correlate with reduced CBF and brain parenchymal microhemorrhages at <24 hours and at 1 week post-injury, both reflecting vascular endothelial dysfunction, and predict global and regional brain volume loss from 1 week to 3-months post-injury on volumetric analysis of 3D T1-weighted

*imaging.*

**Hypothesis 2.B-4:** *MR imaging biomarkers of neuroinflammation at ultra-early (within 48h) and early (2w) time points will correlate with elevated plasma neuroinflammatory biomarkers and predict progressive atrophy from 1 week to 3 months post-injury.*

### **3. Research Approach to Aims 2.A and 2.B.**

**Neuroimaging biomarkers:** The overall MRI protocol is based on established TRACK-TBI standards for structural imaging, DTI and rs-fMRI that have been harmonized across 3T scanners from all 3 MR vendor platforms. Standard operating procedures for acquisition, QA, QC, and data management of this 3T MRI protocol are available from the TRACK-TBI Imaging Core and have been adopted by other multicenter TBI imaging studies. The proposed MRI protocol includes one additional sequence not represented in the TRACKTBI protocol described in Aim 1: ASL perfusion imaging of CBF, as well as an additional 64-direction diffusion-weighted shell for the DTI protocol at  $b=3000$  s/mm<sup>2</sup> to create the multishell diffusion MRI sequence

**The entire proposed MRI protocol can be acquired in 60 minutes.** Standardization of structural MRI, diffusion MRI and rs-fMRI across sites will be performed using the same procedures already established for the TRACK-TBI study (See TRACK-TBI Neuroimaging MOP V3\_TRACK-TBI\_MRI\_Manual\_27JULY2018).

Arterial Spin Labeled Perfusion MRI Protocol: The ASL perfusion protocol is adopted from the new Alzheimer Disease Neuroimaging Initiative 3 (ADNI3) standards for all 3 MR vendors. In brief, the 5-minute sequence consists of 3D pseudocontinuous ASL (PCASL) on 3T GE scanners and 2D pulsed ASL (PASL) on Siemens and Philips scanners, with an additional proton density reference scan using the same ASL readout at a longer TR. These ASL acquisitions for all 3 MR vendor platforms conform to the most recent best practice guidelines for ASL perfusion imaging reported by the International Society for Magnetic Resonance in Medicine.<sup>2</sup>

MR Imaging Analysis: Analytic methods for volumetrics (to detect progressive atrophy over serial scans), DTI, and rs-fMRI will be the same as described for Phase 1-Aim 1A of TRACK-TBI Precision Medicine. Presence and number of microhemorrhages on high-resolution 3D T2\* susceptibility imaging will follow the methods described for TRACK-TBI Precision Medicine Phase 1-Aim 1.B. Free water content (FISO) measurements from NODDI of multishell diffusion MR imaging will also follow the methods of Phase1-Aim 1.C.

ASL Perfusion MRI Analysis: CBF maps will be calculated from the difference between the labeled and control ASL perfusion images using the method of Buxton et al. (1998) for 3D PCASL imaging<sup>3</sup> and the method of Wong et al. (1998) for 2D PASL imaging.<sup>4</sup> For correction of proton density (PD) effects and radiofrequency coil inhomogeneities, a scaling factor from the separately acquired PD scan will be employed to yield absolute CBF quantitation in units of mL/min/100 mL.<sup>2</sup>

#### **3.1. TRACK-TBI Precision Medicine MRI procedures**

The TRACK-TBI Precision Medicine MRI procedures such as standardization of MR across sites, integration and analysis will align with the TRACK-TBI U01 procedures; however, the de-identified neuroimages from each site will be uploaded to the neuroimaging core repository at UCSF. TRACK-TBI Precision Medicine participants will undergo the same MRI procedures as set forth in the TRACK U01

study, Exhibit 5 with the exception of one additional ASL perfusion sequence not represented in the TRACKTBI protocol (See the ‘Research Approach’ above).

### 3.2. MRI Time Points

CT or initial MRI will be obtained as part of clinical care. 3T MRI will be obtained within 24-48h, at 2 weeks and 3 months from CA+MRI subjects only. All initial and follow-up brain CT scans, and any brain MRI scans that are done for clinical care and their reports will be collected. Images will be read and coded by the Neuroimaging Core radiologist in accordance to the Neuroimaging TBI-CDEs.

The CA + MRI Cohort will receive a 3T MRI according to this protocol at the following time points:

#### MRI Scans Schedule

<i>Schedule of MRI scan (from time of traumatic brain injury)</i>	<b>24 hr</b>	<b>Day 2</b>	<b>Day 14</b>	<b>Day 90</b>
MRI scan	(X)	(X)	X	X

(X)—Option either day 1 or day 2

- **The first ultra early scan- within 48 hours from time of injury**
- **2-Week/14 days (+/- 4 days) post-injury**  
 Subsequent cognitive testing must be completed within 3 days of this MRI.
- **3-Month/90 days (+/- 14 days) post-injury**  
 Subsequent cognitive testing must be completed within 3 days of this MRI.

### 3.3. MRI Sequences

TRACK-TBI Precision Medicine MRI scan sequences at Ultra Early (<48 hours), 2 Weeks and 3-month time points

	MRI scan sequences Names and Order	Purpose	Approximate Time to complete
1	Sagittal 3D T2 FLAIR	To detect focal lesions	4 to 5 min
2	Axial 3D T2* (SWI/SWAN)	To detect microbleeds	3 min
3	Axial DTI b=1300	To evaluate white matter microstructure and structural connectivity	11 min
4	Axial DTI b=3000	To evaluate white matter microstructure and structural connectivity	11 min
5	Sagittal 3D T1 MPRAGE/IR-SPGR	To detect focal lesions & for volumetrics/morphometry	4 to 5 min
6	Axial resting state BOLD fMRI	To evaluate functional connectivity	10 min
7	Arterial spin labeling	Exploratory	4 min
8	Sagittal 3D T2	For volumetrics/morphometry	4 to 5 min

### **3.4. Transmission to Flywheel**

The clinical study coordinator (or delegate) at each clinical study site will upload the DICOM images from the brain MRI exams to Flywheel used for the TRACK Precision Medicine study, as described in other workflow procedures.

For each MRI imaging session, all images acquired that contain any portion of the head will be transmitted. All reconstructed images that contain any portion of the head will be transmitted, including series in different planes and employing different reconstruction kernels. When multiple acquisitions of the head have been obtained due to patient motion or other artifact, all images will be transmitted.

#### **3.4.1. Coding of Subject Exams**

Flywheel will remove subject names from images during the upload process. Images will be identified by the Patient Number assigned to each subject during study enrollment. The naming convention for the collected images in this study will be assigned as follows: each site will start with subject "PM-XX-3001" for the Patient Number where PM = Study Name, XX = Site ID (see section 1 above), and the last four digits will start at "3001" and increase consecutively with each enrollment at the site. MRI scans will have an additional label at the end to distinguish the time point and whether the scan is a patient or phantom. The additional labels are as follows: (PM-XX-XXXX\_Ultra-early for MRI within 48 hours, \_2WK for 2-week MRI, \_3MO for 3-month MRI, and \_PHA for phantom).

#### **3.4.2. Technical parameters in the DICOM header**

Technical information regarding the scanner (manufacturer, model, etc.) and protocol (e.g. TE, TR, spatial resolution, slice thickness range, etc.) for each MRI exam will be retained in the DICOM header and will thus be transmitted with the images to Flywheel.

### **3.5. Image Storage and Backup**

#### **3.5.1. Site Archiving**

All study specific head MRI exams will be archived locally at the study sites according to standard site procedure.

#### **3.5.2. Flywheel**

All study images will be stored in Flywheel. Flywheel system maintains an audit trail of system access and system activities and is backed up regularly.

### **3.6. Controlled Access to Flywheel**

#### **3.6.1. Flywheel Accounts**

The Flywheel is a web-based system. To access Flywheel, users will be provided with account information (i.e. user ID and password combination). This is unique to the individual and is used as an electronic signature in compliance with 21 CFR part 11.

## APPENDIX A DATA UPLOAD TO FLYWHEEL

The Flywheel website will have the most updated instructions. For upload instructions, please visit: <https://docs.flywheel.io/hc/en-us/articles/360008109094>.

**NOTE:** Please only use the Chrome browser to upload images to Flywheel.

## Image requirements

Your DICOM images must meet the following requirements to use the web uploader:

- Your images are the DICOM filetype. To upload other image types, [see our Importing Overview article](#).
- DICOMs are not compressed into a zip file. The DICOM uploader also does not accept folders. You must upload single files.

### **Tip: Select multiple files from your computer**

To select a range of files, click on a file in your browser, hold down the **Shift** key, and select the last file. All files in between are selected and can be uploaded to Flywheel.

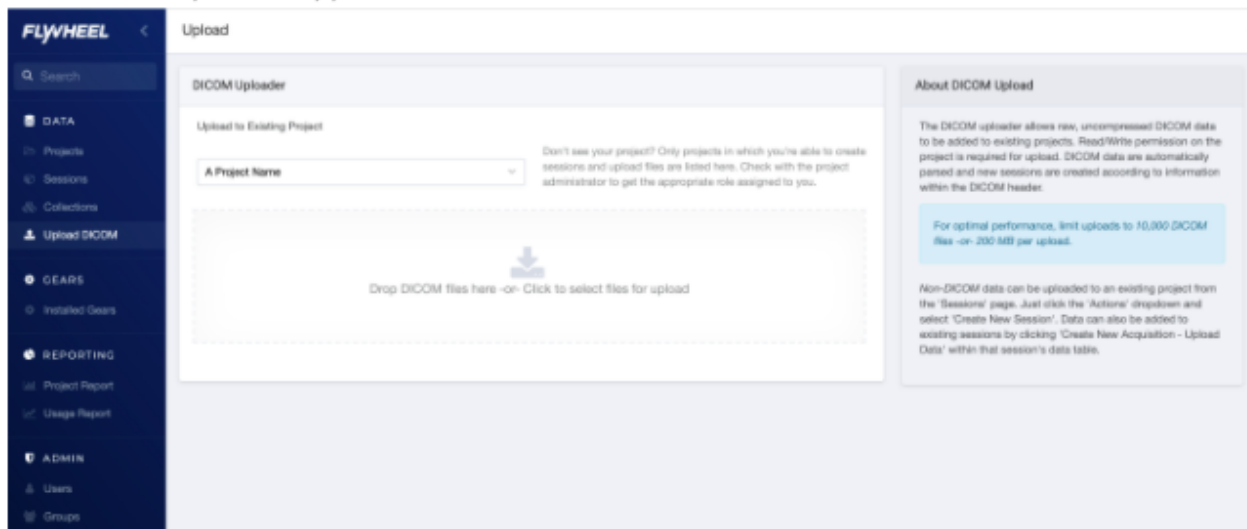
- The total upload size is less than **10,000 DICOM files** or **200 MB**.  
*This is due to various browser limitations.*
- Data does not require custom de-identification. The DICOM Uploader can perform basic de-identification by removing the **PatientID**, **PatientName**, and **PatientBirthDate** headers from the DICOM file before upload. [Learn more about other ways to customize what Flywheel de-identifies](#).

## Steps

After you have verified that your data meets the above requirements, upload your files:

1. Sign in to Flywheel as a user with permissions to upload files to a Project.
2. Click **Upload DICOM** in the left menu.

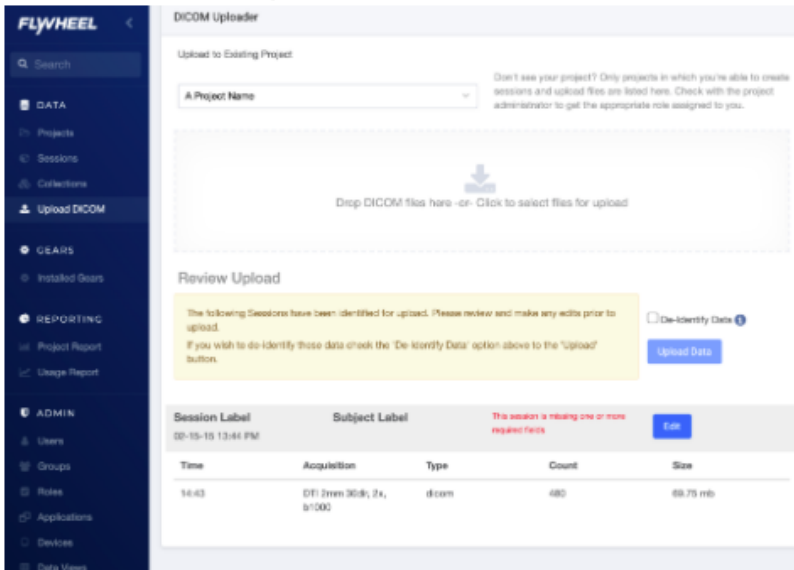
*The DICOM Uploader appears.*



3. Select a Project from the dropdown menu.  
*Only Projects you have permission to upload files to are included in this list.*
4. Drag and drop the files you are uploading or click anywhere on the uploader to launch the file selector.  
*The uploader immediately begins parsing and organizing your Files into Sessions. Properly uploaded files are displayed and grouped by Acquisition. Non-DICOM files and other files not loaded correctly display an error message. .*

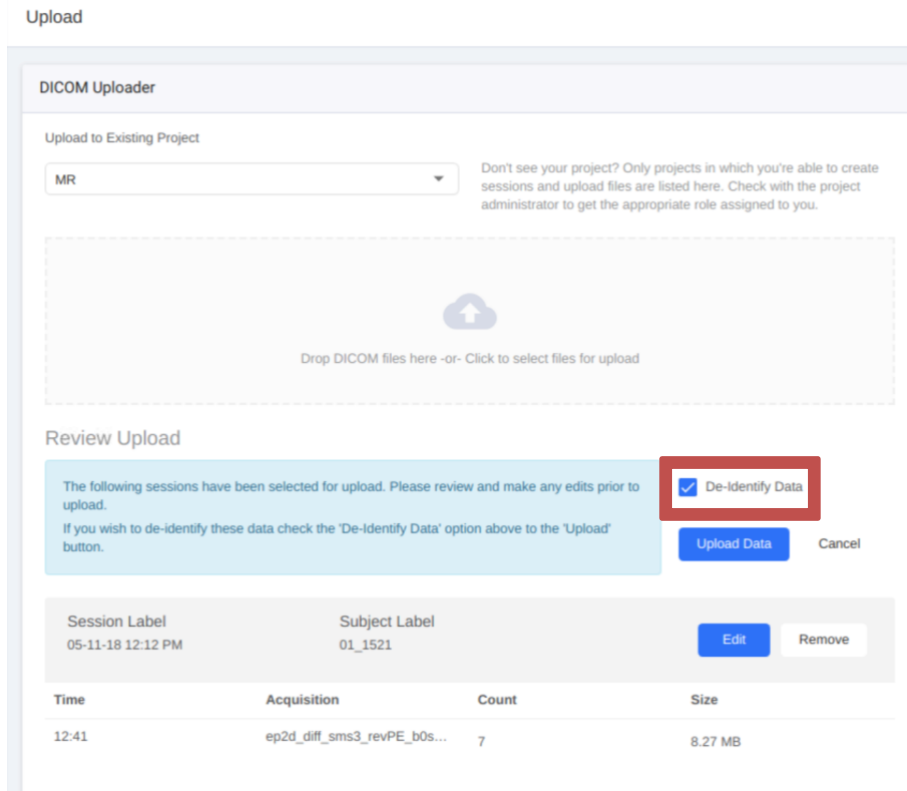


5. Review the upload summary information and make sure that labels are correct. If they are not correct, click **Edit** and update the fields.



6. Enable de-identify data to remove the PatientID, PatientName, and PatientBirthDate headers from the DICOM file. This means that those fields are not imported when you upload the image. This also converts the Age to months.

**NOTE: Be sure to check the de-identify data box**



## References

1. O'Neil, M.E., Carlson, K., Storzbach, D., Brenner, L., Freeman, M., Quinones, A., Motu'apuaka, M., Ensley, M. and Kansagara, D. (2013). VA Evidence-based Synthesis Program Reports. In: *Complications of Mild Traumatic Brain Injury in Veterans and Military Personnel: A Systematic Review*. Department of Veterans Affairs (US): Washington (DC).
2. Alsop, D.C., Detre, J.A., Golay, X., Gunther, M., Hendrikse, J., Hernandez-Garcia, L., Lu, H., MacIntosh, B.J., Parkes, L.M., Smits, M., van Osch, M.J., Wang, D.J., Wong, E.C. and Zaharchuk, G. (2015). Recommended implementation of arterial spin-labeled perfusion MRI for clinical applications: A consensus of the ISMRM perfusion study group and the European consortium for ASL in dementia. *Magnetic resonance in medicine* 73, 102-116.
3. Buxton, R.B., Frank, L.R., Wong, E.C., Siewert, B., Warach, S. and Edelman, R.R. (1998). A general kinetic model for quantitative perfusion imaging with arterial spin labeling. *Magnetic resonance in medicine* 40, 383-396.
4. Wong, E.C., Buxton, R.B. and Frank, L.R. (1998). Quantitative imaging of perfusion using a single subtraction (QUIPSS and QUIPSS II). *Magnetic resonance in medicine* 39, 702-708.