Re: Recommended MRI protocol for Day-2 and Day-30 stroked mice

To all SPAN sites:

As the MRI "shakedown" run approaches, the MRI group, coordinating center, and central repository have continued to revise the imaging protocol and our plans for analysis. The overall goal is to enable a fully automated pipeline for stroke lesion segmentation, brain hemispheric volumes, and midline shift. Because we will be imaging so many animals, it is important that each site employ methods that are best suited to the hardware and sequences at that site, subject to very specific restrictions. Below, we outline the basic imaging protocol, an explicit example protocol specific to a Bruker scanner (used by 5 of 6 sites), and some best practices and potential pitfalls. Methods will be evaluated at each site during the "shakedown run", and then protocols will be tweaked as necessary.

## Basic Imaging protocol

We will collect 4 types of scans at each of the two time points:

1. Anatomical T2-weighted imaging using FSE/TSE/RARE method (fast spin echo)
    - **Purpose:** normalization/segmentation and comparison with later time point at 30 days
    - Use a long TR (e.g., 6000) and an effective echo time of 60 ms or longer
2. A series of spin-echo images in order to create T2 maps
    - **Purpose:** lesion segmentation
    - Use a minimum echo time of 15 ms or shorter to provide a low-contrast volume for analysis
    - Use a maximum echo time of 70 ms or longer (lesion T2 will be about 70 ms)
    - Suggested echo times for multi-echo imaging using a volume transmitter: 0 to 100 ms in steps of 10 ms
    - Suggested echo times for single-echo imaging using a surface coil transmitter: 15, 45, 75 ms
3. A series of diffusion-weighted images in order to create ADC maps
    - **Purpose:** lesion segmentation and CSF discrimination
    - Suggested b-values: 0, 500, 1000 along the z direction
4. A series of gradient-echo images in order to create T2* maps
    - **Purpose:** detection of hemorrhage
    - Suggested echo times: 5 to 30 ms in steps of 5 ms

Additionally, we collect one more scan at the 30 day time point to provide anatomical T1 contrast:

5. MDEFT/MPRAGE. T1 contrast can be obtained using a 2D turbo-FLASH sequence with an inversion preparation pulse. Use a TI of about 1100 ms, a short TE (3-4 ms), an short interpulse separation in the turboFLASH series (12 ms) and a low flip angle (< 10 deg) to obtain an acquisition time/per slice of about 1.5 seconds. Use a repetition time per slice of about 5 sec to obtain 30 slices in 2.5 minutes, and then use averages to boost signal as needed (e.g., 2 averages will yield an image volume in 5 minutes).

All scans except for #1 above should use "conventional" imaging with one readout per excitation in order to avoid distortions. Furthermore, all image data above should use the same matrix, resolution, and geometry:

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| Field of view | 19.2 mm in-plane x 15 mm in slice direction |
| Matrix | 128 x 128 x 30 slices |
| Resolution | 150 x 150 um with 500 um slice thickness |
| BW | 50k |

Individual sites have some latitude to use methods appropriate to their hardware and sequences. In particular, some sites will want to use multi-echo imaging for the generation of spin-echo T2 maps, whereas other sites may not have an available sequence or may need to use single-echo imaging due to the use of a surface coil for RF transmission and reception. Importantly, once each site defines their protocol, it should remain fixed for the duration of the SPAN study.

## Explicit example protocol on a Bruker scanner (numbering scheme matches basic protocol above)

Hardware: 9.4T magnet, Bruker volume transmit coil, Bruker 4-channel phased array surface receiver coil
Software: PV5.1

The numbering scheme below matches the "Basic Imaging Protocol" above. The time per animal, including all setup, should be less than 1 hour.

0. Setup (these setup scans are not needed in the upload)
    - Sagittal localizer (sequence = FLASH) to position animal accurately along bore
    - Tri-plane localizer with large FOV = 30 mm (sequence =RARE) for adjustments
    - Tri-plane localizer with small FOV = 15 mm (sequence =RARE) for geometry planning
1. Anatomical T2-weighted image volume (3.5 min)
    - Sequence = RARE (rapid acquisition with relaxation enhancement)
    - TR/TE=6000/15, 8 segments, 4 averages, effective TE=60
2. Multi-echo (spin-echo) scan to enable T2 map (6.5 min)
    - Sequence = MSME (multi-slice multi-echo); TR = 4500
    - 10 spin echo times from 10 to 100 ms
      Or
    - Sequence = MSME using 1 echo per scan
    - 3 spin-echo TE values = 15, 45, 75 ms
3. Diffusion-weighted scan to enable ADC map (9.5 min)
    - Sequence = DtiStandard (*), TR/TE=1500/25
    - 3 b-values: 0, 500, 1000
4. Multi-echo (gradient-echo) scan to enable T2* map (2.5 min)
    - Sequence = MGE (multi-gradient-echo), TR=1500
    - 4 gradient echo times from 3 to 15 ms

(*) Note that sequence DtiStandard enforces a minimum inter-slice delay time to prevent high gradient duty cycles during long runs (at least this is true on PV5.1). This delay can lead to excessively long TR values, so it may be necessary to edit the sequence to shorten this delay, which is not necessary for short diffusion scans.

For the T1-weighted scan at 30 days, there will be an additional sequence:

5. Anatomical T1-weighted image volume (5 min)
    - Sequence in MDEFT (modified driven equilibrium Fourier transform)
    - TI=1100, TE=3.5, repetition per slice = 5000, 2 averages

## Potential pitfalls and best practices

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| Pitfall | Some scans are not co-aligned with others within a single dataset. |
| Best practice | After defining the geometry on the first scan (e.g., RARE anatomical), always copy geometry from the first scan to other scans. |
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| Pitfall | Image volumes within a given "mapping" dataset (e.g., multi-echo data) do not have a consistent scale factor. |
| Best practice | 1) When possible, collect all data within a mapping series using a single scan that collects multiple time points (e.g., multiple TE values or b values) to ensure self-consistent scaling<br>2) If hardware warrants the use of multiple scans (e.g., a multiple spin-echo sequence like MSME should not be used with a transmit surface coil), then take special care to ensure that each scan has the correct relative signal (**). |
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| Pitfall | The stroke lesion appears on the wrong side of the brain, complicating analysis. |
| Best practice | Ensure that all animals are registered correctly during the initiation of the scan. Using “feet first” when the animal is “head first) will cause a parity change in the image coordinate system. |
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| Pitfall | A surface coil provides insufficient spatial coverage or signal to noise ratio (SNR) across the whole brain, leading to a failed segmentation. |
| Best practice | While surface coils provide excellent SNR in general, ensure that 1) the surface coil in use is large enough to provide full brain coverage under optimal conditions, and 2) there is reproducible method to accurately position the coil on the animal head. If initial images indicate poor volume coverage due to a shifted coil or animal head, remove the animal and reposition the coil before continuing the scan. |

(**) On Bruker scanners, set the parameter “Reco_map_mode = ABSOLUTE_MAPPING”

## Upload to LONI Repository

MRI Data will be uploaded to LONI in DICOM format. These files will contain much of the information that is needed to analyze the data, but unfortunately some information will be missing. For instance, Bruker DICOM files do not incorporate b-values. Moreover, it would simplify identification of each image series if it was labeled in some manner. To help facilitate, please include the following text strings into your “protocol name”, which is a standard DICOM field that will be carried along with the data.

1. RARE anatomical image volume: protocol name includes “RARE_anatomy”
2. T2-weighted image scan(s): protocol name includes “T2_map”
3. Diffusion-weighted image scan(s): protocol name includes “ADC_map”
4. Gradient-echo scan(s): protocol name includes “T2Star_map”

Additionally, specific information describing items 2-4 above should be copied into a text file to accompany each dataset upload. See an example below for the text file.

T2 information
Sequence = multi-echo multi-slice or single-echo multi-slice
TE = specify 10 values for multi-echo or 3 values for single-echo (in units of ms)

ADC information
Sequence = DtiStandard or whatever was used
b-values = specify 3 values used (in units of s/mm2)

T2* information
Sequence = multi-gradient-echo multi-slice or whatever was used
TE = specify 4 values used (in units of ms)

Thank you – the MRI working group.