**Meeting Minutes**

|  |  |
| --- | --- |
| Meeting: | SPAN Steering Committee Meeting |
| Date and Time: | 10/28/20 11am-12pm PST |
| Present: | Patrick Lyden, Jessica Lamb, Andreia Lopes, Mariia Kumskova, Rakesh Patel, Nirav Dhanesha, Pradip Kamat, Senthil Gounder, M Khan, Tao Qin, Anjali Chauhan, Anil Chauhan Francesca Bosetti, Louise McCullough, Andrew Goh, David Hess, Lee, Jarek Aronowski, Andre Rogatko, Ryan Cabeen, Cenk Ayata, Ali Herman, Lauren Sansing, Basavaraju Sanganahalli, Shuning Huang, Raymond Koehler, Enrique Leira, Marcio Diniz, Karisma Nagarkatti |

**Agenda Items**

1. **Recruitment Update** 
   1. Sites need to enroll an equal number of males and females per surgery day.
   2. Doppler drops after occlusion and mortality rates are where they should be. Neuroscores do not indicate severe strokes.
   3. Suggestion to ask sites to save Laser Doppler readouts. Sites should be saving files in such a way that you can retrieve source data. In the meantime. CC to draft an SOP regarding source verification of original data.
   4. Video Stats: CC is waiting on 501 Assigned videos. CC asks every site PI to meet with team to incentivize teams to turn in scores for assigned videos to answer the question whether 3 raters per video is needed. Since we are now beginning 30-day assessments on top of weekly baseline and day 7 assessments, the volume of work will be growing. Sites are encouraged to stay ahead of the work flow.
2. **Report on RIC from IW** 
   1. Dr. Dhanesha shared his experience with RIC. Initially planned asleep surgeries but realized it would not be possible unless you perform treatment at night. IW liked to switch to the awake surgery because IW spent less amount of time doing the surgery. Surgery planned early in the morning. Last treatment was at 10pm at night. IW turned in post-mortem images for deceased mice. Initially optimizing with mice anesthesia helped to determine lowest amount of Isofluorane.
   2. Questions about RIC: How did you time Day2 MRI and RIC treatment, 2 anesthesia doses that day for the mouse. What order should the network decide on? Might not be possible to standardize depending. Sites were advised to consider how they will handle Day 2 RIC and MRI to avoid multiple anesthetics.
   3. Calibration: Image of Manometer at IW shown. Manometer reads in 26.61 kiloPascals converts to 200mmHg. CC requests that sites once RIC is assembled please photo document the manometer reading.
   4. RIC Rotation: YL->UT->JH->MG
3. **Protocol Deviations –not addressed**
   1. Age of mice—sites are asked to pay careful attention to the ages of the mice and randomize mice that are within the target age range.
4. **M/F Housing**-How are sites separating M/F for surgeries scheduled on the same day? **-not addressed**
5. **Reminder**
   1. Enrollment Forms SOP 4: Enroll the mouse in the REDCap database at least one week before the planned MCAo date
   2. ITT Forms SOP 5: Submit ITT forms at least 48 business hours (preferably 72-96 hours) prior to day of surgery
   3. Video Uploads into IDA: Accurate Labeling
   4. REDCap data entry accuracy
6. **Holiday Planning and Planned surgeries** 
   1. Nov 15-Nov 31st surgeries? 12/15-12/31 MRI 1/15-1/31
   2. CC closed and will not be performing randomizations on the following days: 11/26-11/27; 12/25; 12/28-12/31
7. **Plan for Stage 2** 
   1. Dr. Lyden asked all SPANners to begin thinking about Stage 2 trial design. Stage 1 Recap: young f/m mice with filament occlusion. Stage 2 could include aged animals with filament occlusion, or young animals with comorbidities, eg diabetes or htn.
   2. McCullough Experience with Aged Animals
      1. Dr. McCullough believes they are a good model for SPAN. It is a difficult population to study. They will not survive 90 min. Occlusion; 60 min. occlusion they will survive. With aged mice, Dr. Chauhan tries to limit the duration of anesthesia and try to minimize the surgery duration within 10 min or so. Her lab gives subcutaneous fluids twice a day. It is important to watch the aged males, they do not groom so UTH washes them down otherwise they will get urosepsis. Aged mice are fed wet mash with the chow. McCullough believes it is reasonable to go to co-morbidities first such as obese or diabetic.
      2. Cost: Ordering aged mice is expensive about $ 200-250 per mouse. If you have an NIA grant, they will send you 10 mice a month for free. Never isolate them. NIA willing to go above the limit but would need lead time of 1 year because they have commitments and would need to breed for SPAN.
      3. Age of Aged Animals: McCullough suggests 18-20mths old. Natural mortality is 50% by 26 months. Can tolerate IP injections. Repeated anesthesia is difficult, UTH does not touch them for the first 5-7 days after MCAo. Need to establish a plan then for RIC and 48hr MRI
      4. JH will have aged mice by Fall 2021.
      5. IW: Will variations in aging the animals amplify inter-subject variability in a way that can affect the experiment in a significant away?
         1. NIA animals if you order those they are much leaner than those aged in house at UTH. There are many dietary and environmental factors that affect that. Aged mice have smaller strokes, may be somewhat similar to what is seen clinically.
      6. IW experience with Aged Mice: Mortality of 60-70% in the first weeks. They do worse in behavioral outcomes and difficult to perform surgery. If done awake it might be a different outcome but we are unsure.
   3. Thoughts on some sites doing the thrombolysis model.
      1. Aronowski Lab has performed in rats not done in mice. Inject old clot, in order to formulate the clot you need to push through the syringe a tiny catheter many times. You are leaving exclusively a clot that when you inject depending on how it is morphed (difficult to make it consistent) makes it very difficult. Might be reasonable to make a fresh clot, insert a catheter instead of a filament to the level of MCA you have a little bit of the thrombin, aspirate the clot, you don’t want to inject the thrombin in the vasculature, and push in a fresh clot. It’s a reasonable approach.
      2. IW used to make it the previous day and store at 4C. Delivering the clot very slowly. This was done in mice.
      3. McCullough lab has performed it in aged mice.
   4. Co-morbid SHRs, Zucker mice, DBDB mice
      1. Diabetic Stage 2 or Hypertensive rat stage 2?
      2. MGH: We cannot hope to reproduce stage 1 in stage 2. We would need to tailor the study structure for Stage 2.
      3. Yale: Aging is going to be important. Cost is a concern, perhaps partnering with NIA and planning ahead would have some similarities for the first ½ of their life would be a benefit with the acknowledgement that they are less like humans. In terms of co-morbidities Yale thinks hypertension is critical to model. A pragmatic approach would be to permit sites with the expertise to perform the clot tpa approach.
      4. JH: With the aged mice we will be focused more on behavior outcome than on infarct volume because infarcts are smaller as LM mentioned. Peak sensitivity is at 10-14 days and we will lose sensitivity at 30 days. We may need to shorten the survival period to 10-14 days. Takes 20-30 minutes to complete 10 corner turns in aged mice.
      5. Augusta: Biased to autologous clot model in mice, spike with human fibrinogen, in young mice. Clot model has not been done in 18 mth mice at AG.
   5. Ordering Mice from NIA
      1. MGH suggestion: Ordering 10 aged mice per month per lab. Sites can do other studies in parallel.
      2. Sites agree that CC should proceed with arranging the NIA order for aged mice.
      3. Aronowski believes that Network should consider another species.
   6. Statistics planning for Stage 2: Challenge is that all those models assume that you have a homogenous population. How much will previous information be useful for the future information. Once you have a couple of animals from the new population, we would be learning the standard deviation and variability and then develop the model from there.
8. **NINDS** Thoughts**:** 
   1. Dr. Bosetti: TPA is important to do to make sure there is no interference with any of the agents being tested because this is all being explored in the context of reperfusion. It is important to have a cohort that includes tpa could be in stage 2 or stage 3.

**Work Items**

1. Each site PI to send CC a trail design proposal for Stage 2 and a list of models each site has used in the past
2. Each site PI to meet with their team to ensure scores are sent in for assigned videos and that M/F are being enrolled simultaneously in equal distribution
3. Sites to consider Stage 2
4. CC to start process with NIA to order 10 aged mice per month per lab.
5. CC to query sites about how many sites can do mice and rat surgeries in the same week
6. CC to query sites on how they are housing M/F for surgeries scheduled in the same day

**Next Meeting date: 11/25/20 11am PST**