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| Budget code | 10309 |
| Time estimate | **To be agreed with BABS based upon consultation.** |
| Analysis goals | **Please provide an outline of the goals of the analysis**:  ATAC-seq: peak calling and differential analysis between genotypes and/or timepoints  RNA-seq: differential gene expression analysis between genotypes and/or timepoints  ChIP-seq: peak calling |
| Analysis details | **Please provide details of the data analysis required.**  Three replicates will be provided for each genotype for the ATAC-seq and RNA-seq experiments:   1. Wild-type 24h DAPT neurons – WT R1, WT R2, WT R3 2. 24h DAPT neurons treated with DMSO for 24h – 24h DMSO R1, 24h DMSO R2, 24h DMSO R3 3. 24h DAPT neurons treated with BRM014 inhibitor for 24h – 24h BRM014 R1, 24h BRM014 R2, 24h BRM014 R3 4. 24h DAPT neurons treated with BRM014 inhibitor for 48h – 48h BRM014 R1, 48h BRM014 R2, 48h BRM014 R3 5. 24h DAPT neurons treated with DMSO for 48h – 48h DMSO R1, 48h DMSO R2, 48h DMSO R3   Three replicates will be provided for each genotype for the ChIP-seq experiment:   1. ASCL1 ChIP wild-type 24h DAPT neurons – ASCL1 ChIP WT R1, ASCL1 ChIP WT R2, ASCL1 ChIP WT3 2. ASCL1 ChIP 24h DAPT neurons treated with DMSO for 48h – ASCL1 ChIP 48h DMSO R1, ASCL1 ChIP 48h DMSO R2, ASCL1 ChIP 48h DMSO R3 3. ASCL1 ChIP 24h DAPT neurons treated with BRM014 inhibitor for 48h – ASCL1 ChIP 48h BRM014 R1, ASCL1 ChIP 48h BRM014 R2, ASCL1 ChIP 48h BRM014 R3 4. mSWI/SNF complex ChIP wild-type 24h DAPT neurons – mSWI/SNF ChIP WT R1, mSWI/SNF ChIP WT R2, mSWI/SNF ChIP R3 5. mSWI/SNF complex ChIP ASCL1ko 24h DAPT neurons – mSWI/SNF ChIP ASCL1ko R1, mSWI/SNF ChIP ASCL1ko R2, mSWI/SNF ChIP ASCL1ko R3   Comparisons to be performed for ATAC-seq and RNA-seq (differential analysis):   1. WT vs 24h DMSO 2. WT vs 24h BRM014 3. 24h DMSO vs 24h BRM014 4. WT vs 48h DMSO 5. WT vs 48h BRM014 6. 48h DMSO vs 48h BRM014   For the ATAC-seq data, peak calling should be performed for each replicate. Following peak calling, a peak which was called in at least 2 out of the 3 replicates for each genotype should be put in a final set of peaks, which is then used for the differential analysis.  For the ChIP-seq data, peak calling should be performed for each replicate relative to its corresponding input. Following peak calling, a peak which was called in at least 2 out of the 3 replicates for each genotype should be put in a final set of peaks, which is then used for the differential analysis.  *Based on the preliminary results of these analyses, integration with previously generated data sets might be necessary.*  Integration of the ATAC-seq and ChIP-seq data with the RNA-seq data has been difficult to perform so far because both ASCL1 and the mSWI/SNF complexes bind and regulate chromatin accessibilityat genomic loci that are located far (>10kb) from the nearest annotated promoter. These loci are also enriched for both H3K4me and H3K27ac (normally found at enhancers and distal regulatory elements) and less enriched for H3K4me3 (usually found at promoters). The histone modification data sets used for these comparisons have been taken from ENCODE. In order to be able to associate these distal genomic loci with the RNA-seq data, we would like to use Activity-By-Contact (ABC) algorithm. This algorithm predicts enhancer-promoter interactions based on ATAC-Seq and H3K27ac ChIP-Seq data.  <https://www.nature.com/articles/s41588-019-0538-0?proof=t>  <https://www.sciencedirect.com/science/article/pii/S2001037019303630> |

**By submitting this form, you are confirming that your PI has agreed on the project and that the cost-code can be used for this purpose.**

Please be aware that the number of hours listed to do the analysis is an estimate and numbers may change. If the project takes longer than estimated you will be informed, and a new estimation will require approval before continuing. Likewise, if a project takes less time than the initial estimate you will be charged accordingly.

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