

## Life Sciences Reporting Summary

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For further information on the points included in this form, see [Reporting Life Sciences Research](#). For further information on Nature Research policies, including our [data availability policy](#), see [Authors & Referees](#) and the [Editorial Policy Checklist](#).

## ▶ Experimental design

## 1. Sample size

Describe how sample size was determined.

We collected prefrontal cortices from embryonic stages available under the regulations by Reproductive Study Ethics Committee. To exclude batch effect for analysis, we collected two to three replicates for three stages of samples. Final dataset scale was determined according to the quality control criteria as described in the methods.

## 2. Data exclusions

Describe any data exclusions.

Cells detected with less than 1,000 genes and genes with normalized expression level less than 1 or expressed less than 3 single cells were removed out before initial clustering analysis.

## 3. Replication

Describe whether the experimental findings were reliably reproduced.

All replications were consistent for data results.

## 4. Randomization

Describe how samples/organisms/participants were allocated into experimental groups.

The samples were allocated into each experimental groups based on the gestational stage. See methods 'Tissue sample collection and dissection'.

## 5. Blinding

Describe whether the investigators were blinded to group allocation during data collection and/or analysis.

The investigators were blinded to group allocation during data collection and analysis.

Note: all studies involving animals and/or human research participants must disclose whether blinding and randomization were used.

## 6. Statistical parameters

For all figures and tables that use statistical methods, confirm that the following items are present in relevant figure legends (or in the Methods section if additional space is needed).

- |                          |  |
|--------------------------|--|
| n/a                      | Confirmed  |
| <input type="checkbox"/> | <input checked="" type="checkbox"/> The <u>exact sample size</u> ( $n$ ) for each experimental group/condition, given as a discrete number and unit of measurement (animals, litters, cultures, etc.)                                    |
| <input type="checkbox"/> | <input checked="" type="checkbox"/> A description of how samples were collected, noting whether measurements were taken from distinct samples or whether the same sample was measured repeatedly   |
| <input type="checkbox"/> | <input checked="" type="checkbox"/> A statement indicating how many times each experiment was replicated   |
| <input type="checkbox"/> | <input checked="" type="checkbox"/> The statistical test(s) used and whether they are one- or two-sided (note: only common tests should be described solely by name; more complex techniques should be described in the Methods section) |
| <input type="checkbox"/> | <input checked="" type="checkbox"/> A description of any assumptions or corrections, such as an adjustment for multiple comparisons  |
| <input type="checkbox"/> | <input checked="" type="checkbox"/> The test results (e.g. $P$ values) given as exact values whenever possible and with confidence intervals noted   |
| <input type="checkbox"/> | <input checked="" type="checkbox"/> A clear description of statistics including <u>central tendency</u> (e.g. median, mean) and <u>variation</u> (e.g. standard deviation, interquartile range)  |
| <input type="checkbox"/> | <input checked="" type="checkbox"/> Clearly defined error bars   |

See the web collection on [statistics for biologists](#) for further resources and guidance.

## ► Software

Policy information about [availability of computer code](#)

### 7. Software

Describe the software used to analyze the data in this study.

TopHat(version 2.0.12) was applied to align clean reads to the hg19 human transcriptome downloaded from UCSC.The Seurat (version 1.2.1) package implemented in R was applied to identify major cell types. randomForest (version4.6.12) was applied to identify subtypes.The Monocle(version2.6.1) package was applied to analyze cell lineage developmental relationships. GSEA was applied to identify priori defined gene sets that show statistically significant differences between two given clusters.

For manuscripts utilizing custom algorithms or software that are central to the paper but not yet described in the published literature, software must be made available to editors and reviewers upon request. We strongly encourage code deposition in a community repository (e.g. GitHub). [Nature Methods guidance for providing algorithms and software for publication](#) provides further information on this topic.

## ► Materials and reagents

Policy information about [availability of materials](#)

### 8. Materials availability

Indicate whether there are restrictions on availability of unique materials or if these materials are only available for distribution by a for-profit company.

No restrictions.

### 9. Antibodies

Describe the antibodies used and how they were validated for use in the system under study (i.e. assay and species).

Description,supplier,catalog number,lot number,dilutions,species reactivity,application; Rabbit polyclonal to FOXP2,Abcam,ab16046,GR191926-1,1:500,"Mouse, Rat, Human", "WB, IHC-FoFr, IHC-Fr, ICC/IF, IHC-P";  
 Mouse monoclonal [SATBA4B10] to SATB2,Abcam,ab51502,GR70015-8,1:250, "Mouse, Rat, Human, Zebrafish", "IP, ICC/IF, IHC-FoFr, WB, IHC-Fr, IHC-P";  
 Mouse monoclonal [MEM-28] to CD45,Abcam,ab8216, GR302332-1,1:100,"Human", "Flow Cyt, IP, IHC-P, WB, ICC/IF";  
 Goat polyclonal to AIF1,Abcam,ab5076, 1428158,1:200,"Rabbit, Guinea pig, Cow, Dog, Human, Pig, Common marmoset", "Electron Microscopy, IHC-Fr, IHC-P, WB, IHC-FrFl, ICC, ICC/IF";  
 Rabbit monoclonal [EPR7003] to SFRP1,Abcam,ab126613,GR82085-19,1:500, "Human", "WB, IHC-P, ICC/IF";  
 Mouse monoclonal [1G10] to A2BP1 / RBFOX1,Abcam,ab183348,GR298959-2,1:500, "Mouse, Rat, Cow, Human", "ICC/IF, WB";  
 Rabbit polyclonal to TTF1,Abcam,ab86023,785740,1:300,"Mouse, Human"; "ICC/IF, WB";  
 Rabbit polyclonal to NEUROD2,Abcam,ab104430,GR94291-4,1:500,"Mouse, Human", "IHC-Fr, IP, IHC-P, WB";  
 Rabbit Polyclonal to PAX6,BioLegend,901301,B201255,1:500,"Human, Mouse, Mammalian", "WB, IHC, IF";  
 Chicken polyclonal to EOMES, Millipore AB15894, 2697506, 1:500,"Mouse, Rat, Human, Avian", "WB, IHC";  
 Goat polyclonal to SOX2,Santa Cruz,sc-17320,H1406,1:250,"Mouse, Rat, Human", "WB, IP, IF, IHC, IHP, ELISA";  
 Mouse monoclonal to NEUROD1, Abcam,ab60704,GR3183945-2,1:100,"Mouse, Human, Apternotus leptorhynchus", "IHC-Fr, IHC-P, WB, ELISA, ICC/IF, Flow Cyt" ;  
 Rabbit monoclonal [EPR18114] to HMGA2, Abcam, ab207301, 1:100,"Human", "IHC-P, ICC/IF" ;  
 Rabbit polyclonal to HOPX, Santa Cruz,sc-30216, D1615,1:1000,"Mouse, Rat, Human", "WB, IP, IF, IHC, IHP, ELISA";  
 Mouse monoclonal [B56] to Ki67, BD Biosciences, 550609, 19679, 1:100,"Human, Mouse, Rat, Rhesus", "Flow Cyt, IHC-Fr".

## 10. Eukaryotic cell lines

a. State the source of each eukaryotic cell line used.

No eukaryotic cell lines were used.

b. Describe the method of cell line authentication used.

No eukaryotic cell lines were used.

c. Report whether the cell lines were tested for mycoplasma contamination.

No eukaryotic cell lines were used.

d. If any of the cell lines used are listed in the database of commonly misidentified cell lines maintained by [ICLAC](#), provide a scientific rationale for their use.

No commonly misidentified cell lines were used.

## ► Animals and human research participants

Policy information about [studies involving animals](#); when reporting animal research, follow the [ARRIVE guidelines](#)

## 11. Description of research animals

Provide details on animals and/or animal-derived materials used in the study.

No animals were used.

Policy information about [studies involving human research participants](#)

## 12. Description of human research participants

Describe the covariate-relevant population characteristics of the human research participants.

Human pre-frontal cortex of gestational weeks 8-26 (GW 8, 9, 10, 12, 13, 16, 19, 23 and 26 weeks) were obtained from the aborted embryos under the agreement of puerperae.