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TemporalVAE for Temporal Prediction on Small Datasets

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TemporalVAE

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We use this protocol and it's working

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Abstract

TemporalVAE is a deep generative model in a dual-objective setting to infer the biological time of cells from a compressed latent space.

TemporalVAE exhibits scalability to millions of cells in the mouse development atlas and high accuracy in atlas-based cell staging on mouse organogenesis across platforms, as well as during human peri-implantation between in vivo and in vitro conditions.

Here we describe the workflow of TemporalVAE, including predicting the biological time of samples and identifying temporally sensitive genes.

Abstract

1 The most important function of TemporalVAE is to predict biological time of cells, we compare the performance of TemporalVAE with multiple existing methods rooted in varied theoretical principles, including **Psupertime**, **Calderon22**, OT-Regressor, Seurat, PCA, LR, and RF.

Here we use three small datasets from Psupertime's experiments, including **Acinar cells**, **Human germline F**, and **Embryonic beta cells**.

Before start: download orginal data file, convert it into a .CSV file with a unified format, and save at folder named demo/data_fromPsupertime

2 **Acinar dataset**
Directly downloaded through the R-package of Psupertime.
R code:

```
setwd('demo/Fig2_TemproalVAE_against_benchmark_methods')

# make Acinar cell dataset ----------------------------------------
---------------------
suppressPackageStartupMessages({
        library('psupertime')
        library('SingleCellExperiment')
})
knitr::opts_chunk$set(collapse = TRUE,comment =
"#>",package.startup.message = FALSE)

# load the data Acinar cells: total 8 donors
data(acinar_hvg_sce)

write.csv(acinar_hvg_sce@assays[[".-
>data"]]@listData[["logcounts"]], file =
"data_fromPsupertime/acinar_hvg_sce_X.csv", row.names = TRUE)
write.csv(acinar_hvg_sce$donor_age,file =
"data_fromPsupertime/acinar_hvg_sce_Y.csv", row.names = FALSE)
```

3 **Embryonic beta cells**
Orginal data can access on **GEO GSE87375** and the developmental stage of beta cells includes **E17.5, P0, P3, P9, P15, P18 and P60**. **We labelled E17.5 as -1**, which is an

embryonic stage before the other stage, and **the other stage as 0, 3, 9, 15, 18, 60**. Then we pre-processed data by the pre-processing function from Psupertime.

Python code:

```
# -*-coding:utf-8 -*-

from demo.Fig2_TemproalVAE_against_benchmark_methods.pypsupertime
import Psupertime
import anndata
import pandas as pd
import os
import numpy as np

os.chdir('demo/Fig2_TemproalVAE_against_benchmark_methods')


def main():
    # for mouse embryonic beta cells dataset:
    result_file_name = "embryoBeta"
    data_org =
pd.read_csv('data_fromPsupertime/GSE87375_Single_Cell_RNA-
seq_Gene_TPM.txt', index_col=0, sep="\t").T
    # get beta cell
    cell_id = data_org.index[1:]
    cell_beta_id = [i for i in cell_id if i[0] == "b"]
    data_org.columns = data_org.iloc[0]
    data_org = data_org[1:]
    data_org = data_org.loc[:, ~data_org.columns.duplicated()]

    adata = anndata.AnnData(data_org)
    adata.var_names_make_unique()
    adata = adata[cell_beta_id].copy()
    temp_time = np.array(adata.obs_names)
    temp_time = [eval(i.split("_")[0].replace("bP",
"").replace("bE17.5", "-1")) for i in temp_time]  # note bE17.5 is
before birth

    adata.obs["time"] = temp_time
    import scanpy as sc
    # sc.pl.highest_expr_genes(adata, n_top=20, )
    sc.pp.filter_genes(adata, min_cells=25)

    adata.var['ERCC'] = adata.var_names.str.startswith('ERCC-')  #
annotate the group of mitochondrial genes as 'ERCC'
    adata = adata[:, ~adata.var.ERCC]
    adata.var['RP'] = adata.var_names.str.startswith('RP')  #
annotate the group of mitochondrial genes as 'RP'
    adata = adata[:, ~adata.var.RP]
```

```
    # sc.pp.calculate_qc_metrics(adata, qc_vars=['ERCC'],
percent_top=None, log1p=False, inplace=True)
    # sc.pl.violin(adata, ['n_genes_by_counts', 'total_counts',
'pct_counts_ERCC'],jitter=0.4, multi_panel=True)
    # sc.pl.scatter(adata, x='total_counts', y='pct_counts_mt')
    # sc.pl.scatter(adata, x='total_counts',
y='n_genes_by_counts')
    preprocessing_params = {"select_genes": "hvg", "log": True}
    # to get hvg gene of humanGermline dataset
    tp = Psupertime(n_jobs=1, n_folds=5,
preprocessing_params=preprocessing_params)
    adata_hvg = tp.preprocessing.fit_transform(adata.copy())
    del tp

    hvg_gene_df = pd.DataFrame(adata_hvg.var_names)
    hvg_gene_df = hvg_gene_df.rename(columns={'Symbol':
'gene_name'})

hvg_gene_df.to_csv(f'{os.getcwd()}/data_fromPsupertime/{result_fil
e_name}_gene_list.csv', index=True)

    x_df = data_org.loc[adata_hvg.obs_names]
    x_df = x_df[hvg_gene_df["gene_name"]]
    x_df = x_df.T

x_df.to_csv(f'{os.getcwd()}/data_fromPsupertime/{result_file_name}
_X.csv', index=True)

    y_df = pd.DataFrame(adata_hvg.obs.time)

y_df.to_csv(f'{os.getcwd()}/data_fromPsupertime/{result_file_name}
_Y.csv', index=True)

    print("Finish save files.")


if __name__ == '__main__':
    main()
```

4 **Human germline F**

Orginal dataset can be access on **GEO GSE86146**. We downloaded the raw count dataset and selected **Female cells** and pre-processed data by the function from

Psupertime.

4.1 Use following R code to combine .txt.gz files:

R code:

```
setwd('demo/Fig2_TemproalVAE_against_benchmark_methods')
# make Human germline dataset ------------------------------------
------------------------

folder_path <- "data_fromPsupertime/GSE86146"

file_list <- list.files(folder_path, pattern = "\\.txt\\.gz$",
full.names = TRUE)

merged_data <- NULL
for (file_path in file_list) {
  data <- read.table(file_path, header = TRUE, sep = "\t", quote =
"",row.names=1)

  if (is.null(merged_data)) {
    merged_data <- data
  } else {
    merged_data <- cbind(merged_data, data)
  }
}

write.csv(merged_data, file =
"data_fromPsupertime/humanGermline_X.csv", row.names = TRUE)
# make label dataframe
new_data <- data.frame()

col_names <- colnames(merged_data)

for (i in 1:length(col_names)) {
  col_name <- col_names[i]
  col_name_parts <- unlist(strsplit(col_name, "_"))

  new_row <- data.frame(
    "sex" = col_name_parts[1],
    "time" = as.numeric(gsub("W", "", col_name_parts[2])),
    row.names = col_name
  )

  new_data <- rbind(new_data, new_row)
}

print(new_data)

write.csv(new_data,file =
```

```
"data_fromPsupertime/humanGermline_Y.csv", row.names = TRUE)
```

4.2 Remove male cell，re-generate data file.
Python code:

```
# -*-coding:utf-8 -*-


from demo.Fig2_TemproalVAE_against_benchmark_methods.pypsupertime
import Psupertime
import anndata
import pandas as pd
import os
import numpy as np

os.chdir('demo/Fig2_TemproalVAE_against_benchmark_methods')


def main():
    # for Human Germline dataset:
    result_file_name = "humanGermline"
    data_x_df =
pd.read_csv('data_fromPsupertime/humanGermline_X.csv',
index_col=0).T
    data_y_df =
pd.read_csv('data_fromPsupertime/humanGermline_Y.csv',
index_col=0)
    # only use female cell
    data_y_df = data_y_df[data_y_df['sex'] != 'M']
    data_x_df = data_x_df.loc[data_y_df.index]
    data_y_df = data_y_df["time"]
    preprocessing_params = {"select_genes": "hvg", "log": True}

    # START HERE
    adata = anndata.AnnData(data_x_df)
    adata.obs["time"] = data_y_df
    print(f"Input Data: n_genes={adata.n_vars}, n_cells=
{adata.n_obs}")

    # to get hvg gene of humanGermline dataset
    tp = Psupertime(n_jobs=1, n_folds=5,
                    preprocessing_params=preprocessing_params)
    adata_hvg = tp.preprocessing.fit_transform(adata.copy())
    hvg_gene = pd.DataFrame(adata_hvg.var_names, columns=
["gene_name"])

hvg_gene.to_csv(f'{os.getcwd()}/data_fromPsupertime/{result_file_n
ame}_gene_list.csv', index=True)
```

```
    x_df = data_x_df.loc[adata_hvg.obs_names]
    x_df = x_df[hvg_gene["gene_name"]]
    x_df = x_df.T

x_df.to_csv(f'{os.getcwd()}/data_fromPsupertime/{result_file_name}
_X.csv', index=True)

    y_df = pd.DataFrame(adata_hvg.obs.time)

y_df.to_csv(f'{os.getcwd()}/data_fromPsupertime/{result_file_name}
_Y.csv', index=True)

    print("Finish save files.")


if __name__ == '__main__':
    main()
```

Run the analysis code and get results

5 To facilitate the use of the Python code, we provide two GitHub repositories. **TemporalVAE** serves as a user-friendly method library, encapsulating all core functionalities of TemporalVAE for straightforward deployment. **TemporalVAE-reproducibility**, while integrating all the functionalities and source code from **TemporalVAE**, further includes demos that enable the reproduction of all results presented in the corresponding article.

5.1 Configure the environment according to the **README.md** provided in the GitHub repository.

5.2 Download code from https://github.com/StatBiomed/TemporalVAE-reproducibility/tree/releaseV1.0/demo and run the methods using the .py files listed in the table below.

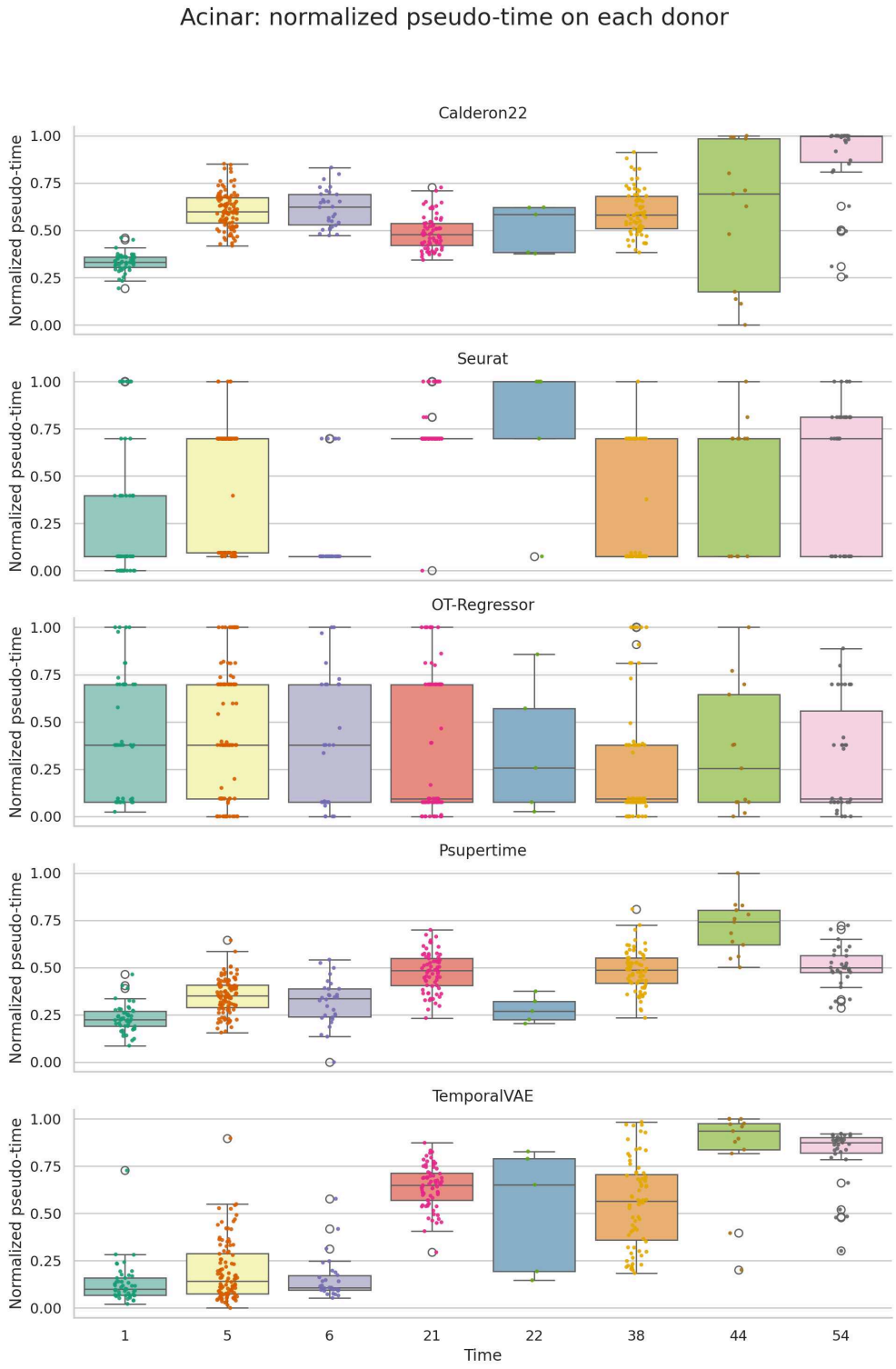
	Method	File name
	Linear regression	exp2_LR_toyDataset.py
	OT-Regressor	exp2_ot_toyDataset.py
	PCA	exp2_pca_toyDataset.py

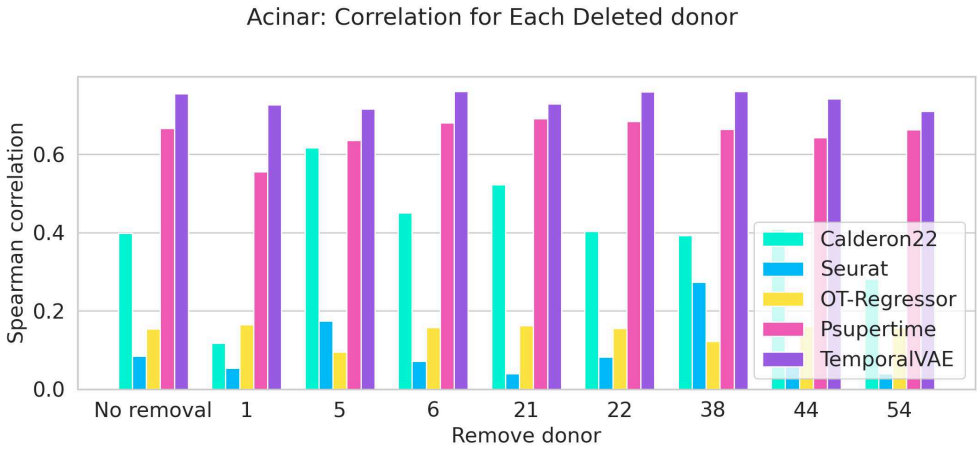
	Method	File name
	Psupertime	exp2_psupertime_toyDataset.py
	Random forest	exp2_randomForest_toyDataset.py
	Calderon22	exp2_science2022_toyDataset.py
	Seurat	exp2_seurat_toyDataset.R
	Vanilla VAE with Linear regression	exp2_VAEwithLR_toyDataset.py
	TemporalVAE	exp2_temporalVAE_toyDataset.py

After run method's .py file, the result will save at *demo/Fig2_TemproalVAE_against_benchmark_methods/{**method**}_results/*, including *{**dataset**}_{**method**}_result.csv* and a prediction density image namd *{**dataset**}_labelsOverPsupertime.png*

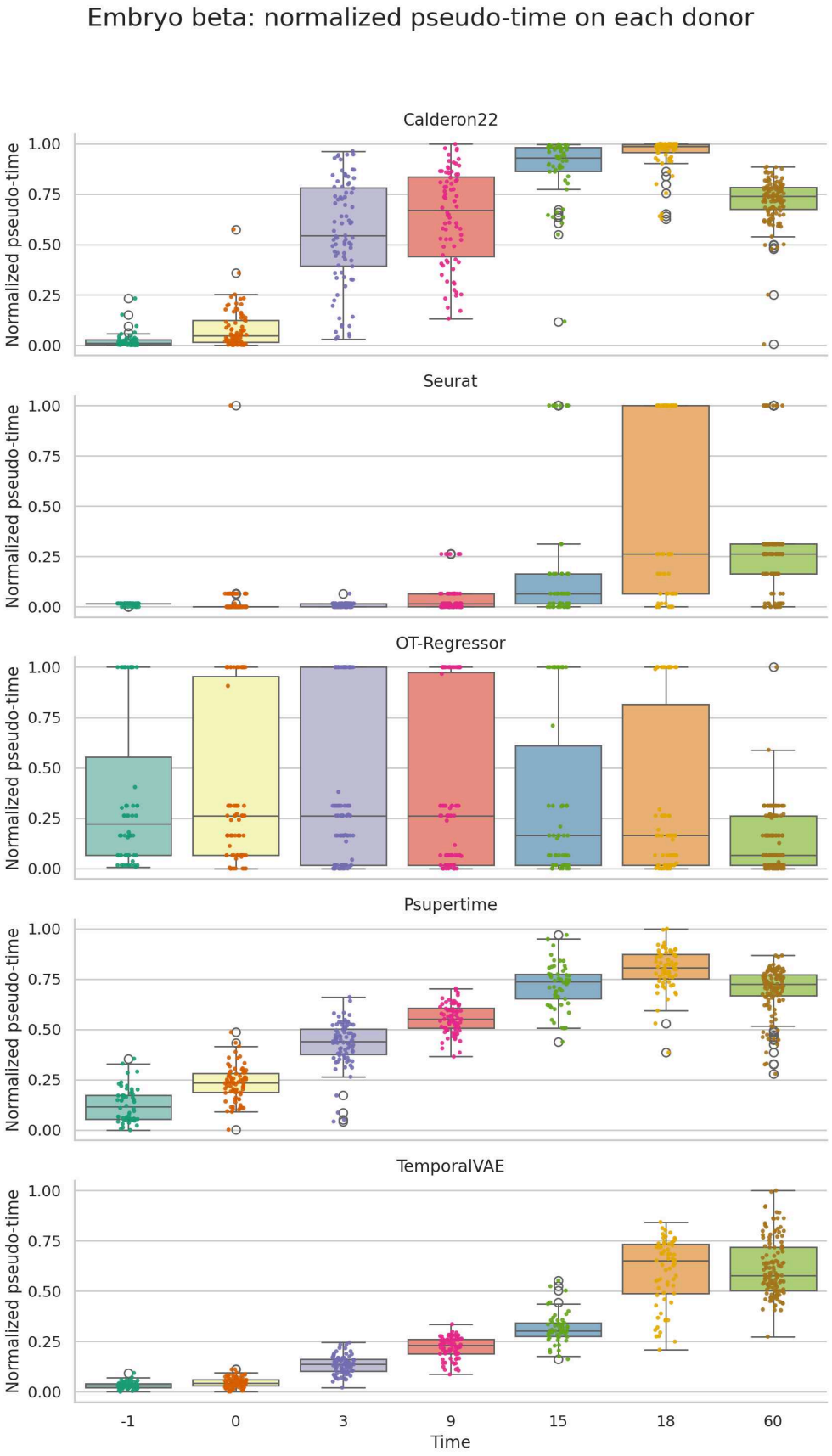
5.3 Run the **plotting code** to obtain comparison graphs of the performance of various methods across three datasets.
The result of each dataset as following:

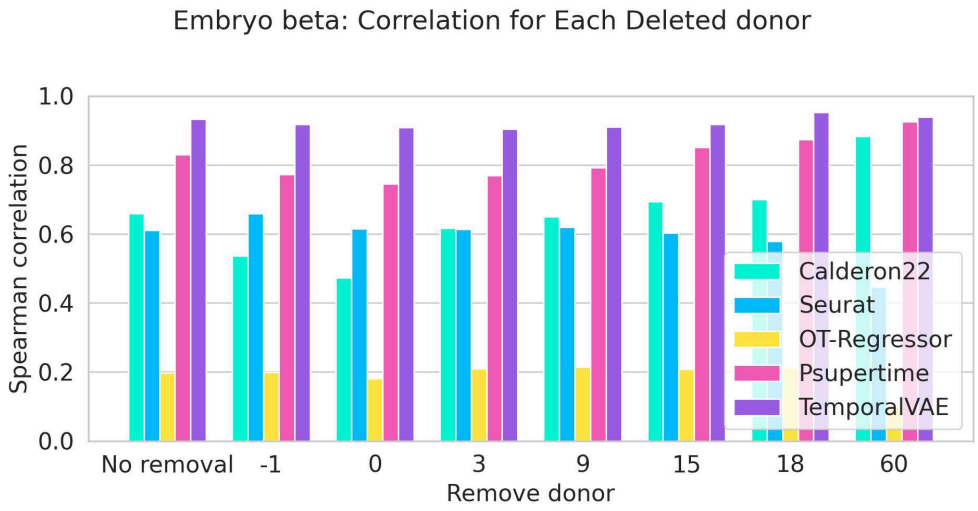
- **Acinar dataset**


Acinar: normalized pseudo-time on each donor
Calderon22
Seurat
OT-Regressor
Psupertime
TemporalVAE
Normalized pseudo-time
0.00
0.25
0.50
0.75
1.00
1
5
6
21
22
38
44
54
Time

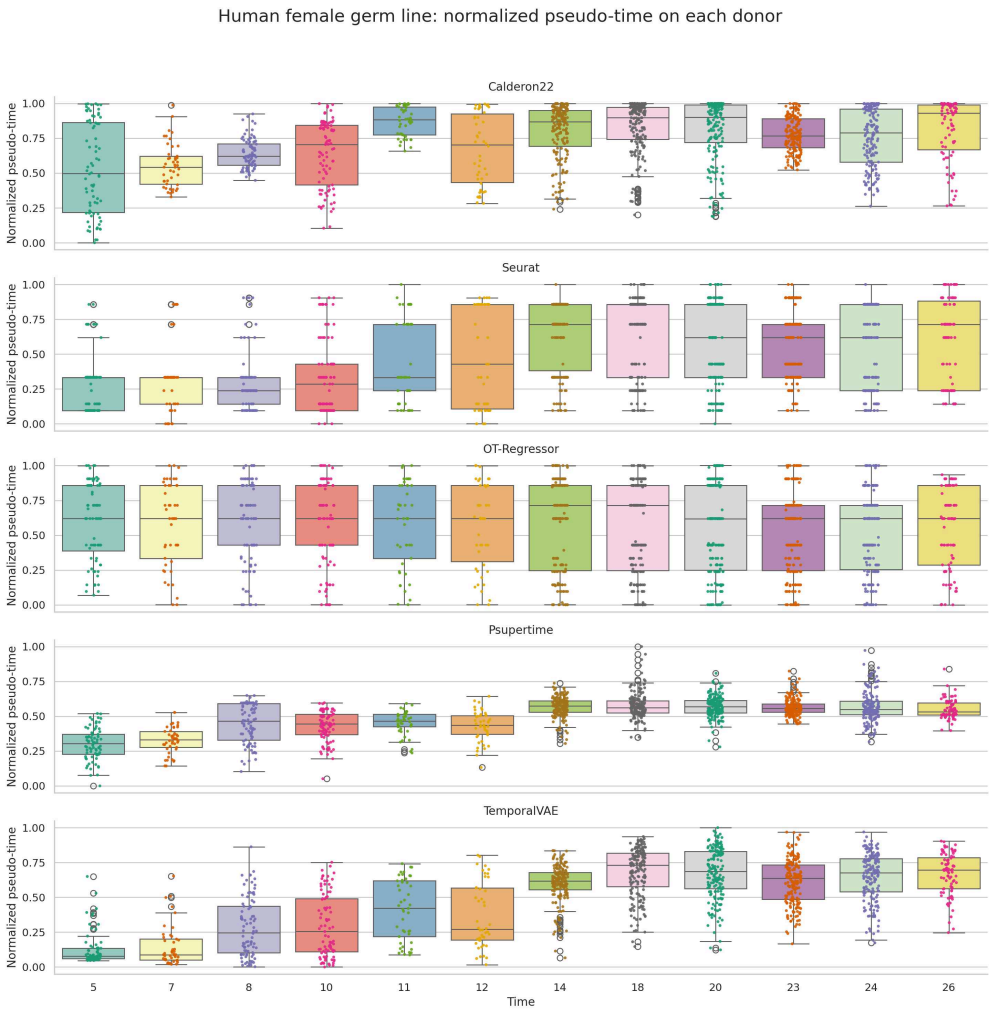


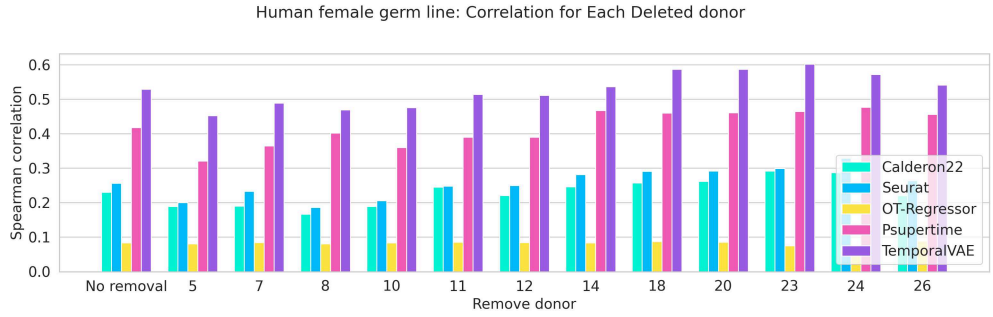
- **Embryonic beta cells**


Embryo beta: normalized pseudo-time on each donor
Calderon22
Seurat
OT-Regressor
Psupertime
TemporalVAE
Normalized pseudo-time
0.00
0.25
0.50
0.75
1.00
-1
0
3
9
15
18
60
Time



- **Human germline F**


Human female germ line: normalized pseudo-time on each donor
Calderon22
Seurat
OT-Regressor
Psupertime
TemporalVAE
Normalized pseudo-time
0.00
0.25
0.50
0.75
1.00
5
7
8
10
11
12
14
18
20
23
24
26
Time


Human female germ line: Correlation for Each Deleted donor
Spearman correlation
0.0
0.1
0.2
0.3
0.4
0.5
0.6
No removal
5
7
8
10
11
12
14
18
20
23
24
26
Remove donor
Calderon22
Seurat
OT-Regressor
Psupertime
TemporalVAE