# 2019ADS2 Week10 T test

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#### 1. Introduction

This R Markdown file contains a tutorial of how to do different form of t test manually and in R. For those problems that need to workout manually, please include the formula and process of how you get the final number.

## 2. one sample t test

Let's say a gene called SOX17, from the published dataset, we found that its average expression in different lines (more than 30) of human embryonic stem cells is 8.9 (unit in RPKM). From your experiment, you used a new human embryonic stem cell line that is generated in ZJE, and you did RNA-seq of three biological replicates to check its expression in this cell line and found out its expression is 15, 30,50 (RPKM). Is there enough evidence to show our embryonic stem cell's SOX17 expression is very different from others under significance level of 0.05?

Please write out the formula you used to calculte the statistics.

- 1. step 1 : State the hypotheses and identify the claim.
- 2. Step 2: what distribution to use?
- 3. Step 3: Find the critical value.
- 4. Step 4: Compute the test value.
- 5. Step 5: Make the decision.
- 6. Step 6:Summarize the results.

Please write out the R code calculte the statistics.

#### 2. two sample t test

# 2.1 paired two sample t test

We followed a set of 5 paitents with acute myeloid leukemia. We want to investigate whether the oncogene AML1 expression is repressed after a new treatment. Thus we tested their AML1 expression before and after the therapy. The gene expression level fo AML1 before the treatment is:

$$x_1, x_2, x_3, x_4, x_5 = c(102, 340, 234, 332, 129)$$

. And the gene expression level fo AML1 after the treatment is:

$$y_1, y_2, y_3, y_4, y_5 = c(74, 56, 70, 104, 11)$$

. Is there enough evidence to support the claim that the new treatment significantly reduce the AML1 expression level in acute myeloid leukemia paitents under significance level of 0.05?

- 1. step 1: State the hypotheses and identify the claim.
- 2. Step 2: what distribution to use?
- 3. Step 3: Find the critical value.
- 4. Step 4: Compute the test value.
- 5. Step 5: Make the decision.
- 6. Step 6:Summarize the results.

Please write out the R code calculte the statistics.

# 2.2 unpaired two sample t test

There are two types of human embryonic stem cells (naive vs primed). We have the RNAseq data for naive hESC (4 biological replicates) and primed hESCs (4 biological replicates). In each RNAseq dataset, there are 23368 genes identified. We want to find out those genes that are significantly differential expressed (either up regulated or down regulated) under significance level of 0.05. Hint, use unpaired t test to find out genes with p-value less than 0.05.

```
geneexp=read.csv("week10_t_test_problemset_testdata.csv")
head(geneexp)
```

```
gname naive_hESC_r1 naive_hESC_r2 naive_hESC_r3 naive_hESC_r4
##
## 1 1/2-SBSRNA4
                           3.657
                                          3.808
                                                         7.239
                                                                        4.607
## 2
             A1BG
                           0.038
                                          0.035
                                                         0.146
                                                                        0.028
## 3
        A1BG-AS1
                           0.032
                                          0.348
                                                         0.361
                                                                        0.299
## 4
             A1CF
                           0.004
                                          0.000
                                                         0.006
                                                                        0.003
## 5
           A2LD1
                           0.490
                                                         0.192
                                          0.404
                                                                        0.137
## 6
              A2M
                           0.087
                                          0.067
                                                         0.089
                                                                        0.063
     primed_hESC_r1 primed_hESC_r2 primed_hESC_r3 primed_hESC_r4
##
## 1
               4.429
                               8.190
                                               3.364
                                                               6.431
## 2
                               0.096
                                               0.000
                                                               0.034
               0.120
## 3
                               0.356
                                               0.201
                                                               0.527
               0.331
## 4
               0.026
                                               0.009
                                                               0.006
                               0.036
## 5
               0.264
                               0.315
                                               0.120
                                                               0.221
## 6
               0.549
                               0.801
                                               0.521
                                                               0.728
```

## tail(geneexp)

##		gname	naive_hES0	C_r1	naive_hES	SC_r2	naive_hES0	C_r3	naive_hESC_r4
##	23363	ZXDC	5	.472	7	7.170	6	.914	6.781
##	23364	ZYG11A	38	.996	40	783	43	.214	34.863
##	23365	ZYG11B	9	.271	Ş	9.741	7	. 855	9.625
##	23366	ZYX	11	.731	7	7.882	7	. 685	8.729
##	23367	ZZEF1	9	.343	Ş	9.973	7	.814	7.089
##	23368	ZZZ3	20	.996	14	1.505	9	. 251	11.654
##		primed	hESC_r1 p	rimed	d_hESC_r2	prime	ed_hESC_r3	pri	med_hESC_r4
##	23363		5.866		6.931		4.513		5.958
##	23364		4.714		7.006		3.506		4.975
##	23365		8.533		9.096		6.318		7.359
##	23366		3.450		2.543		1.304		3.481
##	23367		2.608		3.334		1.208		2.705
##	23368		18.732		16.479		16.563		17.609

# dim(geneexp)

**##** [1] 23368 9

# 2.3 One step further - Multiple testing correction (Advanced thinking, optional)

## 2.3.1 Why Multiple Testing Matters?

Genomics usually have Lots of Data which means there will be lots of Hypothesis Tests in one experiment. For example, a typical RNAseq experiment might result in performing 20000 separate hypothesis tests (like what we did before). If we use a standard p-value cut-off of 0.05, we'd expect 1000 (20000\*0.05) genes to be deemed 'significant' by chance (not reasonable). Thus we usually will perform multiple testing correction after we calculate p-value for genomics. You can refer to this coursera online course video if you are interested in https://www.coursera.org/lecture/statistical-genomics/multiple-testing-8-25-NsJfs

#### R Markdown

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