

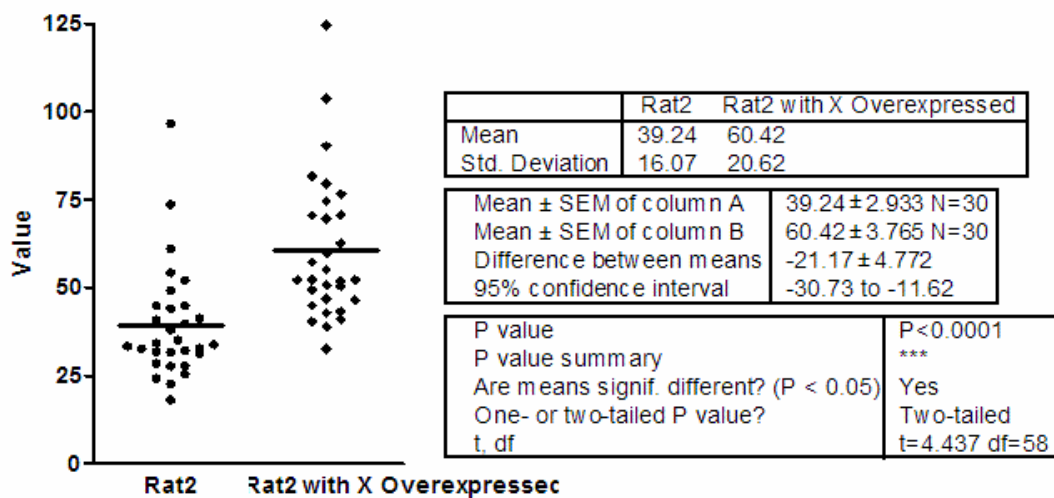
Data Set:**Velocity of Rat2 cells (um/hr):**

73.44, 96.48, 33.696, 60.84, 48.96, 41.04, 31.86, 44.64, 17.892, 43.92, 31.428, 23.976, 39.6, 27.576, 30.996, 32.472, 34.056, 40.68, 25.128, 28.332, 33.156, 22.356, 32.508, 44.64, 27.504, 31.572, 54, 51.84, 34.848, 37.8;

**Velocity of Rat2 cells with protein X over-expressed (um/hr):**

69.48, 70.56, 46.44, 43.2, 74.52, 52.2, 103.68, 45, 62.64, 38.88, 46.8, 76.68, 49.32, 52.2, 51.84, 70.56, 81.72, 79.56, 59.76, 40.32, 124.56, 52.2, 50.76, 32.616, 42.84, 55.08, 50.4, 57.24, 41.04, 90.36.

Set  $\alpha=0.05$ . Use *Prism*<sup>®</sup> 4 to do two-sample t-test, and have the computer printout:



a)

Rat2 is a fibroblast cell line, which is suitable for single cell tracking. Our lab interests in protein X's function in cell migration and actin dynamics. A stable Rat2 cell with protein X over-expressed was constructed by retrovirus infection and FACS. Our hypothesis is that over-expression of protein X in Rat2 cells affects cytoskeleton and influences cell migration.

b)

Dependent variables: cell culture environment, cell culture condition, genetic background of Rat2 cell line (before retrovirus infection), cell-confluence during tracking experiments, parameter-setting of tracking program, *etc.*

Independent variables: over-expressed protein X's function in cell migration, *etc.*

c)

Figure out whether over-expression protein X influences cell migration or not.

d)

The required assumptions are random sampling, independent sampling and normal population distribution. In the experiment,  $n=30$ , which is large, and with large  $n$ , sampling distribution of means will be normal. Cells were picked up randomly from a 5-field-movie. The effect of protein X on cell velocity is independent during the experiments.

e)

Null hypothesis: There is no difference between the population mean cell migration speed of Rat2 cells with protein X over-expressed vs. control Rat2 cells.

Alternative hypothesis: protein X over-expression makes the population mean cell migration speed of Rat2 cells different (two-sided).

f)

Alpha level is 0.05.

g)

Degrees of freedom for t is 58.

h)

Value of t-critical is 2.

i)

p value of observed result is  $<0.0001$ .

j)

According the result, I rejected the null hypothesis.

k)

-30.73 to -11.62 is the 95% confidence interval for the population difference.

l)

Data were analyzed with an independent samples t-test, conducted with the PC-based program PRISM (GraphPad, San Diego, CA). Alpha was set at 0.05. The mean cell migration velocity was significantly greater in protein X over-expressed vs. control Rat2 cells [ $t(58)=4.437$ ,  $p<0.0001$ ].

m)

An alternative hypothesis is that the over-expression of protein X speeds up cell migration velocity to two fold. If I want to detect this difference, according to the previous experiment  $d=20.62/20.62=1$ ,  $\alpha=0.05$  (two-tailed),  $\beta=0.20$ , I would need to have 18 subjects in each group.