The Effect of Vitamin C on Tooth Growth in Guinea Pigs

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Synopsis

Based on the `ToothGrowth` data from the R `datasets` package, we analyze the effect of vitamin C on the length of odontoblasts in guinea pigs. We want to study whether the amount of vitamin C or the method it is delivered to guinea pigs affect the length of the odontoblasts. We conjecture that they do.

Data

The dataset contains the length of odontoblasts (cells responsible for tooth growth) in 60 guinea pigs that received different doses of vitamin C by different methods. Each animal received one of three dose levels of vitamin C (0.5, 1, and 2 mg/day) by one of two delivery methods, orange juice or ascorbic acid (a form of vitamin C and coded as VC).

```
set.seed(0);
data("ToothGrowth");
```

Packages

The following packages will be used to perform the analysis.

```
library(dplyr);
library(ggplot2);
```

Exploratory analysis

We start by having a look at the size of our data set: dim = 60 x 3. Hence we have, as expected, 3 observations for 60 subjects. Let us have a look at the head, the tail and the structure of our data

```
##    len supp dose
## 1  4.2   VC  0.5
## 2 11.5   VC  0.5
## 3  7.3   VC  0.5
```

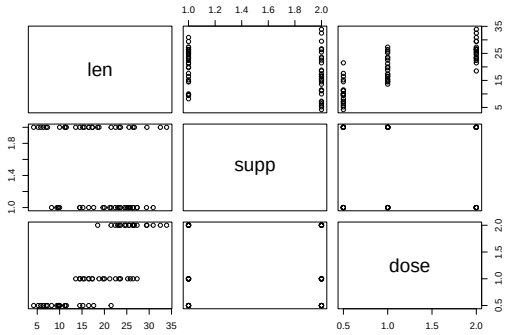
```
##     len supp dose
## 58 27.3   OJ    2
## 59 29.4   OJ    2
## 60 23.0   OJ    2
```

```
## 'data.frame':    60 obs. of  3 variables:
##  $ len : num  4.2 11.5 7.3 5.8 6.4 10 11.2 11.2 5.2 7 ...
##  $ supp: Factor w/ 2 levels "OJ","VC": 2 2 2 2 2 2 2 2 2 2 ...
##  $ dose: num  0.5 0.5 0.5 0.5 0.5 0.5 0.5 0.5 0.5 0.5 ...
```

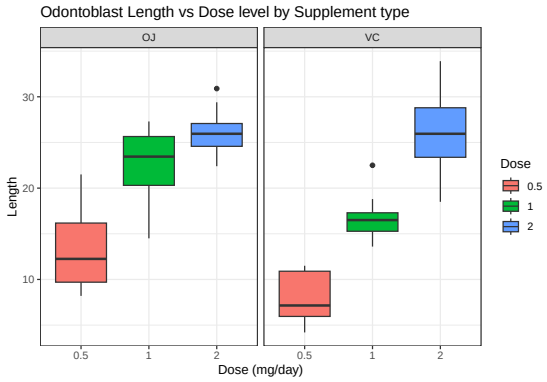
respectively. The dataset is already clean and we may provide a basic summary of the data:

```
##       len        supp         dose
##  Min.   : 4.20   OJ:30   Min.   :0.500
##  1st Qu.:13.07   VC:30   1st Qu.:0.500
##  Median :19.25           Median :1.000
##  Mean   :18.81           Mean   :1.167
##  3rd Qu.:25.27           3rd Qu.:2.000
##  Max.   :33.90           Max.   :2.000
```

By keeping in mind our working questions, we can have a quick look at potential correlations:



Apparently, the dose level has a clear effect, while we cannot draw conclusions about the delivery method, yet. The following additional boxplot, grouped by delivery method, confirms the effect of dosage and suggest a possible effect of delivery method at dose level 0.5 and 1:



Hypothesis tests

In view of the relative small size of samples and under the reasonable hypothesis that the underlying data are iid Gaussian, we are going to perform two group T tests for equality of means. However, it is important to consider the multiple hypothesis testing issue here as we are going to perform 5 tests. We will implicitly apply the Bonferroni correction, so for a level $\alpha = 0.05$ we will consider significant the p-values under 0.01.

Compare tooth growth by dose level

Our first hypothesis is that a higher dose level of vitamin C leads to a higher length of odontoblasts. To support this hypothesis we compare level 1 with level 0.5 and level 2 with level 1, the null hypothesis always being that there is no significant difference in the means and the alternative being that a higher level of vitamin C entails a longer length of odontoblasts.

First we split the three groups according to the rule

- group 1: dose level = 0.5 mg/day,
- group 2: dose level = 1 mg/day,
- group 3: dose level = 2 mg/day

and we compute their sample means:

```
##   grp1   grp2   grp3
## 10.605 19.735 26.100
```

Then we perform a T test to check whether there is a significant difference between the means at level $\alpha = 0.05$. For the sake of space, we only report the p-value. The full output of the test can be found in the appendices. If we compare group 2 (dose level = 1 mg/day) with group 1 (dose level = 0.5 mg/day):

```
t.test(grp2, grp1, alternative = "greater")$p.value
```

```
## [1] 6.341504e-08
```

We can reject the null hypothesis that the two means coincide, since the p-value is much smaller than 0.01. Similarly, if we compare group 3 (dose level = 2 mg/day) with group 2 (dose level = 1 mg/day):

```
t.test(grp3, grp2, alternative = "greater")$p.value
```

```
## [1] 9.532148e-06
```

then we can reject the null hypothesis of equal means, since the p-value is much smaller than 0.01. There is no need, in this case, to test group 3 versus group 1.

We conclude that a higher level of vitamin C is related with a longer length of the odontoblasts at level 0.05.

Compare tooth growth by delivery method

Our second hypothesis is that also the delivery method affects tooth growth. Namely, we suspect that subjects that received vitamin C at dose levels 0.5 and 1 mg/day through orange juice have longer odontoblasts. Instead, we believe that the supply method has no significant effect at a dosage of 2 mg/day.

We separate the three groups by dosage, OJ versus VC, and we compute the respective sample means:

```
## OJgrp1 OJgrp2 OJgrp3 VCgrp1 VCgrp2 VCgrp3
##  13.23  22.70  26.06   7.98   7.98  26.14
```

Then we perform a T test to check whether there is a significant difference between the means at level $\alpha = 0.05$. If we compare the OJ group with the VC group:

```
t.test(suppOJgrp1,suppVCgrp1,alternative = "greater")$p.value
```

```
## [1] 0.003179303
```

```
t.test(suppOJgrp2,suppVCgrp2,alternative = "greater")$p.value
```

```
## [1] 0.0005191879
```

then we can reject the hypothesis of equal means at level 0.05 for the 0.5 and the 1 mg/day dose levels, while

```
t.test(suppOJgrp3,suppVCgrp3,alternative = "greater")$p.value
```

```
## [1] 0.5180742
```

so we cannot reject the null hypothesis of equal means for the highest dosage.

We conclude that there is a relationship between the delivery method and the length of the odontoblasts at lower dosage (0.5 or 1 mg/day): subjects that received vitamin C via orange juice have, on average, longer odontoblasts than those who received it via ascorbic acid.

Appendices

Full outcomes of the T tests to check the effect of dosage.

Group 2 (dose level = 1 mg/day) vs group 1 (dose level = 0.5 mg/day):

```
t.test(grp2, grp1, alternative = "greater")
```

```
##
##  Welch Two Sample t-test
##
## data:  grp2 and grp1
## t = 6.4766, df = 37.986, p-value = 6.342e-08
## alternative hypothesis: true difference in means is greater than 0
## 95 percent confidence interval:
##  6.753323      Inf
## sample estimates:
## mean of x mean of y
##    19.735    10.605
```

Group 3 (dose level = 2 mg/day) vs group 2 (dose level = 1 mg/day):

```
t.test(grp3, grp2, alternative = "greater")
```

```
##
##  Welch Two Sample t-test
##
## data:  grp3 and grp2
## t = 4.9005, df = 37.101, p-value = 9.532e-06
## alternative hypothesis: true difference in means is greater than 0
## 95 percent confidence interval:
##  4.17387     Inf
## sample estimates:
## mean of x mean of y
##    26.100    19.735
```

Full outcomes of the T tests to check the effect of delivery method by dosage

Orange juice vs ascorbic acid at dosage 0.5

```
t.test(suppOJgrp1,suppVCgrp1,alternative = "greater")
```

```
##
##  Welch Two Sample t-test
##
## data:  suppOJgrp1 and suppVCgrp1
## t = 3.1697, df = 14.969, p-value = 0.003179
## alternative hypothesis: true difference in means is greater than 0
## 95 percent confidence interval:
##  2.34604     Inf
## sample estimates:
## mean of x mean of y
##     13.23      7.98
```

Orange juice vs ascorbic acid at dosage 1

```
t.test(suppOJgrp2,suppVCgrp2,alternative = "greater")
```

```
##
##  Welch Two Sample t-test
##
## data:  suppOJgrp2 and suppVCgrp2
## t = 4.0328, df = 15.358, p-value = 0.0005192
## alternative hypothesis: true difference in means is greater than 0
## 95 percent confidence interval:
##  3.356158      Inf
## sample estimates:
## mean of x mean of y
##     22.70     16.77
```

Orange juice vs ascorbic acid at dosage 2

```
t.test(suppOJgrp3,suppVCgrp3,alternative = "greater")
```

```
##
##  Welch Two Sample t-test
##
## data:  suppOJgrp3 and suppVCgrp3
## t = -0.046136, df = 14.04, p-value = 0.5181
## alternative hypothesis: true difference in means is greater than 0
## 95 percent confidence interval:
##  -3.1335     Inf
## sample estimates:
## mean of x mean of y
##     26.06     26.14
```

Readers interested in a literate version of this report, with explicit R code, may refer to the R markdown file on my GitHub repository.