

Mouse behavioral assay - LORR

I am Renjini Ramadasan Nair. The following is a snippet of the R-code I had written to analyze mouse behavioral data pertaining to my project. To summarize, the mice ($N \geq 6$) were subjected to increasing concentrations of volatile anesthesia (ISO or HAL) and their induction and emergence EC50s recorded, for the righting reflex assay. The change between induction and emergence concentrations are plotted as bar, whisker and scatter plots, followed by significance analysis by T-test. This data has been published as part of a research paper in the journal of Anesthesiology.

```
library (ggplot2)
library(gridExtra)
library (forcats)
library (readxl)

# Import the excel sheet as a data frame

lorr1 = as.data.frame (read_excel("together.xls", sheet = "lorr1"))

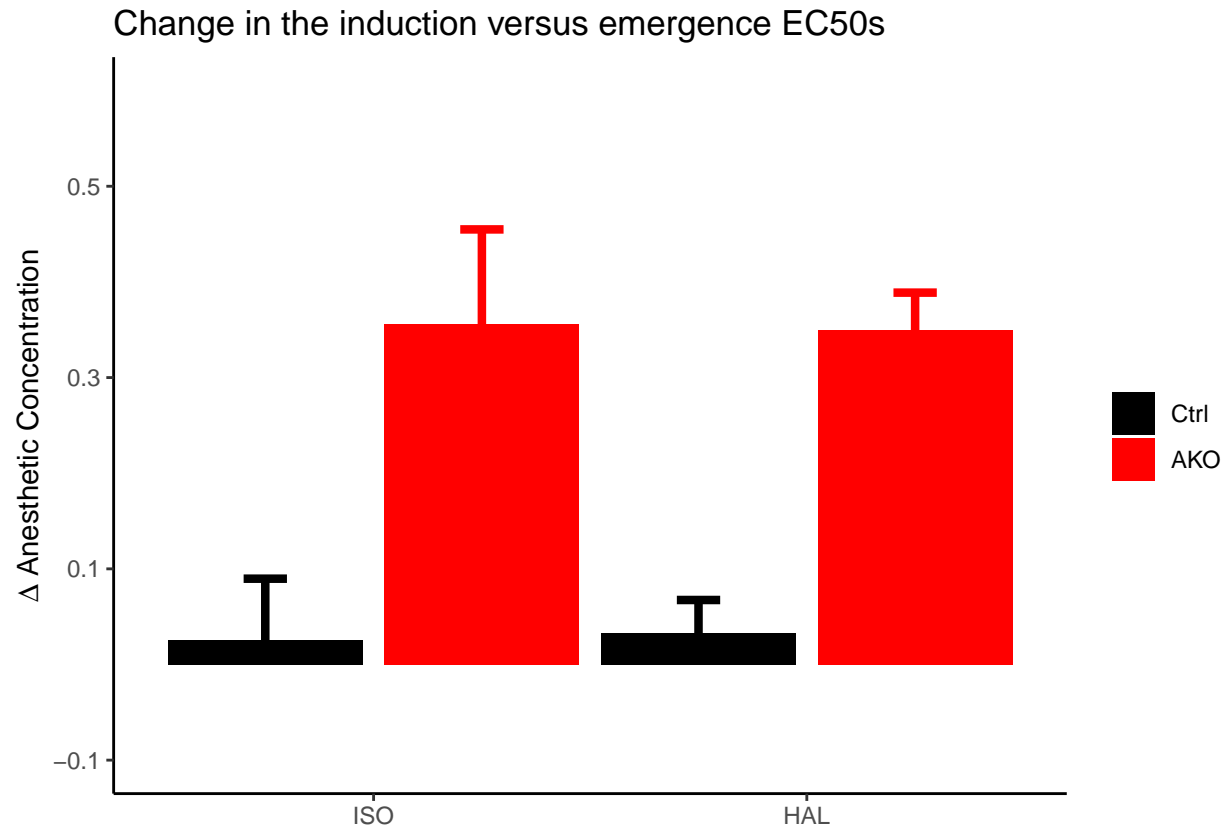
lorr1[, 1:2] = data.frame(apply(lorr1[, 1:2], 2, as.factor))

lorr1$genotype <- factor(lorr1$genotype, levels = c('Ctrl', 'AKO'))
str(lorr1)

## 'data.frame':   28 obs. of  5 variables:
## $ genotype   : Factor w/ 2 levels "Ctrl","AKO": 1 1 1 1 1 1 1 2 2 2 ...
## $ anesthetic: Factor w/ 2 levels "HAL","ISO": 2 2 2 2 2 2 2 2 2 2 ...
## $ change      : num  0.0233 -0.0206 0.0065 -0.0562 0.1082 ...
## $ induction   : num  1.109 0.983 0.889 0.904 1.12 ...
## $ emergence   : num  1.086 1.004 0.882 0.96 1.012 ...

# Plot the mean for the control and mutant (AKO) mice with ISO and HAL (error = SD)
lorr1plot = ggplot (lorr1, aes(x=anesthetic, y=change, fill = genotype)) +
  theme_classic() +
  stat_summary(geom = "col", fun.y=mean, aes (fill=genotype), position = position_dodge(width = 1)) +
  labs (y= expression (Delta~"Anesthetic Concentration"), x=element_blank(), fill=element_blank(), title=
  scale_fill_manual(values = c("black", "red"))+
  stat_summary(fun.ymin = function(x) mean(x)*0.5,fun.ymax = function(x) mean(x) + sd(x), position = position_dodge(width = 1)) +
  guides (color = "none") +
  scale_color_manual(values = c("black", "red")) +
  scale_x_discrete(limits=c("ISO","HAL")) +
  scale_y_continuous(limits = c(-0.1, 0.60), breaks = seq(-0.1, 0.60, 0.20))

lorr1plot
```



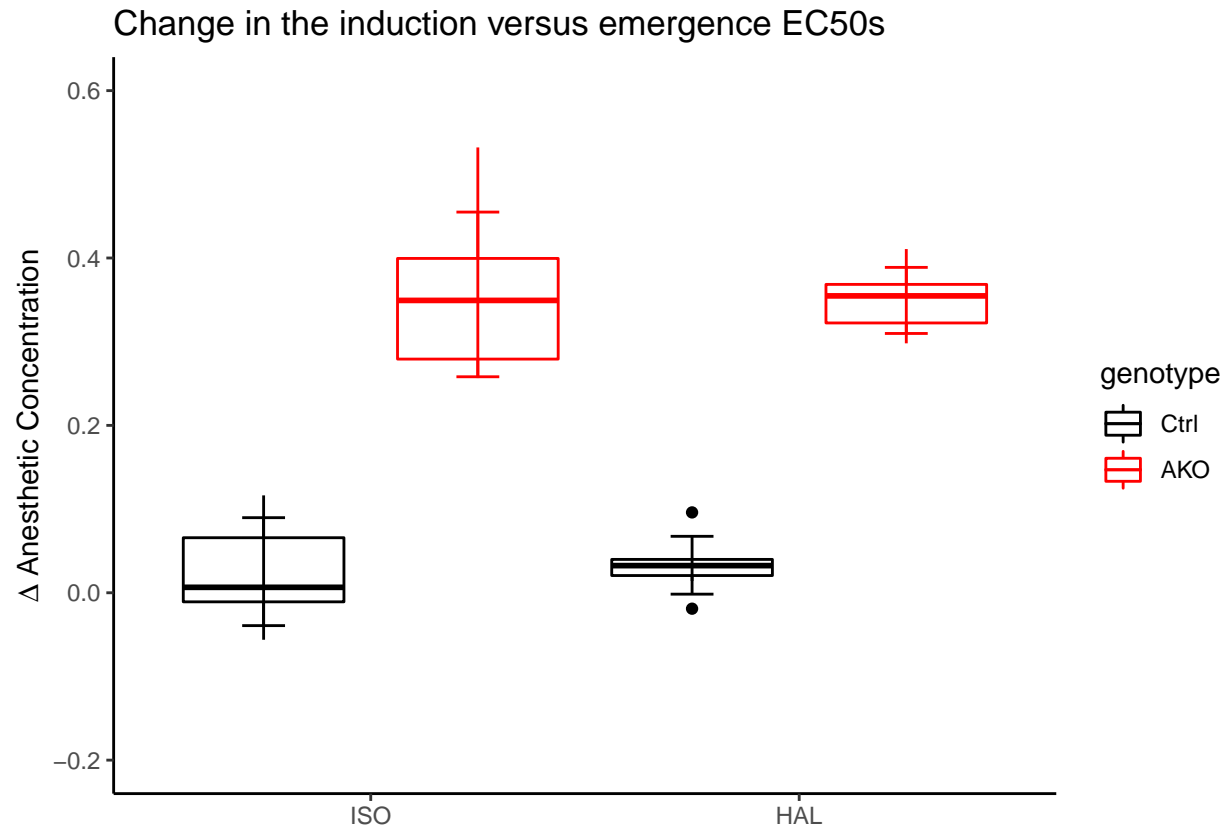
```
# Save the plot as a png file
```

```
ggsave (filename = "lorriplot1.png", plot = lorriplot, dpi = 600, width = 4.5, height = 4, units = "in")
```

```
# Data plotted as boxplots (error bars = SD)
```

```
lorriwhiskerplot = ggplot (lorri1, aes(x=anesthetic, y=change, color = genotype)) + theme_classic() +  
geom_boxplot(position = position_dodge(width = 1)) +  
labs (y= expression (Delta~"Anesthetic Concentration"), x=element_blank(), title = "Change in the induction  
stat_summary(fun.data = mean_sdl, geom = "errorbar", fun.args=list (mult=1), aes (color = genotype), w  
scale_y_continuous(limits = c(-0.20, 0.60), breaks = seq(-0.20, 0.60, 0.20))
```

```
lorriwhiskerplot
```

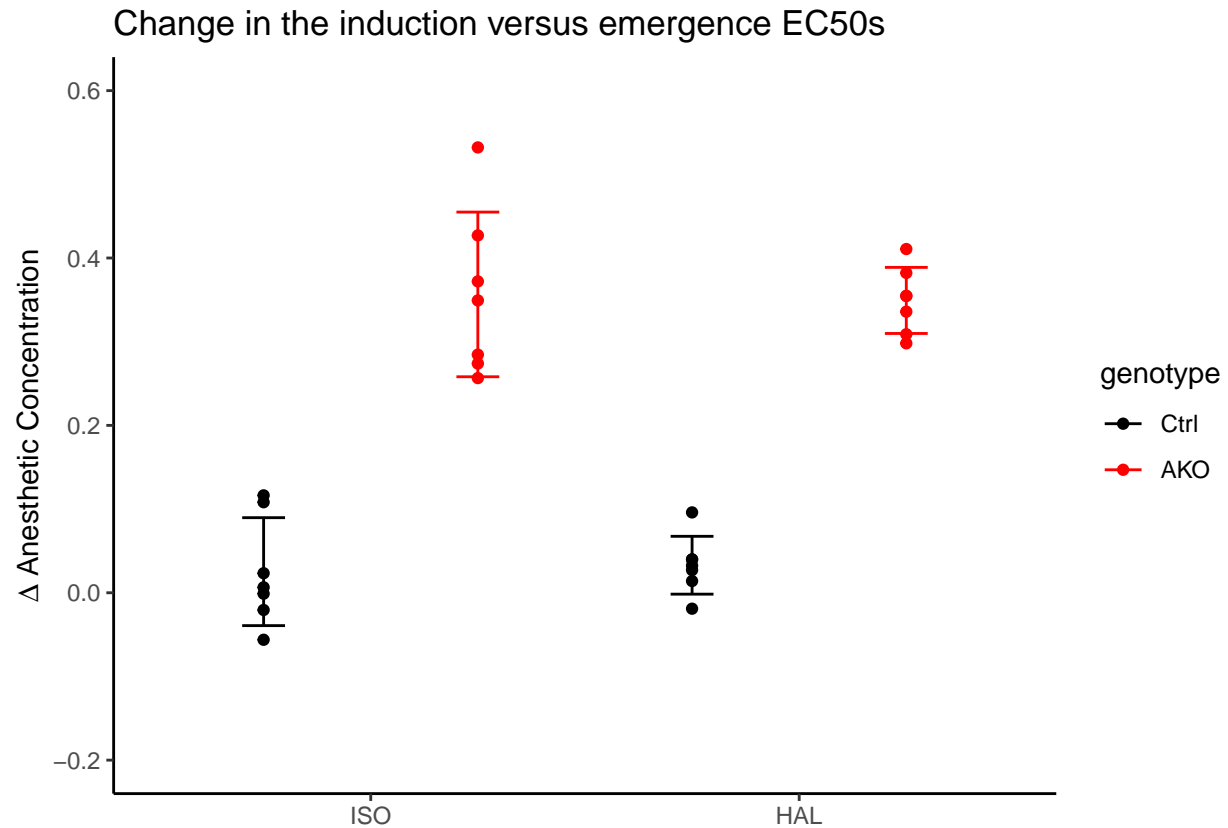


```
ggsave (filename = "lorriscatterplot1.png", plot = lorr1whiskerplot, dpi = 600, width = 4.5, height = 4)
```

```
# Data plotted as scatter plots (error bars = SD)
```

```
lorriscatterplot = ggplot (lorr1, aes(x=anesthetic, y=change, color = genotype)) + theme_classic() +  
geom_point(position = position_dodge(width = 1)) +  
labs (y= expression (Delta~"Anesthetic Concentration"), x=element_blank(), title = "Change in the induc  
stat_summary(fun.data = mean_sdl, geom = "errorbar", fun.args=list (mult=1), aes (color = genotype), w  
scale_y_continuous(limits = c(-0.20, 0.60), breaks = seq(-0.20, 0.60, 0.20))
```

```
lorriscatterplot
```



```
ggsave (filename = "lorriscatterplot1.png", plot = lorriscatterplot, dpi = 600, width = 4.5, height = 4
```

```
# Calculate the statistical significance for the ISO data by performing a two-tailed T-test.
```

```
lorr1iso = lorr1[lorr1$anesthetic == 'ISO', ]
lorr1isot=t.test(change ~ genotype, data=lorr1iso, var.equal = TRUE)
lorr1isot
```

```
##
## Two Sample t-test
##
## data: change by genotype
## t = -7.4492, df = 12, p-value = 7.744e-06
## alternative hypothesis: true difference in means is not equal to 0
## 95 percent confidence interval:
## -0.4281562 -0.2343731
## sample estimates:
## mean in group Ctrl mean in group AKO
## 0.02522156 0.35648617
```

```
# Calculate the statistical significance for the HAL data by performing a two-tailed T-test.
```

```
lorr1hal = lorr1[lorr1$anesthetic == 'HAL', ]
lorr1halt=t.test(change ~ genotype, data=lorr1hal, var.equal = TRUE)
lorr1halt
```

```
##
## Two Sample t-test
```

```
##
## data:  change by genotype
## t = -15.958, df = 12, p-value = 1.911e-09
## alternative hypothesis: true difference in means is not equal to 0
## 95 percent confidence interval:
##  -0.3596267 -0.2732218
## sample estimates:
## mean in group Ctrl  mean in group AKO
##           0.03288958           0.34931381
```