

# Mortality of zoea 1 larvae of *Taliepus dentatus* reared at different temperatures

## Reproducible report

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This document reproduces and supports all data work and statistical analysis of the larval mortality in the paper: swimming performance and morphometrics of the kelp-crab *Taliepus dentatus* zoea 1 exposed to different rearing temperatures.

## 1. Libraries

```
library(cowplot)
library(scales)
library(dplyr)
library(gridExtra)
library(tidyverse)
library(ggsci)
library(Rmisc)
library(survminer)
library(survival)
library(MASS)
```

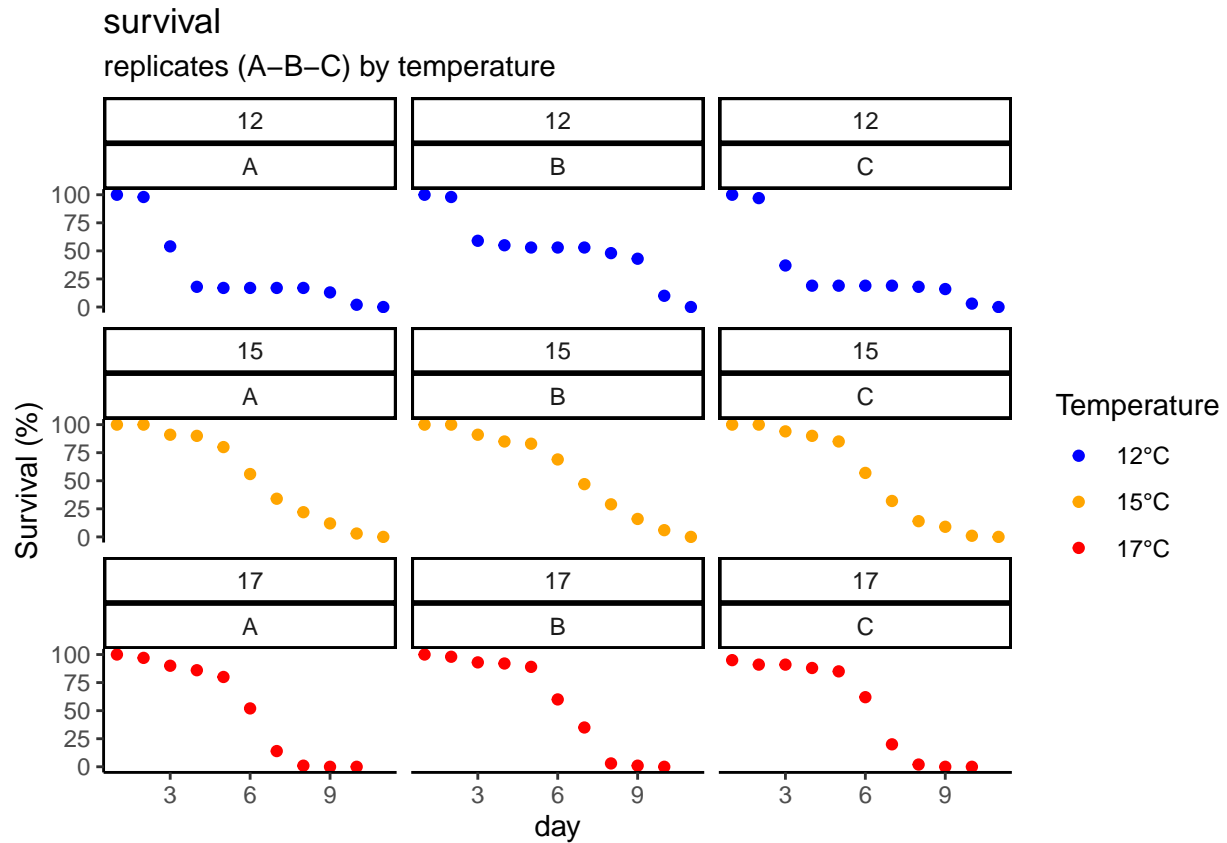
## 2. Import of data base and definition of factors

```
#database import
Mort<-read.delim("~/Desktop/ECOLAB!/paper tesis/mortalidad/mort.txt")
Mort$bino<-Mort$Vivas/100

#Factor
Mort$Temperature<-as.factor(Mort$Temperatura)

#survival plot by temp by each replicate.
Grafico_frascos<-ggplot(Mort, aes(x=Dia, y=Vivas, colour=Temperature))+
  geom_point() + theme_classic() +
  ggtitle("survival")+facet_wrap(~Temperature*Frasco)+
  labs(x="day", subtitle =
        "replicates (A-B-C) by temperature", y= "Survival (%)")
Grafico_frascos +
  scale_colour_manual(values =
```

```
c("blue", "orange", "red"),
labels=c("12°C", "15°C", "17°C"))
```



## 2.1 table with statistics for LT50 and Kaplan-meier Analysis

```
#Estadísticos básicos (SD, SE, CI, MEAN)
LT50_summary<- summarySE(Mort, measurevar="bino",
                          groupvars=c("Temperatura", "Dia"))
print(LT50_summary)
```

##	Temperatura	Dia	N	bino	sd	se	ci
## 1	12	1	3	1.000000000	0.000000000	0.000000000	0.000000000
## 2	12	2	3	0.976666667	0.005773503	0.003333333	0.01434218
## 3	12	3	3	0.500000000	0.115325626	0.066583281	0.28648474
## 4	12	4	3	0.306666667	0.210792157	0.121700908	0.52363675
## 5	12	5	3	0.296666667	0.202319879	0.116809436	0.50259044
## 6	12	6	3	0.296666667	0.202319879	0.116809436	0.50259044
## 7	12	7	3	0.296666667	0.202319879	0.116809436	0.50259044
## 8	12	8	3	0.276666667	0.176162803	0.101707642	0.43761266
## 9	12	9	3	0.240000000	0.165227116	0.095393920	0.41044691
## 10	12	10	3	0.050000000	0.043588989	0.025166115	0.10828105
## 11	12	11	3	0.000000000	0.000000000	0.000000000	0.000000000
## 12	15	1	3	1.000000000	0.000000000	0.000000000	0.000000000

```
## 13      15      2 3 1.000000000 0.000000000 0.000000000 0.000000000
## 14      15      3 3 0.920000000 0.017320508 0.010000000 0.04302653
## 15      15      4 3 0.883333333 0.028867513 0.016666667 0.07171088
## 16      15      5 3 0.826666667 0.025166115 0.014529663 0.06251609
## 17      15      6 3 0.606666667 0.072341781 0.041766547 0.17970695
## 18      15      7 3 0.376666667 0.081445278 0.047022453 0.20232129
## 19      15      8 3 0.216666667 0.075055535 0.043333333 0.18644828
## 20      15      9 3 0.123333333 0.035118846 0.020275875 0.08724005
## 21      15     10 3 0.033333333 0.025166115 0.014529663 0.06251609
## 22      15     11 3 0.000000000 0.000000000 0.000000000 0.00000000
## 23      17      1 3 0.983333333 0.028867513 0.016666667 0.07171088
## 24      17      2 3 0.953333333 0.037859389 0.021858128 0.09404794
## 25      17      3 3 0.913333333 0.015275252 0.008819171 0.03794583
## 26      17      4 3 0.886666667 0.030550505 0.017638342 0.07589166
## 27      17      5 3 0.846666667 0.045092498 0.026034166 0.11201597
## 28      17      6 3 0.580000000 0.052915026 0.030550505 0.13144821
## 29      17      7 3 0.230000000 0.108166538 0.062449980 0.26870058
## 30      17      8 3 0.020000000 0.010000000 0.005773503 0.02484138
## 31      17      9 3 0.003333333 0.005773503 0.003333333 0.01434218
## 32      17     10 3 0.000000000 0.000000000 0.000000000 0.00000000
```

### 3. GLM analysis and LT50% plot.

```
#separation of the table by temp
mort_12<-LT50_summary[1:11, ]
mort_15<-LT50_summary[12:22, ]
mort_17<-LT50_summary[23:32,]

#GLM analysis for each temp

model1<-glm(data=mort_12, bino ~ Dia, family=quasibinomial())
summary(model1)

##
## Call:
## glm(formula = bino ~ Dia, family = quasibinomial(), data = mort_12)
##
## Deviance Residuals:
##      Min       1Q   Median       3Q      Max
## -0.5179  -0.2999  -0.0738   0.3400   0.5964
##
## Coefficients:
##              Estimate Std. Error t value Pr(>|t|)
## (Intercept)    2.1710     0.6380   3.403 0.00784 **
## Dia           -0.4791     0.1117  -4.289 0.00202 **
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
##
## (Dispersion parameter for quasibinomial family taken to be 0.1436587)
##
##      Null deviance: 5.4993  on 10  degrees of freedom
## Residual deviance: 1.5399  on  9  degrees of freedom
```

```
## AIC: NA
##
## Number of Fisher Scoring iterations: 5
```

```
model2<-glm(data=mort_15, bino ~ Dia, family=quasibinomial())
summary(model2)
```

```
##
## Call:
## glm(formula = bino ~ Dia, family = quasibinomial(), data = mort_15)
##
## Deviance Residuals:
##      Min       1Q   Median       3Q      Max
## -0.185582  -0.053923  -0.005199   0.088830   0.186640
##
## Coefficients:
##              Estimate Std. Error t value Pr(>|t|)
## (Intercept)  5.84038    0.37027   15.77 7.28e-08 ***
## Dia         -0.89940    0.05508  -16.33 5.39e-08 ***
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
##
## (Dispersion parameter for quasibinomial family taken to be 0.01151942)
##
##      Null deviance: 8.21286  on 10  degrees of freedom
## Residual deviance: 0.13672  on  9  degrees of freedom
## AIC: NA
##
## Number of Fisher Scoring iterations: 6
```

```
model3<-glm(data=mort_17, bino ~ Dia, family=quasibinomial())
summary(model3)
```

```
##
## Call:
## glm(formula = bino ~ Dia, family = quasibinomial(), data = mort_17)
##
## Deviance Residuals:
##      Min       1Q   Median       3Q      Max
## -0.287545  -0.221565  -0.146333  -0.006668   0.263467
##
## Coefficients:
##              Estimate Std. Error t value Pr(>|t|)
## (Intercept)  6.6331    0.9512    6.974 0.000116 ***
## Dia         -1.1205    0.1559   -7.186 9.37e-05 ***
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
##
## (Dispersion parameter for quasibinomial family taken to be 0.05103717)
##
##      Null deviance: 8.41356  on 9  degrees of freedom
## Residual deviance: 0.38809  on 8  degrees of freedom
## AIC: NA
```

```
##
## Number of Fisher Scoring iterations: 6
```

```
#Dose.p function to find moment at which 50% of the population has
#died based in the glm models made above
```

```
print(dose.p(model1, p = 0.5))
```

```
##           Dose      SE
## p = 0.5: 4.531844 0.6121778
```

```
print(dose.p(model2, p = 0.5))
```

```
##           Dose      SE
## p = 0.5: 6.493628 0.11405
```

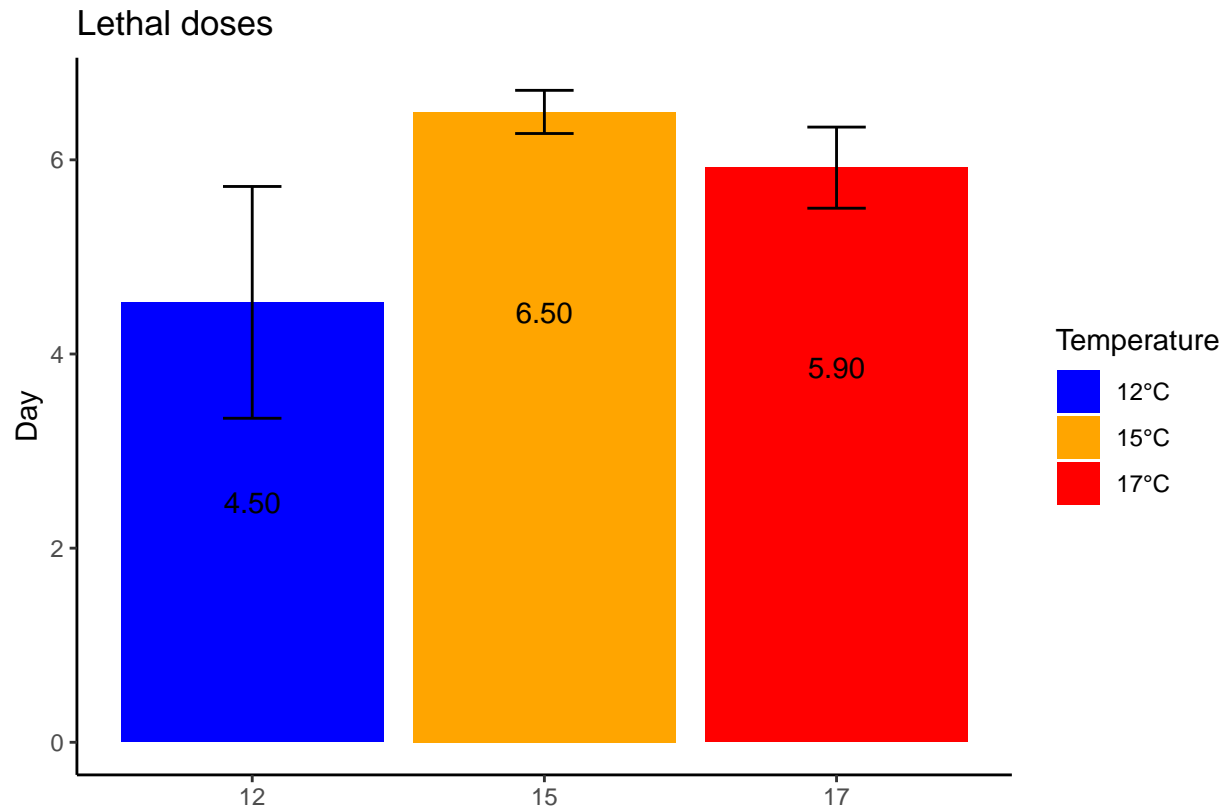
```
print(dose.p(model3, p = 0.5))
```

```
##           Dose      SE
## p = 0.5: 5.919705 0.2142654
```

```
# LT50% plot with values given by dose.p
```

```
lt50<-c(4.531844, 6.493628, 5.919705)
CI<-c(0.6121778*1.95, 0.11405*1.95, 0.2142654*1.95)
temp<-c(12, 15,17)
data_final<-data.frame(lt50, CI, temp)
data_final$temp<-as.factor(data_final$temp)

lt50_plot<-ggplot(data_final, aes(x=temp, y=lt50, fill=temp))+
  geom_bar(stat="identity") + theme_classic() +
  labs(title = "Lethal doses",
       x="", y="Day", fill= "Temperature") +
  scale_fill_manual(values =c("blue", "orange", "red"),
                    labels=c("12°C", "15°C", "17°C"))
lt50_plot + geom_errorbar(aes(ymin=lt50-CI, ymax=lt50+CI),
                        width=.2, position=position_dodge(0.05)) +
  geom_text(aes(label=c("4.50", "6.50", "5.90")), position =
            position_dodge(0.9), vjust = 10)
```



### 3.1 Kaplan-Meier Analysis

```
#Vector for creating correct data table
repV<- Mort$Muertas
repV[repV=="0"]<-1

#Datatable KAP for analysis
KAP<-Mort %>%
  mutate(count = repV) %>%
  uncount(count)

# Adding Status column (binary in which 1=no death; 2=death on that day)
KAP$STATUS <- ifelse (KAP$Muertas==0, 1, 2)
survObj <- Surv(time=KAP$Dia, event=KAP$STATUS==2)

#Kaplan-Meier analysis
sfit <- survfit(Surv(Dia, STATUS)~Temperatura, data=KAP)

#Ploting Kaplan-meier results
ggsurvplot(sfit, conf.int=TRUE, pval=TRUE, risk.table=FALSE,
  legend.labs=c("12°C", "15°C","17°C"),
  legend.title="Temperature",
  palette=(c("blue", "orange", "red")),
```

```
title="Kaplan-Meier Curve for Zoea 1 Survival to Temperature",  
risk.table.height=.15, ylab="Survival (%)", xlab="Day")
```

## Kaplan–Meier Curve for Zoea 1 Survival to Temperature

