##Install Packages  
library(tidyverse)  
library(ggplot2)  
library(nls.multstart)##Prepare and insert file here  
tpcs <- read.csv("~/Desktop/filename.csv")tpcs <- tpcs %>%  
  filter(Final>=1) %>%  
  mutate(r = log(Final/Initial)) %>%  
  mutate(r\_scale=10,  
         log\_r=log(r+r\_scale)) %>%  
  ungroup##Display data  
ggplot(tpcs, aes(Temp, log\_r)) +  
  geom\_point() +  
  theme\_bw(base\_size = 12) +  
  labs(x = 'Temperature (ºC)',  
       y = 'Metabolic rate',  
       title = 'Respiration across temperatures')TPC\_fits <- tpcs %>%  
  group\_by(Clone) %>%  
  do(TPC\_fit = nls\_multstart(log\_r ~ a + (E\_a/(8.6\*10^-5))\*(1/298.15-1/(Temp+273.15)) - log(1+exp((E\_d/(8.6\*10^-5))\*(1/Th-1/(Temp+273.15)))),  
                             data = .,  
                             iter = 500,  
                             start\_lower = c(a=-10, E\_a=0.1, E\_d=0.5, Th=285),  
                             start\_upper = c(a=10, E\_a=4, E\_d=10, Th=330),  
                             supp\_errors = 'Y',  
                             na.action = na.omit,  
                             lower = c(a=-10, E\_a=0, E\_d=0, Th=0))) %>%  
  rowwise() %>%  
  mutate(a=coef(TPC\_fit)[[1]],  
         E\_a=coef(TPC\_fit)[[2]],  
         E\_d=coef(TPC\_fit)[[3]],  
         Th=coef(TPC\_fit)[[4]],  
         T\_opt=E\_d\*Th/(E\_d+8.6e-5\*Th\*log(E\_d/E\_a-1)))pars <- dplyr::select(TPC\_fits, -TPC\_fit)TPC\_eqn<-function(a, E\_a, E\_d, Th, Temperature, r\_scale){exp(a + (E\_a/(8.6\*10^-5))\*(1/298.15-1/(Temperature+273.15)) - log(1+exp((E\_d/(8.6\*10^-5))\*(1/Th-1/(Temperature+273.15)))))-r\_scale}TPC\_predicted<-expand.grid(Temperature=seq(0, 50, length.out=500)) %>%  
  left\_join(dplyr::select(TPC\_fits, -TPC\_fit)) %>%  
  left\_join(distinct(dplyr::select(tpcs, r\_scale))) %>%  
  mutate(r=TPC\_eqn(a, E\_a, E\_d, Th, Temperature, r\_scale)) #%>%  
#filter(r>=-1)TPC\_summary\_spread<-TPC\_predicted %>%  
  mutate(CT\_min=ifelse(lag(r)<0 & r>0, Temperature, NA),  
         CT\_max=ifelse(lag(r)>0 & r<0, Temperature, NA),  
         r\_peak=max(r)) %>%  
  filter(![is.na](http://is.na/)(CT\_min) | ![is.na](http://is.na/)(CT\_max)) %>%  
  dplyr::select(CT\_min, CT\_max, r\_peak) %>%  
  ungroup() %>%  
  gather(param, param\_val) %>%  
  drop\_na %>%  
  distinct %>%  
  spread(param, param\_val) %>%  
  left\_join(dplyr::select(TPC\_fits, E\_a, E\_d, T\_opt)) %>%  
  mutate(T\_opt=T\_opt-273.15, T\_range=CT\_max-CT\_min, TPC\_asymmetry=abs((T\_opt-CT\_min)-(CT\_max-T\_opt))) %>%  
  arrange(r\_peak)# Plot TPCg\_1<-  
  ggplot()+  
  geom\_line(data=TPC\_predicted, aes(Temperature, r), size=1)+  
  labs(x="Temperature (C)", y=Intrinsic~growth~rate~(r)~(cells~cell^-1~d^-1)) +  
  theme(plot.background=element\_blank(), panel.background=element\_blank(),#panel.grid=element\_blank(),  
        panel.border=element\_rect(color = "black", size=1, fill=NA),  
        axis.text=element\_text(size=14), axis.title=element\_text(size=16),  
        axis.title.x.top=element\_blank(), axis.text.x.top=element\_blank(),  
        axis.ticks.length.x.top=unit(0, "cm"),  
        axis.ticks.length=unit(-0.15, "cm"),  
        plot.margin=unit(c(0,0,0,1), "cm"),  
        aspect.ratio=0.55,  
        legend.key=element\_blank(),  
        legend.position="right",  
        legend.text=element\_text(size=12)  
  )