Owen et al. - Figure 5

This script will reproduce Figure 5 from *Owen et al. - Sample size planning for insect critical thermal limits studies.*

Load required packages

```
if (!require("pacman")) install.packages("pacman")
pacman::p_load(tidyverse,
               tidyr,
               readr,
               data.table,
               viridis,
               here)

# Load ThermalSampleR package
# To download on first use, remove # from line below
# devtools::install_github("CJMuS/ThermalSampleR")
library(ThermalSampleR)
```

Set ggplot theme (makes nice plots)

```
theme_set(theme_classic() +
            theme(panel.border = element_rect(colour = "black", fill = NA),
                  axis.text = element_text(colour = "black"),
                  axis.title.x = element_text(margin = unit(c(2, 0, 0, 0), "mm")),
                  axis.title.y = element_text(margin = unit(c(0, 4, 0, 0), "mm")),
                  legend.position = "none"))
```

Load raw thermal limits data

```
# Also, load raw data (without CI's)
raw_data <- readr::read_csv(here::here("./data_clean/ct_min_data_clean.csv"))

# Make insect_sp column into a factor
raw_data <- raw_data %>%
  dplyr::mutate(col = as.factor(insect_sp))
```

Perform analysis

Here, let's subset the data to only the first 20 individuals tested for False Codling Moth (*Thaumatotibia leucotreta*) (FCM) fed on a standard versus trehalose diet. The aim of these experiments was to determine

whether trehalose, an anti-freeze compound, lowered the CTmin of FCM. Lowering the CTmin of FCM would be beneficial for the management of FCM (a crop pest). Thereafter, we perform bootstrap re-sampling of the data, and calculate summary statistics to evaluate whether testing additional individuals is required for this experiment.In this study, we may be interested in whether (1) diet treament had an effect on FCM CTmin (i.e. do FCM fed on a trehalose diet have lower CTmin's than FCM fed on a standard artificial diet?), and (2) if so, what is the magnitude (effect size) of the diet treatment (i.e. by how much do the CTmin's differ between standard vs trehalose diets?)

```
# Subset data
raw_data_ex <- raw_data %>%
  dplyr::filter(insect_sp %in% c("Thaumatotibia leucotreta_std_diet_CO_30",
                                 "Thaumatotibia leucotreta_tre_diet_CO_30"))

# Let's assume we only had the first 20 samples (n = 20), for each species
raw_data_ex <- raw_data_ex %>%
  dplyr::filter(between(sample_size, 1, 20))

# Check subset worked (n = 20 per species)
raw_data_ex %>% dplyr::count(insect_sp)
```

```
## # A tibble: 2 x 2
##   insect_sp                                    n
## * <chr>                                    <int>
## 1 Thaumatotibia leucotreta_std_diet_CO_30    20
## 2 Thaumatotibia leucotreta_tre_diet_CO_30    20
```

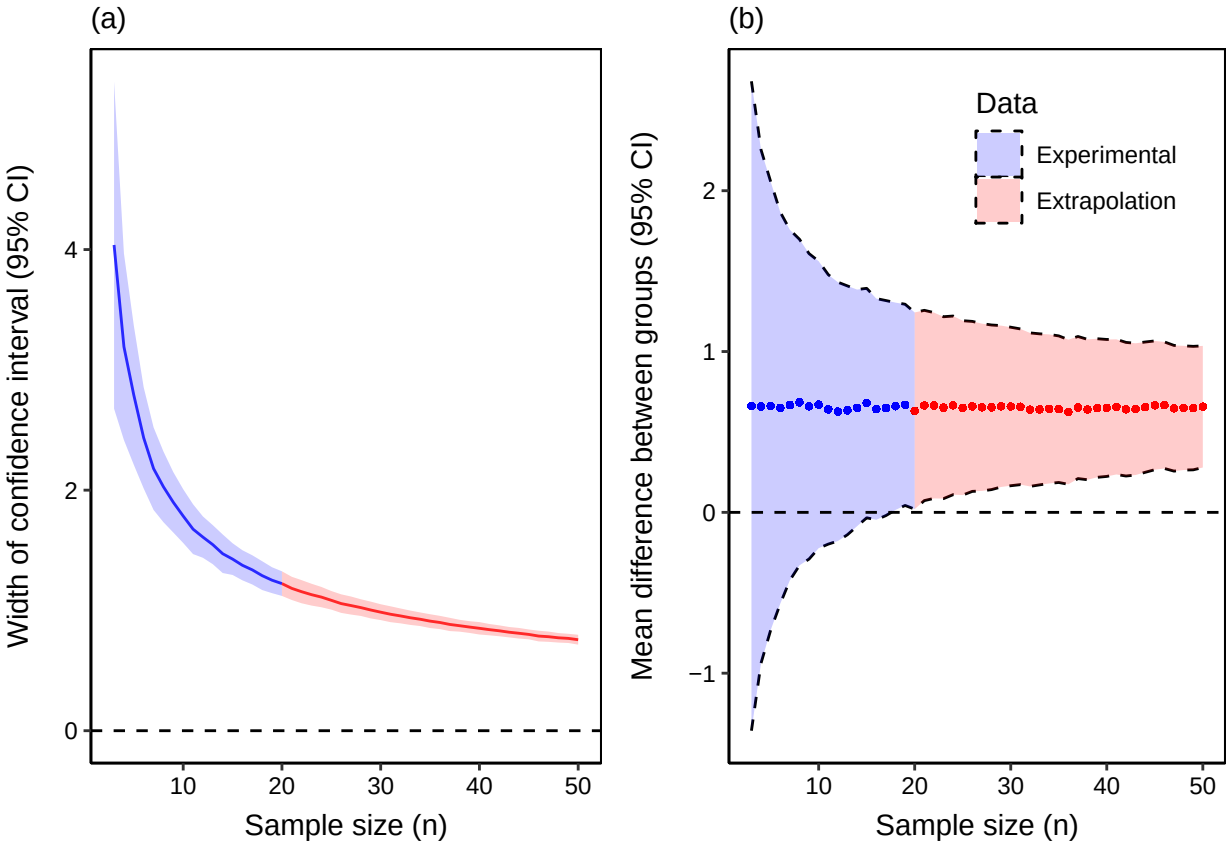
Use `ThermalSampleR` to evaluate sample size required

```
# Set reproducible seed
set.seed(2012)

# Perform bootstrap re-sampling
sims_15 <- ThermalSampleR::boot_two(
  # Data frame containing raw thermal limits data
  data = raw_data_ex,
  # Column name containing species ID's
  groups_col = col,
  # Name of first species
  group1 = "Thaumatotibia leucotreta_std_diet_CO_30",
  # Name of second species
  group2 = "Thaumatotibia leucotreta_tre_diet_CO_30",
  # Column name of thermal limits data
  response= ct_val,
  # Sample size to extrapolate towards
  n_max = 50,
  # Number of bootstrap iterations
  iter = 499)
```

Make figure

```
# Plot bootstrap samples
fig6 <- ThermalSampleR::plot_two_groups(
  # Data frame containing bootstrap re-samples
  x = sims_15,
  # Minimum number of samples to evaluate
  n_min = 3,
  # How many individuals were tested?
  n_max = 20,
  legend.position = c(0.7, 0.85))
fig6
```



Save figure to PC

```
# Save figure to file
ggplot2::ggsave(here::here("./figures/figure_5.png"),
                width = 8,
                height = 5)
```