

# Antidepressant effects of esketamine in adolescents with major depressive disorder and suicidal ideation: a case series

## Reproducible analysis report

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## 1 Note

This R Markdown file will exactly reproduce our data tidying, transformation, analysis and graphing processes in the R software and produce an illustrative PDF file. Code-only scripts are also available in the Open Science Framework repository for this study, at <https://osf.io/2ujqn/>. The reproduction of this `markdown` file requires that the dataset available in the OSF repository is located in the same directory of this file, and that the user has the necessary dependencies to create a PDF using the `bookdown` package. Further information is available at the *package website*. Please refer to the repository for a detailed description of each variable contained herein. All files in the repository, including this one, are licensed under CC-BY-4.0.

To help with reproducibility, the `{renv}` R package was used. The `renv.lock` file and the package folder are also available and should be imported to the same location on your computer as this script. For further information on how to use `{renv}` visit the package website: <https://rstudio.github.io/renv/articles/renv.html>.

## 2 Load the required packages

```
# Required Packages
library(tidyverse)
library(haven)
library(lsr)
library(rstatix)
library(boot)
library(kableExtra)
library(patchwork)
library(RColorBrewer)
library(ggsci)
library(car)
library(purrr)
library(broom)
```

## 3 Load the dataset

The first step is to load the dataset into R. You can load the dataset directly from the GitHub repository.

```
## Load from GitHub repository

urlRemote <- "https://raw.githubusercontent.com/"
pathGithub <- "brenosouzamarques/LANP_Esketamine_Adolescents/master/"
fileName <- "Dataset.csv"

df <- paste0(urlRemote, pathGithub, fileName) %>%
  read.csv()
```

## 4 Visualizing distribution of dependent variables

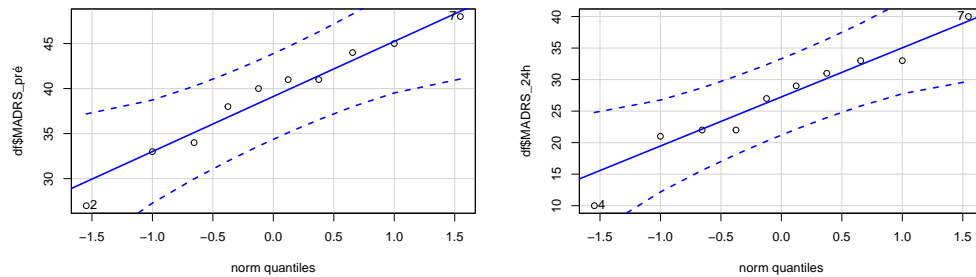
Q-Q Plots are used to assess the symmetry of dependent variables, that is, **MADRS total score** and **MADRS item 10** at baseline and 24-h post-infusion.

### 4.1 MADRS total score at baseline and 24h post-treatment

```
qqplot_madrs_pre <- car::qqPlot(df$MADRS_pré)
qqplot_madrs_post <- car::qqPlot(df$MADRS_24h)

print(qqplot_madrs_pre, qqplot_madrs_post)

## [1] 2 7
```

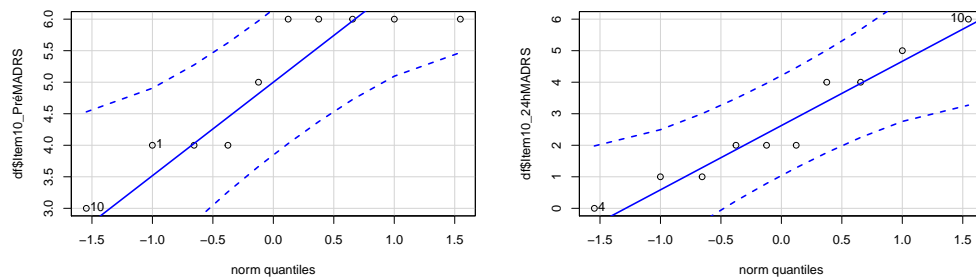


## 4.2 MADRS item 10 score at baseline and 24h post-treatment

```
qqplot_item10_pre <- car::qqPlot(df$Item10_PréMADRS)
qqplot_item10_post <- car::qqPlot(df$Item10_24hMADRS)

print(qqplot_item10_pre, qqplot_item10_post)
```

```
## [1] 10 1
```



## 5 Descriptives

Total depressive symptoms and suicidal ideation scores are presented as means and standard deviations.

### 5.1 MADRS total scores

```
madsr_total <- c(df$MADRS_pré, df$MADRS_24h)
time <- rep(c("baseline", "post"), each = 10)
data_madsr_total <- tibble(time, madsr_total)
kbl(data_madsr_total %>% group_by(time) %>%
  summarise(
    Mean = mean(madsr_total),
    "Std dev" = sd(madsr_total),
    Maximum = max(madsr_total),
    Minimum = min(madsr_total)),
  caption = "MADRS total score",
  booktabs = T) %>% kable_styling(latex_options = c("hold_position"))
```

Table 1: MADRS total score

time	Mean	Std dev	Maximum	Minimum
baseline	39.1	6.297266	48	27
post	26.8	8.403703	40	10

## 5.2 MADRS item 10 scores

```

item_10_total <- c(df$Item10_PréMADRS, df$Item10_24hMADRS)
time <- rep(c("baseline", "post"), each = 10)
data_madrs_total <- tibble(time, item_10_total)
kbl(
  data_madrs_total %>%
  group_by(time) %>%
  summarise(
    Mean = mean(item_10_total),
    "Std dev" = sd(item_10_total),
    Maximum = max(item_10_total),
    Minimum = min(item_10_total),
    caption = "MADRS item 10 score",
    booktabs = T) %>% kable_styling(latex_options = c("hold_position"))

```

Table 2: MADRS item 10 score

time	Mean	Std dev	Maximum	Minimum
baseline	5.0	1.154700	6	3
post	2.7	1.946507	6	0

# 6 Hypothesis tests

## 6.1 T-tests

We first aim to determine if the difference between pre-and post-treatment outcomes is significant. We test this hypothesis with a paired samples t-test for each outcome, as follows.

```
### T-test - MADRS Total Scores
```

```

ttest_total_madrs <- t.test(x = df$MADRS_pré,
  y = df$MADRS_24h,
  paired = T)

ttest_item10_madrs <- t.test(x = df$Item10_PréMADRS,
  y = df$Item10_24hMADRS,
  paired = T)

table_madrs <- map_df(list(ttest_total_madrs, ttest_item10_madrs), tidy)

table_madrs$'Dependent Variable' <- c("MADRS total score",

```

```

                                "MADRS item 10")

kbl(table_madrs[c("Dependent Variable",
                  "estimate",
                  "statistic",
                  "p.value")],
     caption = "T-tests",
     booktabs = T) %>% kable_styling(latex_options = c("hold_position"))

```

Table 3: T-tests

Dependent Variable	estimate	statistic	p.value
MADRS total score	12.3	4.221349	0.0022350
MADRS item 10	2.3	3.146129	0.0118109

## 6.2 Effect sizes

Effect sizes allow further interpretation of how significant are these changes from baseline to 24 hours post-ketamine treatment.

```

effectsize_madrs <- lsr::cohensD(df$MADRS_pré,
                                df$MADRS_24h,
                                method = "paired")

effectsize_item10 <- lsr::cohensD(df$Item10_PréMADRS,
                                  df$Item10_24hMADRS,
                                  method = "paired")

kbl(tribble(~Outcome, ~"Effect Size (Cohen's D)",
            "MADRS total score", effectsize_madrs,
            "MADRS item 10", effectsize_item10),
     caption = "Effect sizes",
     booktabs = T) %>% kable_styling(latex_options = c("hold_position"))

```

Table 4: Effect sizes

Outcome	Effect Size (Cohen's D)
MADRS total score	1.3349078
MADRS item 10	0.9948934

## 7 Bootstrapped 95% confidence intervals

### 7.1 Create a bootstrap function

This functions creates a main element, the `data_dens` data frame. It stores the bootstrapped samples of means as they are calculated using the bias-corrected and accelerated (Bca) method. These are later used to plot density graphs that compare bootstrapped distributions of pre-and post-treatment symptoms.

```

boot_funs <- function(df, f1, f2){
  a <- df[[f1]]

```

```

b <- df[[f2]]
boot_a <- boot(a,
  function(x, j) mean(x[j], na.rm = T), #Boot baseline variable
  R = 10000)
boot_b <- boot(b,
  function(x, j) mean(x[j], na.rm = T), #Boot 7 days variable
  R = 10000)
boot_ci_a <- boot.ci(boot.out = boot_a, #Calculate baseline CI
  conf = 0.95,
  type = "bca")
boot_ci_b <- boot.ci(boot.out = boot_b, #Calculate post-treatment CI
  conf = 0.95,
  type = "bca")
print(boot_ci_a) # Return baseline bootstrapped CI
print(boot_ci_b) # Return post-treatment bootstrapped CI

## Code to extract bootstrapped sample means
densitiy_a <- boot_a$t
densitiy_b <- boot_b$t
dens <- c(densitiy_a, densitiy_b)
time <- rep(c("Baseline", "Post-Treatment"), each = 10000)
data_dens <-< data.frame(dens, time)
data_dens$time <- factor(data_dens$time)

```

```

}

```

## 7.2 Run bootstraps and create ggplot graph objects

### 7.2.1 Bootstrap - MADRS total score

```
### Bootstrap CI - MADRS total score
boot_funs(df, "MADRS_pré", "MADRS_24h")

## BOOTSTRAP CONFIDENCE INTERVAL CALCULATIONS
## Based on 10000 bootstrap replicates
##
## CALL :
## boot.ci(boot.out = boot_a, conf = 0.95, type = "bca")
##
## Intervals :
## Level      BCa
## 95%   (34.8, 42.3 )
## Calculations and Intervals on Original Scale
## BOOTSTRAP CONFIDENCE INTERVAL CALCULATIONS
## Based on 10000 bootstrap replicates
##
## CALL :
## boot.ci(boot.out = boot_b, conf = 0.95, type = "bca")
##
## Intervals :
## Level      BCa
## 95%   (21.2, 31.1 )
## Calculations and Intervals on Original Scale

plot_madrs <- ggplot(data_dens)+
  geom_density(aes(x = dens, fill = time),
               color = "white",
               alpha = 0.9)+
  labs(x = "Total MADRS scores")+
  scale_fill_brewer(name = "Time of assessment:",
                   palette = "Dark2")+
  theme_classic()+
  theme(text = element_text(family = "serif"),
        axis.text.x = element_text(size = 14),
        axis.title.x = element_text(size = 14),
        axis.text.y = element_text(size = 14),
        axis.title.y = element_text(size = 14),
        legend.text = element_text(size = 13),
        legend.title = element_text(size = 13)) ##ggplot2 graph
```

## 7.2.2 Bootstrap - MADRS item 10

```
### Bootstrap CI - MADRS item 10

boot_funs(df, "Item10_PréMADRS", "Item10_24hMADRS")

## BOOTSTRAP CONFIDENCE INTERVAL CALCULATIONS
## Based on 10000 bootstrap replicates
##
## CALL :
## boot.ci(boot.out = boot_a, conf = 0.95, type = "bca")
##
## Intervals :
## Level      BCa
## 95%      ( 4.2,  5.5 )
## Calculations and Intervals on Original Scale
## BOOTSTRAP CONFIDENCE INTERVAL CALCULATIONS
## Based on 10000 bootstrap replicates
##
## CALL :
## boot.ci(boot.out = boot_b, conf = 0.95, type = "bca")
##
## Intervals :
## Level      BCa
## 95%      ( 1.6,  3.8 )
## Calculations and Intervals on Original Scale

plot_madrs10 <- ggplot(data_dens)+
  geom_density(aes(x = dens, fill = factor(time)),
               color = "white",
               alpha = 0.9)+
  labs(x = "Item 10 MADRS scores",
       y = element_blank())+
  scale_fill_brewer(name = "Time of assessment:",
                    palette = "Dark2")+
  theme_classic()+
  theme(text = element_text(family = "serif"),
        axis.text.x = element_text(size = 14),
        axis.title.x = element_text(size = 14),
        axis.text.y = element_text(size = 14),
        axis.title.y = element_text(size = 14),
        legend.text = element_text(size = 13),
        legend.title = element_text(size = 13)) ##ggplot2 graph
```



## 8 Graphics

This first part creates an annotated plot using the `ggplot2` density plots for the bootstrapped sample means and the `patchwork` package.

```
## Density plots of bootstrapped sample means

## Creates patchwork object
graficos <- plot_madrs + plot_madrs10 &
  theme(legend.position = "bottom",
        text = element_text(family = "serif"),
        title = element_text(size = 12),
        legend.title = element_text(size = 12))

## Creates guided patchwork object
graficos_f <- graficos + plot_layout(guides = "collect")

## Edits and annotates patchwork object
plota <- graficos_f + plot_annotation(
  title = 'Supp. Fig. 1 - Bootstraped sample means for total and item 10 MADRS scores',
  subtitle = 'Density plots of results at baseline and 24 hours post-treatment')

## Saves plot in high resolution
ggsave("density_plot.png", plota, width=25, height=15, units="cm", dpi=450)
```

This next part uses a long formatted dataset to compare pre-and post-infusion results for each patient.

```
## Create dataset for total MADRS scores
madrs_total <- c(df$MADRS_pré, df$MADRS_24h)
time <- rep(c("baseline", "post"), each = 10)
id <- rep(1:10, 2)
id <- factor(id)
madrs_data_plot <- tibble(id, time, madrs_total)

## plot total MADRS scores
madrs_plot <- madrs_data_plot %>% ggplot()+
  geom_boxplot(aes(x = time, y = madrs_total),
    width = .3,
    alpha = .3,
    color = "grey")+
  scale_y_continuous(breaks = seq(from = 10,
    to = 50,
    by = 5))+
  scale_x_discrete(labels = c("Baseline", "24 hours post-infusion"))+
  geom_point(aes(x = time, y = madrs_total,
    color = id,
    group = id),
    size = 1.5)+
  geom_line(aes(x = time, y = madrs_total,
    color = id,
    group = id,
    alpha = .4),
    size = 1.5)+
  scale_color_npg()+
  theme_classic()+
```

```

labs(x = "Time of assessment",
     y = "Total MADRS scores")+
theme(axis.title.x = element_text(size = 14),
      axis.text.x = element_text(size = 13),
      axis.title.y = element_text(size = 15),
      axis.text.y = element_text(size = 13),
      legend.position = "none")

## Create dataset for MADRS Item 10

si_pre <- df$Item10_PréMADRS
si_post <- df$Item10_24hMADRS

madr_item10 <- c(si_pre, si_post)
time <- rep(c("baseline", "post"), each = 10)
id <- rep(1:10, 2)
id <- factor(id)
madr_item10_plot <- tibble(id, time, madrs_item10)

item10_plot <- madrs_item10_plot %>% ggplot()+
  geom_boxplot(aes(x = time, y = madrs_item10),
              width = .3,
              alpha = .3,
              color = "grey")+
  scale_y_continuous(breaks = c(0, 1, 2, 3, 4, 5, 6))+
  scale_x_discrete(labels = c("Baseline", "24 hours post-infusion"))+
  geom_point(aes(x = time, y = madrs_item10,
                 color = id,
                 group = id),
             size = 1.5,
             position = position_dodge(0.08))+
  geom_line(aes(x = time, y = madrs_item10,
                color = id,
                group = id,
                alpha = .4),
            size = 1.5,
            position = position_dodge(0.08))+
  scale_color_npg()+
  theme_classic()+
  labs(x = "Time of assessment",
       y = "MADRS item 10 scores")+
  theme(axis.title.x = element_text(size = 14),
        axis.text.x = element_text(size = 13),
        axis.title.y = element_text(size = 15),
        axis.text.y = element_text(size = 13),
        legend.position = "none",
        text = element_text(family = "serif"))

```

## 8.1 Join and save plots

```

## Creates patchwork object
graficos_i10 <- madrs_plot + item10_plot &
  theme(text = element_text(family = "serif"),

```

```

        title = element_text(size = 16),
        legend.title = element_text(size = 14))

## Edits and annotates patchwork object
graficos_i10 <- graficos_i10 + plot_annotation(
  title = 'Total and item 10 MADRS scores at baseline and 24h post-treatment',
  subtitle = 'Scores are given for each patient')

## Saves plot in high resolution
ggsave("MADRS_plot.png", graficos_i10, width=25, height=15, units="cm", dpi=500)

```