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 on reproducibility

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

```r
# 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.

```r
set.seed(2509)
## 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
```
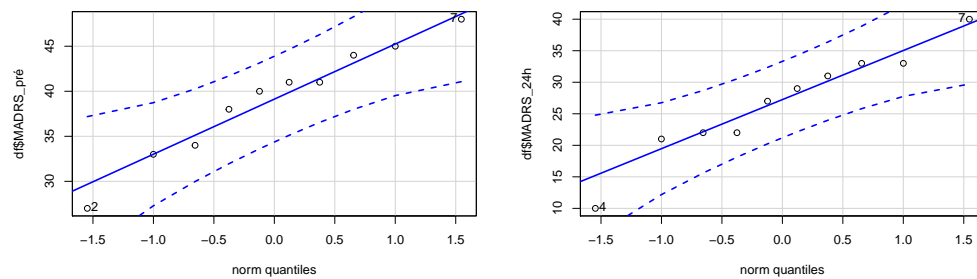
4.2 MADRS item 10 score at baseline and 24h post-treatment

```
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.
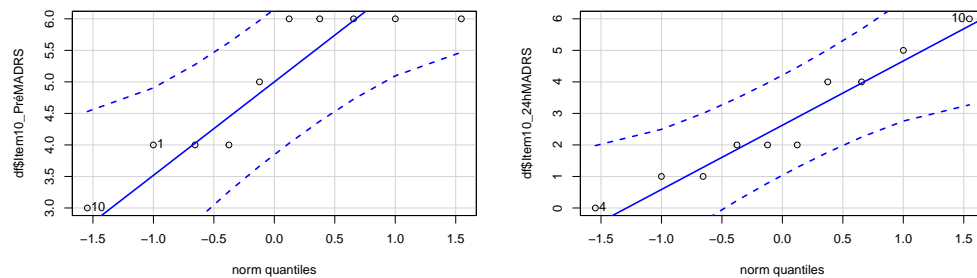
5.1 MADRS total scores

```r
madrs_total <- c(df$MADRS_pré, df$MADRS_24h)
time <- rep(c("baseline", "post"), each = 10)
data_madrs_total <- tibble(time, madrs_total)
kbl(data_madrs_total %>% group_by(time) %>%
      summarise(
        Mean = mean(madrs_total),
        "Std dev" = sd(madrs_total),
        Maximum = max(madrs_total),
        Minimum = min(madrs_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

```r
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),
    Minimun = 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	Minimun
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.

```r
### 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",
                  "conf.low",
                  "conf.high",
                  "p.value")],
    caption = "T-tests",
    booktabs = T) %>% kable_styling(latex_options = c("hold_position"))
```

Table 3: T-tests

Dependent Variable	estimate	statistic	conf.low	conf.high	p.value
MADRS total score	12.3	4.221349	5.708616	18.891384	0.0022350
MADRS item 10	2.3	3.146129	0.646234	3.953766	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.

```r
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.

```r
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%    (34.9, 42.4 )
## 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%   ( 4.1,  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%   ( 1.5,  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

8.1 Density plots of bootstrapped sample means

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

```r
## 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)
```

8.2 Main plots: pre-and post-treatment effects for depressive symptoms and suicidal ideation

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

```r
## 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,
                                            face = "bold"),
              axis.text.x = element_text(size = 13),
              axis.title.y = element_text(size = 15,
                                            face = "bold"),
              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

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

item10_plot <- madrs_item_plot %>% ggplot()+
```

```r
        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,
                                          face = "bold"),
              axis.text.x = element_text(size = 13),
              axis.title.y = element_text(size = 15,
                                          face = "bold"),
              axis.text.y = element_text(size = 13),
              legend.position = "none",
              text = element_text(family = "serif"))
```

8.2.1 Join and save plots!

```r
## 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') &
  theme()


## Saves plot in high resolution
ggsave("MADRS_plot.png", graficos_i10, width=20, height=10, units="cm", dpi=450)
ggsave("MADRS_plot.tiff", graficos_i10, width=20, height=10, units="cm", dpi=450)
```