

# *Piper auritum* in rural communities from Yucatán, México

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## 1. Configuración de la sesión y carga de paqueterías

```
## environment
set.seed(123999)
options(scipen = 10000, digits = 3, ggrepel.max.overlaps = Inf)
library(tidyverse, warn.conflicts = F)
library(patchwork)
library(vegan);
library(FactoMineR)
```

## 2. Cargando sesiones de R

```
base::load(file = 'rdbases/Figs_2-y-4_Usos-y-Manejos.RData')
base::load(file = 'rdbases/Fig.5-PCA-aceites-esenciales.RData')
source(file = 'scripts/plot_setts.R')
```

## 3. Figura 2: Índices de sutrop

```
# 1. Reading raw databae: excel data; check for
# additional details ----- Data were taken
# from the Excel database: check data out for any
# additional detail

sutrop <- xlsx::read.xlsx(file = "data/p_auritum_database.xlsx",
  sheetIndex = 3, startRow = 3)

sutrop %>%
  dplyr::as_tibble()

## Filter data and plot: Uso culinario
culinario <- sutrop %>%
  tidyr::pivot_longer(cols = c(-1:-2), names_to = "comunidad",
    values_to = "sutrop") %>%
```

```

dplyr::filter(Usos == "Culinario") %>%
dplyr::mutate(sutrop = dplyr::if_else(sutrop ==
  0, true = 0.01, false = sutrop)) %>%
ggplot(aes(x = comunidad, y = sutrop, fill = Tipo)) +
theme_bw(base_size = 16) + labs(y = "Índice de Sutrop\n",
x = element_blank()) + geom_bar(position = position_dodge(0.9),
stat = "identity", width = 0.8) + scale_fill_manual(name = element_blank(),
values = RColorBrewer::brewer.pal(n = 5, name = "Dark2"),
limits = c("vaporcitos", "con huevos", "con ibes",
  "Bebida", "cool"), labels = c("Vaporcitos",
  "+Huevos", "+Íbes", "Bebida", "Cool")) +
scale_y_continuous(expand = c(0, 0), limits = c(0,
1.05), breaks = seq(from = 0, to = 1, by = 0.1),
labels = c("0.0", "", 0.2, "", 0.4, "", 0.6,
  "", 0.8, "", 1.0)) + scale_x_discrete(name = element_blank(),
limits = c("Ebtun", "Ek.Balam", "Hunuku", "Pixoy",
  "Santa.Rita", "Uayma"), labels = c("Ebtún",
  "Ek Balam", "Hunuku", "Pixoy", "Santa Rita",
  "Uayma")) + facet_grid(cols = vars(factor(Usos,
levels = c("Culinario"), labels = c("Uso culinario")))) +
guides(fill = guide_legend(ncol = 3)) + plot_setts +
theme(legend.position = "bottom") + coord_flip() +
theme(panel.grid.major.x = element_line(colour = "gray20",
linetype = "dashed", linewidth = 0.1))

## Uso Medicinal: plots
medicinal <- sutrop %>%
tidyr::pivot_longer(cols = c(-1:-2), names_to = "comunidad",
values_to = "sutrop") %>%
dplyr::filter(Usos == "Medicinal") %>%
dplyr::mutate(sutrop = dplyr::if_else(sutrop ==
  0, true = 0.005, false = sutrop)) %>%
ggplot(aes(x = comunidad, y = sutrop, fill = Tipo)) +
theme_bw() + labs(y = "Índice de Sutrop\n", x = element_blank()) +
geom_bar(position = position_dodge(0.9), stat = "identity",
width = 0.8) + scale_fill_manual(name = element_blank(),
values = RColorBrewer::brewer.pal(n = 12, name = "Paired"),
limits = c("Colesterol", "Diabetes", "Dolor", "Fiebre",
  "Producir leche", "Gastritis ", "Hemorroides",
  "Nervios", "Pelagra", "Presión", "Purgante",
  "Tos"), labels = c("Colesterol", "Diabetes",
  "Dolor", "Fiebre", "Galactogogo", "Gastritis",
  "Hemorroides", "Nervios", "Pelagra", "Presión",
  "Purgante", "Tos")) + scale_y_continuous(expand = c(0,
0), limits = c(0, 0.31), breaks = seq(0, 0.3, 0.03),
labels = c("0.0", "", 0.06, "", 0.12, "", 0.18,
  "", 0.24, "", 0.3)) + scale_x_discrete(name = element_blank(),
limits = c("Ebtun", "Ek.Balam", "Hunuku", "Pixoy",
  "Santa.Rita", "Uayma"), labels = c("Ebtún",
  "Ek Balam", "Hunuku", "Pixoy", "Santa Rita",
  "Uayma")) + facet_grid(cols = vars(factor(Usos,
levels = c("Medicinal"), labels = c("Uso medicinal")))) +

```

```

guides(fill = guide_legend(ncol = 4)) + theme(legend.position = "bottom") +
coord_flip() + plot_setts + theme(panel.grid.major.x = element_line(colour = "gray20",
linetype = "dashed", linewidth = 0.1))

## Merge into one plot with patchwork:
figure2 <- (culinario + theme(axis.title.x = element_text(size = 14))) |
  (medicinal + theme(axis.text.y = element_blank(),
    axis.title.x = element_text(size = 14)))
figure2

## Save as a single plot
ggplot2::ggsave(filename = "Figures/Figure-2-Sutrop-Usos.png",
  dpi = 300, width = 14, height = 6)

```

#### 4. Figura 4: Manejos de *Piper auritum*

```

# MANEJOS Y USOS DE Piper auritum: Valores
# porcentuales -----

## 1. Leemos la base de datos, convertimos a
## formato largo, y posteriormente graficamos
data <- xlsx::read.xlsx(file = "data/p_auritum_database.xlsx",
  sheetIndex = 4, startRow = 3) %>%
  tidyr::pivot_longer(cols = c(-1:-2), names_to = "comunidad",
    values_to = "sutrop")

data %>%
  dplyr::as_tibble()

## Filter data and plot: Origen
origenes <- data %>%
  dplyr::filter(practice == "Origen") %>%
  dplyr::mutate(sutrop = dplyr::if_else(sutrop ==
    0, true = 0.2, false = sutrop)) %>%
  ggplot(aes(x = comunidad, y = sutrop, fill = type)) +
  theme_bw() + labs(y = "\nValores porcentuales (%) \n",
    x = element_blank()) + geom_bar(position = position_dodge(0.9),
    stat = "identity", width = 0.8) + scale_fill_manual(name = element_blank(),
    values = RColorBrewer::brewer.pal(n = 5, name = "Dark2"),
    limits = c("nacío_solo", "del_monte", "de_otro_huerto"),
    labels = c("Nació sola", "Monte", "Otro huerto")) +
  scale_y_continuous(expand = c(0, 0), limits = c(0,
    31), breaks = seq(0, 30, 3), labels = c("0.0",
    "", 6, "", 12, "", 18, "", 24, "", 30)) + scale_x_discrete(name = element_blank(),
    limits = c("Ebtun", "Ek_Balam", "Hunuku", "Pixoy",
    "Santa_Rita", "Uayma"), labels = c("Ebtún",
    "Ek Balam", "Hunuku", "Pixoy", "Santa Rita",
    "Uayma")) + facet_grid(cols = vars(factor(practice,
    levels = c("Origen"), labels = c("Origen")))) +
  guides(fill = guide_legend(nrow = 1)) + plot_setts +

```

```

theme(legend.position = "bottom", panel.grid.major.y = element_blank(),
      panel.grid.major.x = element_line(color = "gray40",
        linewidth = 0.1, linetype = "dashed")) +
coord_flip()

## Manejos: Plotting:
manejos <- data %>%
  dplyr::filter(practice == "Manejo") %>%
  dplyr::mutate(sutrop = dplyr::if_else(sutrop ==
    0, true = 0.005, false = sutrop)) %>%
  ggplot(aes(x = comunidad, y = sutrop, fill = type)) +
  theme_bw() + labs(y = "\nÍndice de Sutrop\n",
    x = element_blank()) + geom_bar(position = position_dodge(0.9),
    stat = "identity", width = 0.8) + scale_fill_manual(name = element_blank(),
    values = RColorBrewer::brewer.pal(n = 5, name = "Dark2"),
    limits = c("Riego", "Chapeo", "Poda"), labels = c("Riego",
      "Deshierbe", "Poda")) + scale_y_continuous(expand = c(0,
    0), limits = c(0, 1.1), breaks = seq(0, 1, 0.1),
    labels = c("0.0", "", "0.2", "", "0.4", "", "0.6", "",
      0.8, "", "1.0")) + scale_x_discrete(name = element_blank(),
    limits = c("Ebtun", "Ek_Balam", "Hunuku", "Pixoy",
      "Santa_Rita", "Uayma"), labels = c("Ebtún",
      "Ek Balam", "Hunuku", "Pixoy", "Santa Rita",
      "Uayma")) + facet_grid(cols = vars(factor(practice,
    levels = c("Manejo"), labels = c("Manejos")))) +
  guides(fill = guide_legend(nrow = 1)) + plot_setts +
  theme(legend.position = "bottom", panel.grid.major.y = element_blank(),
    panel.grid.major.x = element_line(color = "gray40",
      linewidth = 0.1, linetype = "dashed")) +
  coord_flip()

figure4 <- origenes | (manejos + theme(axis.text.y = element_blank()))
figure4 + plot_annotation(tag_levels = "A", tag_suffix = ")") &
  theme(plot.tag = element_text(size = 14, face = "bold"))

ggplot2::ggsave(file = "Figures/Fig.4-Sutrop.png",
  dpi = 300, device = "png", width = 10, height = 5)

```

## 5. Figura 5: Análisis de componentes principales - PCA de aceites esenciales

```

# 5.1. Loading database -----

# Load the raw database

data <- read.table(file = 'data/db_Fig.5_PCA_aceites_esenciales.tsv', row.names = 1, header = T, sep = '\t')

metadata <- data %>%
  tibble::rownames_to_column(var = "TRATAMIENTO") %>%
  select(1:4); head(metadata)

# 5.2. Transform data and determine the Principal Component Analysis -----

head(data)

```

```

oil.pca <- FactoMineR::PCA(data[,8:ncol(data)], ncp = 4)

# 5.2.1. Get individuals coordinates and merge with their metadata

sam.coords <- data.frame(oil.pca$ind$coord, check.names = F) %>%
  tibble::rownames_to_column(var = "TRATAMIENTO") %>%
  dplyr::inner_join(x = metadata, by = "TRATAMIENTO")
str(sam.coords)
sam.coords %>% tibble::as_tibble()

# 5.2.2. Get variables (essential oils) coordinates of the top 10 contributors for PC1 and PC2

topcontrib <- data.frame(oil.pca$var$contrib, check.names = F) %>%
  tibble::rownames_to_column(var = "oil") %>%
  dplyr::arrange(-Dim.1) %>%
  dplyr::filter(!grepl(pattern = "TR-", x = oil)) %>%
  filter(Dim.1 > 4); topcontrib

# 5.3.3. Get variables (essential oils), and ignore those with unknown names

responsevar <- data.frame(oil.pca$var$coord, check.rows = F, check.names = F) %>%
  tibble::rownames_to_column(var = "ResponseVar"); responsevar

# 5.3. Draw the Principal Component Analysis with ggplot2: PCA -----

p <-
ggplot() + theme_bw ()+
  labs (x = paste ("PC1 ", paste(base::round (oil.pca$eig[1,2], digits = 2), "%", sep = "")),
        y = paste ("PC2 ", paste(base::round (oil.pca$eig[2,2], digits = 2), "%", sep = ""))) +
  # adding vectors: variables
  geom_segment(data = responsevar,
    aes(x = 0, y = 0, xend = Dim.1*5, yend = Dim.2*5),
    arrow = arrow(length = unit(0.15, "cm")),
    color = "black", show.legend = NA) +
  ## adding vectors names: variable names in front of each vector
  ## Plot only the top 10 contributors per PC
  ggrepel::geom_label_repel(data = responsevar %>%
    dplyr::filter(!grepl(pattern = 'TR-', x = ResponseVar)) %>%
    dplyr::filter(ResponseVar %in% topcontrib$oil),
    aes(x=Dim.1*7, y=Dim.2*7, label = ResponseVar),
    segment.colour = NA, col = 'black', fill= "#FFFFFF",
    fontface = "italic", box.padding = 0.6, size = 5) +
  geom_point(data = sam.coords, aes (x = Dim.1, y = Dim.2, shape = SITIO, colour = CATEGORIAS), s
  stat_ellipse(data = sam.coords, aes(x=Dim.1, y=Dim.2, color = CATEGORIAS), type = "norm") +
  scale_color_manual(name = element_blank(), values = c("#E69F00", "#56B4E9")) +
  scale_shape_manual(name = element_blank(), values=c(15,19)) +
  scale_x_continuous(limits = c(-10,10), breaks = seq(from = -10, to = 10, by = 5)) +
  scale_y_continuous(limits = c(-10,15), breaks = seq(from = -10, to = 15, by = 5)) +
  geom_hline(yintercept=0, linetype="dashed", color = "black") +
  geom_vline (xintercept = 0, linetype="dashed", color = "black") +
  guides(fill = guide_legend(nrow = 2)) +
  theme(panel.grid = element_blank(),

```

```

legend.text = element_text(size = 12, color = "black"),
axis.text = element_text(size = 14, color = "black"),
axis.title = element_text(color = "black", size = 14),
legend.key = element_blank(),
## legend.position:legend.position = c(0.85,0.85)
legend.position = "top")

stats_data <- data.frame("Factor" = c("Sitio", "Categoría", "Interacción"),
"P_value" = c(0.018, 0.140, 0.450),
"R2" = c(0.080, 0.003, 0.110))

stats_data %>% tibble::as_tibble()

p + annotation_custom(tableGrob(stats_data, rows=NULL),
xmin=3, xmax=10, ymin=10, ymax=15)
ggplot2::ggsave(file = 'Figures/Fig.5.PCA-acei_esenciales.png', dpi = 300, device = "png", width = '

```

## 6. Análisis multivariado: perMANOVA de contenido de aceites esenciales

```

# 6. Multivariate analysis: perMANOVA
# -----

## perMANOVA: Using the filtered data
## sink (file =
## 'data/permanova-eoils.txt') # to
## save the output to a plain text file
## dist_essoils <- vegan::vegdist(x =
## data[,8:ncol(data)], method =
## 'euclidean') # determine the
## distance matrix
vegan::adonis2(formula = dist_essoils ~ SITIO *
  CATEGORIAS, data = data[, 1:7], parallel = T,
  permutations = 9999) # determine perMANOVA
# sink (file = NULL) # save the outpu

```