Appendix A

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R build: Geospatial 4.4.0

Analysis of Ace2 dataset from this study

Detailed data are available in Table 1 of the manuscript.

1. Load and Process Species Distribution Models (SDMs)

```
# Load MaxEnt SDMs
sdm_pipiens <- raster("../gis/culex_pipiens_meansuitability.nc")
sdm_quinque <- raster("../gis/culex_quinquefasciatus_meansuitability.nc")
# Threshold to binary
threshold <- 0.5
sdm_pipiens_bin <- sdm_pipiens >= threshold
sdm_quinque_bin <- sdm_quinque >= threshold
```

2. Convert SDMs to Polygons

```
# Raster to terra
sdm_pipiens_v <- terra::rast(sdm_pipiens_bin)
sdm_quinque_v <- terra::rast(sdm_quinque_bin)

# Raster to polygons
poly_pipiens <- terra::as.polygons(sdm_pipiens_v, dissolve = TRUE)
poly_quinque <- terra::as.polygons(sdm_quinque_v, dissolve = TRUE)

# Terra to sf
poly_pipiens_sf <- st_as_sf(poly_pipiens)
poly_quinque_sf <- st_as_sf(poly_quinque)

# Filter to presence only
names(poly_pipiens_sf)[1] <- "presence"
names(poly_quinque_sf)[1] <- "presence"
poly_pipiens_sf <- poly_pipiens_sf %>% filter(presence == 1)
poly_quinque_sf <- poly_quinque_sf %>% filter(presence == 1)
```

3. Project to Albers Equal Area and Calculate Overlap

```
# Define CRS
aea_crs <- st_crs("+proj=aea +lat_1=29.5 +lat_2=45.5 +lat_0=23 +lon_0=-96")

# Project
poly_pipiens_sf <- st_transform(poly_pipiens_sf, aea_crs)
poly_quinque_sf <- st_transform(poly_quinque_sf, aea_crs)

# Overlap
overlap_sf <- st_intersection(poly_pipiens_sf, poly_quinque_sf)

# Calculate areas (km²)
area_pipiens_km2 <- sum(st_area(poly_pipiens_sf)) / 1e6
area_quinque_km2 <- sum(st_area(poly_quinque_sf)) / 1e6
area_overlap_km2 <- sum(st_area(overlap_sf)) / 1e6</pre>
```

4. Clip to North America and Reproject to WGS84

```
# US base map
us_states <- st_as_sf(map("usa", plot = FALSE, fill = TRUE))
us_states <- st_transform(us_states, crs = aea_crs)

# Clip extent
bbox_na <- st_as_sfc(st_bbox(c(xmin = -170, xmax = -50, ymin = 5, ymax = 85), crs = st_crs(4326)))
bbox_na_sf <- st_transform(bbox_na, crs = aea_crs)

# Clip
poly_pipiens_sf <- st_intersection(st_make_valid(poly_pipiens_sf), bbox_na_sf)
poly_quinque_sf <- st_intersection(st_make_valid(poly_quinque_sf), bbox_na_sf)
overlap_sf <- st_intersection(st_make_valid(overlap_sf), bbox_na_sf)

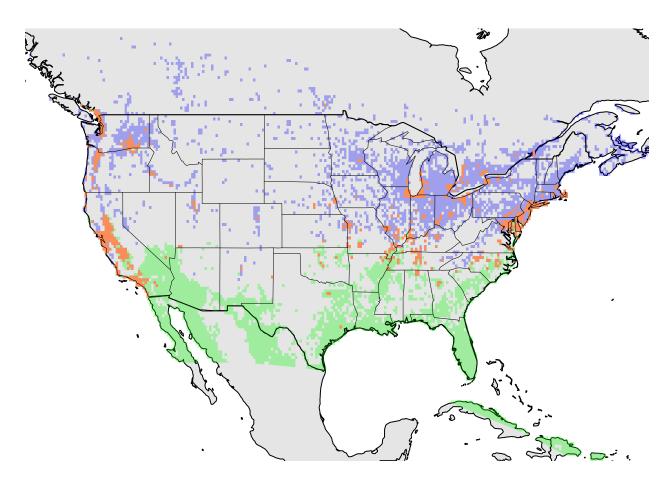
# Reproject to WGS84
poly_pipiens_ll <- st_transform(poly_pipiens_sf, 4326)
poly_quinque_ll <- st_transform(poly_quinque_sf, 4326)
overlap_ll <- st_transform(overlap_sf, 4326)</pre>
```

5. Visualize SDMs and Overlap

```
par(xpd = NA)
map("usa")
map(add = TRUE, col = "grey90", fill = TRUE)

plot(st_geometry(poly_pipiens_ll), col = rgb(0, 0, 1, 0.3), border = NA, add = TRUE)
plot(st_geometry(poly_quinque_ll), col = rgb(0, 1, 0, 0.3), border = NA, add = TRUE)
plot(st_geometry(overlap_ll), col = adjustcolor("#fc8d59", alpha.f = 1), border = NA, add = TRUE)

map("state", add = TRUE, col = "black", lwd = 0.5)
```



6. Merge Nearby Overlapping zones

```
# Reproject overlap
overlap_ll_proj <- st_transform(overlap_ll, aea_crs)</pre>
# Make valid and extract polygons
overlap_valid <- st_make_valid(overlap_ll_proj)</pre>
overlap_polygons <- st_collection_extract(overlap_valid, "POLYGON")</pre>
overlap_parts <- st_cast(overlap_polygons, "POLYGON")</pre>
# Buffer outward
buffer_dist_meters <- 25000</pre>
overlap_buffered <- st_buffer(overlap_parts, dist = buffer_dist_meters)</pre>
overlap_buffered <- st_make_valid(overlap_buffered)</pre>
# Merge touching patches
overlap_combined <- st_union(overlap_buffered)</pre>
overlap_combined <- st_make_valid(overlap_combined)</pre>
# Buffer inward
overlap_combined <- st_buffer(overlap_combined, dist = -buffer_dist_meters)</pre>
overlap_combined <- st_make_valid(overlap_combined)</pre>
# Finalize
overlap_combined <- st_cast(overlap_combined, "MULTIPOLYGON")</pre>
```

7. Visualize Combined Overlap zones

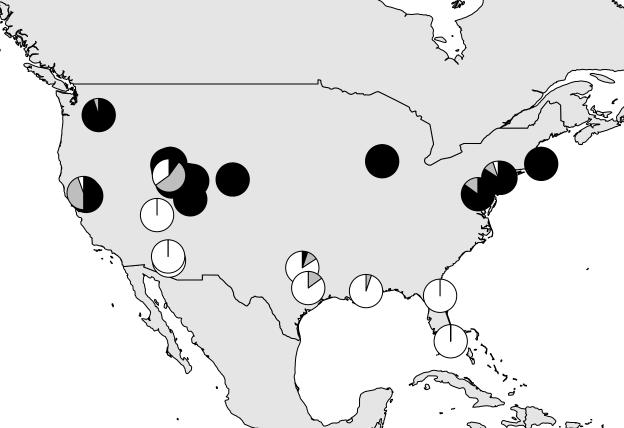
```
#pdf("../figs/overlap_SDM.pdf", width = 8, height = 6)
par(xpd = NA)
map("usa")
map(add = TRUE, col = "grey90", fill = TRUE)
plot(st_geometry(poly_pipiens_ll), col = rgb(0, 0, 1, 0.3), border = NA, add = TRUE)
plot(st_geometry(poly_quinque_ll), col = rgb(0, 1, 0, 0.3), border = NA, add = TRUE)
plot(st_geometry(overlap_combined), col = adjustcolor("#fc8d59", alpha.f = 0.9), border = NA, add = TRU
map("state", add = TRUE, col = "black", lwd = 0.5)
```

8. Add Sampling Points and Population Structure (Pie Charts)

#dev.off()

```
# Load counts
counts <- read.csv("../data/ThisStudy_v9.txt", sep = "\t")
coord <- as.data.frame(counts[,c("long","lat")])</pre>
```

```
# Prepare
countsDf <- as.data.frame(counts[,c(2,1,4,5,6,9,10,7,8)])</pre>
names(countsDf) <- c("locality", "site", "pp", "pq", "qq", "latitude", "longitude", "year", "h_index")</pre>
rownames(countsDf) <- countsDf$site</pre>
# Frequencies
freqsDf <- t(apply(countsDf[,c("pp","pq","qq")], 1, function(row) row / sum(row)))</pre>
freqsDf <- as.matrix(freqsDf)</pre>
# Plot pies
par(xpd = NA)
plot_order <- rev(order(countsDf$h_index))</pre>
map("usa")
map(add = TRUE, col = "grey90", fill = TRUE)
for (i in plot_order) {
  add.pie(z = freqsDf[i,],
          x = coord[i,1],
          y = coord[i, 2],
          clockwise = TRUE,
          labels = "",
          col = c("black", "grey", "white"),
          cex = 1.5, radius = 1.5)
```



Some jitter, added by hand:

Create a jitter data frame by hand:

```
# Create a jitter df
jitterDf <- data.frame(x_jitter = rep(0,26), y_jitter = rep(0,26))
rownames(jitterDf) <- rownames(countsDf)
jitterDf[3,] <- c(0,1)
jitterDf[4,] <- c(5,-1)
jitterDf[5,] <- c(1,-1)
jitterDf[6,] <- c(-2,0)
jitterDf[7,] <- c(-4,-1)
jitterDf[8,] <- c(-4,1)
jitterDf[9,] <- c(2,-3)
jitterDf[10,] <- c(5,-1)
jitterDf[15,] <- c(-7,3.5)
jitterDf[17,] <- c(-6,-1.5)
jitterDf[20,] <- c(1,1)
jitterDf[21,] <- c(1,-1)</pre>
```

Calculate bounds of map based on coordinates + jitter:

```
# Expand x/y limits by a small margin (e.g., 2 degrees)
x_range <- range(coord[, 1] + jitterDf[, 1], na.rm = TRUE)
y_range <- range(coord[, 2] + jitterDf[, 2], na.rm = TRUE)

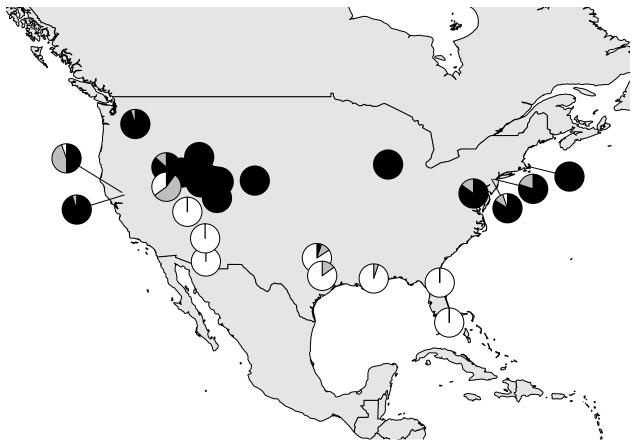
x_margin <- 2
y_margin <- 2

x_lim <- c(x_range[1] - x_margin, x_range[2] + x_margin)
y_lim <- c(y_range[1] - y_margin, y_range[2] + y_margin)</pre>
```

Add jitter with a line from true origin:

```
# Open PDF device
#pdf("../figs/ace2_pies_ThisStudy.pdf", width = 8, height = 6)
# Set xpd to NA to allow for plotting in the margins
par(xpd = NA)
# Plot map with expanded margins
map("usa", xlim = x_lim, ylim = y_lim)
map(add = TRUE, col = "grey90", fill = TRUE)
for (i in plot_order) {
  # Jittered coordinates
  jittered_x <- coord[i, 1] + (jitterDf[i, 1])</pre>
 jittered_y <- coord[i, 2] + (jitterDf[i, 2])</pre>
  # Draw line from true location to jittered pie
  segments(x0 = coord[i, 1],
           y0 = coord[i, 2],
           x1 = jittered_x,
           y1 = jittered_y,
           col = "black", lty = 1, lwd = 1)
  # Plot pie chart at jittered location
  add.pie(z = freqsDf[i,],
```

```
x = jittered_x,
y = jittered_y,
clockwise = TRUE,
labels = "",
col = c("black", "grey", "white"),
cex = 1.5, radius = 1.5)
}
```



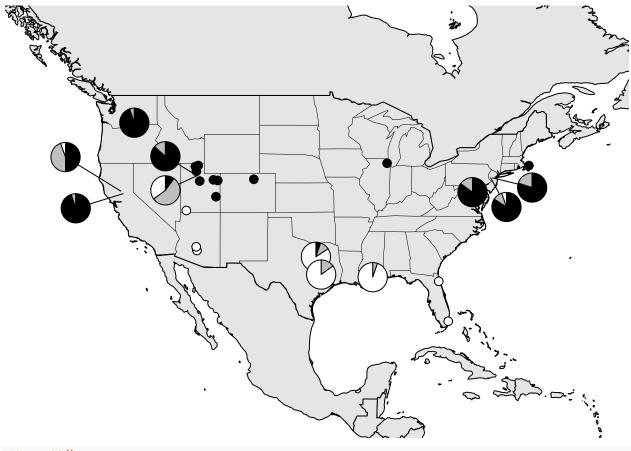
#dev.off()

Add jitter with a line from true origin AND fixed sites as small dots

```
# Open PDF device
#pdf("../figs/ace2_pies_ThisStudy.pdf", width = 8, height = 6)

#ADJUST the jitter df
jitterDf <- data.frame(x_jitter = rep(0,26), y_jitter = rep(0,26))
rownames(jitterDf) <- rownames(countsDf)
#jitterDf[3,] <- c(0,1)
#jitterDf[4,] <- c(5,-1)
#jitterDf[6,] <- c(-2,0)
jitterDf[7,] <- c(-4,-1.5)
jitterDf[8,] <- c(-4,2)
jitterDf[9,] <- c(2,-3)
jitterDf[10,] <- c(5,-1)
jitterDf[15,] <- c(-7,3.5)
```

```
jitterDf[17,] \leftarrow c(-6,-1.5)
#jitterDf[20,] <- c(1,1)
#jitterDf[21,] <- c(1,-1)
# Set xpd to NA to allow for plotting in the margins
par(xpd = NA)
# Add flag for fixed sites and what type of fixed (pp or qq)
freqs_fixed <- freqsDf[, 1] == 1 | freqsDf[, 3] == 1</pre>
freqs_fixed_type <- ifelse(freqsDf[, 1] == 1, "pp",</pre>
                            ifelse(freqsDf[, 3] == 1, "qq", NA))
# Plot map with expanded margins
map("usa", xlim = x_lim, ylim = y_lim)
map(add = TRUE, col = "grey90", fill = TRUE)
map("state", add = TRUE, col = "black", lwd = 0.5)
for (i in plot_order) {
  jittered_x <- coord[i, 1] + (jitterDf[i, 1])</pre>
  jittered_y <- coord[i, 2] + (jitterDf[i, 2])</pre>
  segments(x0 = coord[i, 1],
           y0 = coord[i, 2],
           x1 = jittered_x,
           y1 = jittered_y,
           col = "black", lty = 1, lwd = 1)
  if (freqs_fixed[i]) {
    if (freqs_fixed_type[i] == "pp") {
      # Black dot
      points(jittered_x, jittered_y, pch = 21, bg = "black", col = "black", cex = 1.2)
    } else if (freqs_fixed_type[i] == "qq") {
      # White dot with black border
      points(jittered_x, jittered_y, pch = 21, bg = "white", col = "black", cex = 1.2)
  } else {
    add.pie(z = freqsDf[i,],
            x = jittered_x,
            y = jittered_y,
            clockwise = TRUE,
            labels = "",
            col = c("black", "grey", "white"),
            cex = 1.5, radius = 1.5)
  }
}
```

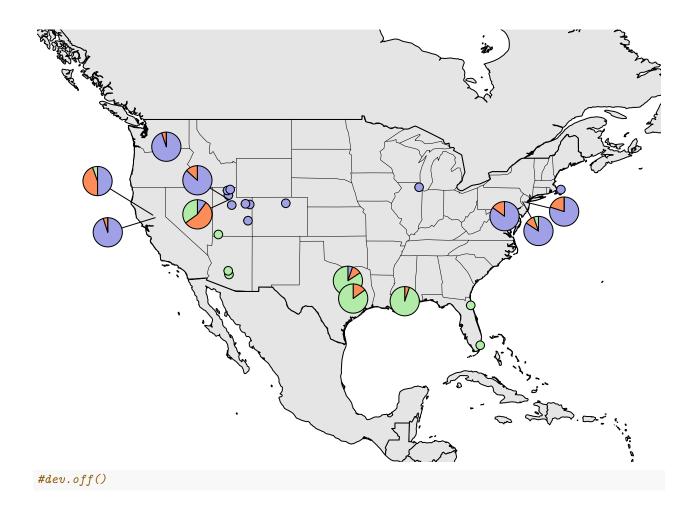


#dev.off()

Plot using colors from SDM:

```
# Open PDF device
#pdf("../figs/ace2_pies_rgb_ThisStudy.pdf", width = 8, height = 6)
#ADJUST the jitter df
jitterDf <- data.frame(x_jitter = rep(0,26), y_jitter = rep(0,26))</pre>
rownames(jitterDf) <- rownames(countsDf)</pre>
#jitterDf[3,] <- c(0,1)
#jitterDf[4,] \leftarrow c(5,-1)
#jitterDf[5,] <- c(1,-1)
\#jitterDf[6,] \leftarrow c(-2,0)
jitterDf[7,] \leftarrow c(-4,-1.5)
jitterDf[8,] \leftarrow c(-4,2)
jitterDf[9,] \leftarrow c(2,-3)
jitterDf[10,] \leftarrow c(5,-1)
jitterDf[15,] \leftarrow c(-7,3.5)
jitterDf[17,] \leftarrow c(-6,-1.5)
#jitterDf[20,] <- c(1,1)
#jitterDf[21,] <- c(1,-1)
# Set xpd to NA to allow for plotting in the margins
par(xpd = NA)
# Add flag for fixed sites and what type of fixed (pp or qq)
```

```
freqs_fixed <- freqsDf[, 1] == 1 | freqsDf[, 3] == 1</pre>
freqs_fixed_type <- ifelse(freqsDf[, 1] == 1, "pp",</pre>
                            ifelse(freqsDf[, 3] == 1, "qq", NA))
# Plot map with expanded margins
map("usa", xlim = x_lim, ylim = y_lim)
map(add = TRUE, col = "grey90", fill = TRUE)
map("state", add = TRUE, col = "black", lwd = 0.5)
for (i in plot_order) {
  jittered_x <- coord[i, 1] + (jitterDf[i, 1])</pre>
  jittered_y <- coord[i, 2] + (jitterDf[i, 2])</pre>
  segments(x0 = coord[i, 1],
           y0 = coord[i, 2],
           x1 = jittered_x,
           y1 = jittered_y,
           col = "black", lty = 1, lwd = 1)
  if (freqs_fixed[i]) {
    if (freqs_fixed_type[i] == "pp") {
      # Black dot
      points(jittered_x, jittered_y, pch = 21, bg = "#a0a0e7", col = "black", cex = 1.2)
    } else if (freqs_fixed_type[i] == "qq") {
      # White dot with black border
      points(jittered_x, jittered_y, pch = 21, bg = "#b1eba7", col = "black", cex = 1.2)
  } else {
    add.pie(z = freqsDf[i,],
            x = jittered_x,
            y = jittered_y,
            clockwise = TRUE,
            labels = "",
            col = c("#a0a0e7", "#fc8d59", "#b1eba7"),
            cex = 1.5, radius = 1.5)
 }
}
```



Predicted level of overlapping suitable habitat?

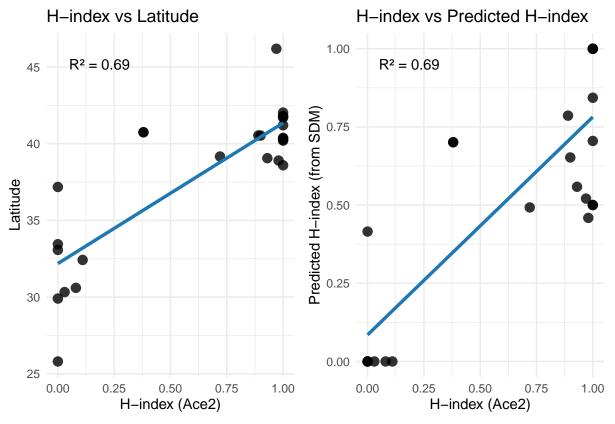
```
if (nrow(intersection) == 0) {
    # No overlap
   overlap_area <- 0
 } else {
    # Sum all overlapping parts
   overlap_area <- sum(st_area(intersection))</pre>
 buffer_area <- st_area(buf)</pre>
  # Return proportion
  as.numeric(overlap_area) / as.numeric(buffer_area)
})
# Step 6: Add predicted overlap to countsDf
countsDf$predicted_overlap <- predicted_overlap_prop</pre>
# Step 7: Quick check
head(countsDf[, c("site", "h_index", "predicted_overlap")])
##
                              site h_index predicted_overlap
## 001.ByroWA.2023 001.ByroWA.2023 0.97
                                                    0.8411216
## 007.CookIL.2023 007.CookIL.2023
                                      1.00
                                                   0.9993222
## 008.CachUT.2023 008.CachUT.2023 1.00
                                                  0.0000000
## 009.BarnMA.2023 009.BarnMA.2023
                                      1.00
                                                   0.8248844
                                    1.00
## 011.BoxEUT.2024 011.BoxEUT.2024
                                                    0.0000000
## 012.0gdeUT.2024 012.0gdeUT.2024
                                    1.00
                                                    0.4750927
Now, add directionality with 0 = quinq, 1 = pip, to estimate "predicted_h_index"
# -----
# New: Predict directional hybrid index from habitat around each sampling site
# Step 1: Prepare the pipiens-only and quinque-only polygons
# (We already have poly_pipiens_ll and poly_quinque_ll, but need to transform)
poly_pipiens_proj <- st_transform(poly_pipiens_ll, crs = aea_crs)</pre>
poly_quinque_proj <- st_transform(poly_quinque_ll, crs = aea_crs)</pre>
overlap_combined_proj <- st_transform(overlap_combined, crs = aea_crs) # already done above
# Step 2: Create pip-only and quinque-only polygons (remove overlap area)
pip_only <- st_difference(poly_pipiens_proj, overlap_combined_proj)</pre>
quinque_only <- st_difference(poly_quinque_proj, overlap_combined_proj)</pre>
# Step 3: Calculate areas for each sample buffer
library(units) # make sure units package is loaded
predicted_h_index <- sapply(1:nrow(sample_buffers), function(i) {</pre>
  buf <- sample_buffers[i, ]</pre>
  pip_intersect <- st_intersection(buf, pip_only)</pre>
  quinque_intersect <- st_intersection(buf, quinque_only)</pre>
  overlap_intersect <- st_intersection(buf, overlap_combined_proj)</pre>
 pip_area <- if (nrow(pip_intersect) == 0) units::set_units(0, "m^2") else sum(st_area(pip_intersect))</pre>
```

```
quinque_area <- if (nrow(quinque_intersect) == 0) units::set_units(0, "m^2") else sum(st_area(quinque
  overlap_area <- if (nrow(overlap_intersect) == 0) units::set_units(0, "m^2") else sum(st_area(overlap
  total_area <- pip_area + quinque_area + overlap_area
  if (as.numeric(total area) == 0) {
    return(NA) # no habitat found
    pred_h <- (pip_area + 0.5 * overlap_area) / total_area</pre>
    return(as.numeric(pred_h)) # strip units at the end
  }
})
# Step 4: Add predicted_h_index to countsDf
countsDf$predicted_h_index <- predicted_h_index</pre>
Plot h index versus latitude, and h index versus predicted h index based on SDM habitat MaxEnt models:
library(ggplot2)
library(patchwork)
# First plot: H-index vs Latitude
# -----
# Fit linear model for H-index vs Latitude
lm_lat <- lm(latitude ~ h_index, data = countsDf)</pre>
r_squared_lat <- summary(lm_lat)$r.squared
r_squared_lat_text <- paste0("R2 = ", round(r_squared_lat, 2))
p1 <- ggplot(countsDf, aes(x = h_index, y = latitude)) +
  geom_point(size = 3, alpha = 0.8, color = "black") +
  geom_smooth(method = "lm", formula = y ~ x, se = FALSE, color = "#1f78b4", linewidth = 1.2) +
  annotate("text", x = 0.05, y = max(countsDf$latitude, na.rm = TRUE) - 1, label = r_squared_lat_text,
  theme minimal() +
  labs(x = "H-index (Ace2)", y = "Latitude", title = "H-index vs Latitude") +
  xlim(0, 1)
# Second plot: H-index vs Predicted H-index
# Fit linear model for H-index vs Predicted H-index
lm_pred <- lm(predicted_h_index ~ h_index, data = countsDf)</pre>
r_squared_pred <- summary(lm_pred)$r.squared</pre>
r_squared_pred_text <- paste0("R2 = ", round(r_squared_pred, 2))
p2 <- ggplot(countsDf, aes(x = h_index, y = predicted_h_index)) +</pre>
  #qeom abline(slope = 1, intercept = 0, linetype = "dashed", color = "grey40") +
  geom_point(size = 3, alpha = 0.8, color = "black") +
  geom_smooth(method = "lm", formula = y ~ x, se = FALSE, color = "#1f78b4", linewidth = 1.2) +
  annotate("text", x = 0.05, y = 0.95, label = r_squared_pred_text, hjust = 0, size = 4) +
  theme minimal() +
  labs(x = "H-index (Ace2)", y = "Predicted H-index (from SDM)",
  title = "H-index vs Predicted H-index") +
```

```
xlim(0, 1) +
ylim(0, 1)

# ------
# Combine side-by-side
# ------

(p1 | p2) +
   plot_layout(guides = 'collect')
```



HWE Fisher's Exact Tests

```
library(dplyr)

# Create an empty list to store results
hwe_results <- list()

# Loop through each site
for (i in 1:nrow(countsDf)) {
    # Extract observed counts
    obs_pp <- countsDf$pp[i]
    obs_pq <- countsDf$pq[i]
    obs_qq <- countsDf$qq[i]

total_genotypes <- obs_pp + obs_pq + obs_qq

# Skip if total is 0
    if (total_genotypes == 0) {</pre>
```

```
hwe_results[[countsDf$site[i]]] <- NA</pre>
    next
  }
  # Check if there are at least two non-zero genotype classes
  non_zero_classes <- sum(c(obs_pp, obs_pq, obs_qq) > 0)
  if (non zero classes < 2) {</pre>
    hwe results[[countsDf$site[i]]] <- NA</pre>
    next
  }
  # Calculate allele frequencies
  p <- (2 * obs_pp + obs_pq) / (2 * total_genotypes)</pre>
  q < -1 - p
  # Expected counts under HWE
  exp_pp <- p^2 * total_genotypes</pre>
  exp_pq <- 2 * p * q * total_genotypes</pre>
  exp_qq <- q^2 * total_genotypes</pre>
  # Round expected counts
  exp_pp <- round(exp_pp)</pre>
  exp_pq <- round(exp_pq)</pre>
  exp_qq <- round(exp_qq)</pre>
  # Create contingency table
  table_hwe <- matrix(</pre>
    c(obs_pp, obs_pq, obs_qq,
      exp_pp, exp_pq, exp_qq),
    nrow = 2, byrow = TRUE
  colnames(table_hwe) <- c("pp", "pq", "qq")</pre>
  rownames(table_hwe) <- c("Observed", "Expected")</pre>
  # Perform Fisher's exact test
  fisher_test <- fisher.test(table_hwe, simulate.p.value = TRUE, B = 1e5)
  # Save the result
  hwe_results[[countsDf$site[i]]] <- list(</pre>
    p_value = fisher_test$p.value,
    observed = c(pp = obs_pp, pq = obs_pq, qq = obs_qq),
    expected = c(pp = exp_pp, pq = exp_pq, qq = exp_qq)
}
# Convert to a dataframe
hwe_summary <- do.call(rbind, lapply(names(hwe_results), function(site) {</pre>
  res <- hwe_results[[site]]</pre>
  if (is.null(res) | all(is.na(res))) {
    return(data.frame(site = site, p_value = NA))
  } else {
    return(data.frame(site = site, p_value = res$p_value))
```

```
}
}))
# View the result
hwe_summary
##
                 site
                        p_value
## 1 001.ByroWA.2023 1.0000000
## 2 007.CookIL.2023
## 3 008.CachUT.2023
## 4 009.BarnMA.2023
                             NA
## 5 011.BoxEUT.2024
## 6 012.0gdeUT.2024
## 7 015.SaltUT.2023 0.8274217
## 8 015.SaltUT.2018 1.0000000
## 9 022.HuntNJ.2023 0.6613734
## 10 023.SomeNJ.2023 1.0000000
## 11 024.FortC0.2023
## 12 025.UteTUT.2024
## 13 026.VernUT.2024
                             NA
## 14 028.ProvUT.2024
## 15 035.SuttCA.2023 1.0000000
## 16 038.RockMD.2023 1.0000000
## 17 041.LincCA.2023 1.0000000
## 18 054.MoabUT.2024
## 19 072.StGeUT.2023
                             NA
## 20 119.PhoeAZ.2024
## 21 123.MariAZ.2023
## 22 124.DalFTX.2024 0.6581934
## 23 130.CollTX.2023 1.0000000
## 24 131.SlidLA.2024 1.0000000
## 25 133.AnasFL.2023
                             NA
## 26 137.MiDaFL.2023
                             NA
```

Create a summary table

```
# Merge HWE p-values into countsDf
countsDf$HWE_p_value <- hwe_summary$p_value[match(countsDf$site, hwe_summary$site)]

# Add total N per site
countsDf$N <- countsDf$pp + countsDf$pq + countsDf$qq

# Function to pool and run HWE
pool_hwe_test <- function(df_zone) {
   total_pp <- sum(df_zone$pp, na.rm = TRUE)
   total_pq <- sum(df_zone$pq, na.rm = TRUE)
   total_qq <- sum(df_zone$qq, na.rm = TRUE)

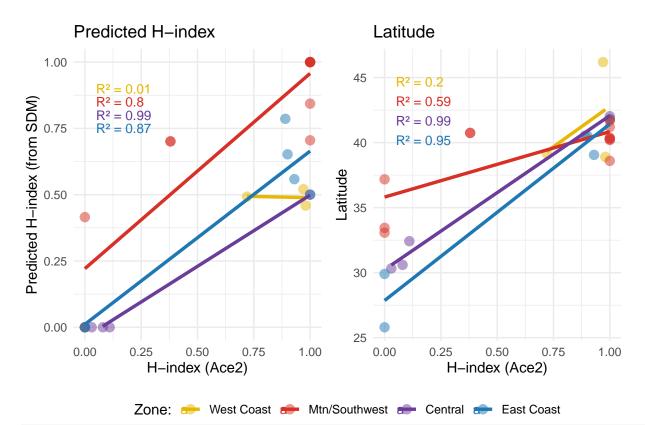
   total_N <- total_pp + total_pq + total_qq</pre>
```

```
if (total_N == 0) {
    return(data.frame(total_pp, total_pq, total_qq, total_N, pooled_HWE_p_value = NA))
  p <- (2 * total_pp + total_pq) / (2 * total_N)</pre>
 q < -1 - p
  exp pp <- round(p^2 * total N)</pre>
  exp_pq \leftarrow round(2 * p * q * total_N)
  exp_qq <- round(q^2 * total_N)</pre>
 table_hwe <- matrix(c(total_pp, total_pq, total_qq,</pre>
                         exp_pp, exp_pq, exp_qq),
                       nrow = 2, byrow = TRUE)
  colnames(table_hwe) <- c("pp", "pq", "qq")</pre>
  rownames(table_hwe) <- c("Observed", "Expected")</pre>
 fisher_test <- fisher.test(table_hwe, simulate.p.value = TRUE, B = 1e5)</pre>
 return(data.frame(total_pp, total_pq, total_qq, total_N, pooled_HWE_p_value = fisher_test$p.value))
}
countsDf$zone <- NULL</pre>
countsDf$zone <- case_when(</pre>
  grep1("CA|OR|WA", countsDf$site) ~ "West Coast",
  grep1("NJ|MD|MA|DE|CT|NY|VA|FL", countsDf$site) ~ "East Coast",
 grepl("TX|LA|IL", countsDf$site) ~ "Central",
  grepl("UT|CO|AZ", countsDf$site) ~ "Desert West",
 #grepl("UT/CO", countsDf$site) ~ "Mountain",
 TRUE ~ "Other"
# Apply by zone
summary_zones <- countsDf %>%
  group_by(zone) %>%
  group_modify(~ pool_hwe_test(.x)) %>%
 ungroup()
# Save summary zones to CSV
#write.csv(summary_zones, file = "../data/summary_zones_hindex_pred_HWE.csv", row.names = FALSE)
# Select and reorder columns
summary sites <- countsDf %>%
  select(site, pp, pq, qq, N, latitude, longitude, h_index, predicted_h_index, HWE_p_value, predicted_o
# Save summary sites as CSV
#write.csv(summary_sites, file = "../data/summary_sites_hindex_pred_HWE.csv.csv", row.names = TRUE)
```

Plot h index versus predicted h index, latitude, etc...

```
library(ggplot2)
library(dplyr)
library(patchwork)
# Define hybrid zones:
countsDf$zone <- NULL</pre>
countsDf$zone <- case when(</pre>
  grep1("CA|OR|WA", countsDf$site) ~ "West Coast",
  grepl("NJ|MD|MA|DE|CT|NY|VA|FL|QC|DC|TN|GA|NC", countsDf$site) ~ "East Coast",
  grep1("TX|LA|IL|LA|MN|ON|KS|MO|AR|AL|MS", countsDf$site) ~ "Central",
  grepl("UT|CO|AZ", countsDf$site) ~ "Mtn/Southwest",
  #grepl("UT/CO", countsDf$site) ~ "Mountain",
  TRUE ~ "Other"
# Control order by making it a factor
countsDf$zone <- factor(countsDf$zone, levels = c("West Coast", "Mtn/Southwest", "Central", "East Coast</pre>
# Define zone color palette
zone_colors <- c(</pre>
   "Mtn/Southwest" = "#d73027",
   "Central" = "#6a3d9a",
   "West Coast" = "#e6b800",
  "East Coast" = "#1f78b4",
   "Other" = "grey60"
                               # grey
)
# Create shared color scale
zone_scale <- scale_color_manual(values = zone_colors, name = "Zone:")</pre>
# Shared theme for both plots
shared_theme <- theme_minimal(base_size = 10) +</pre>
    plot.title = element_text(size = 12, face = "bold"),
   legend.position = "bottom", # <-- Legend at bottom</pre>
    axis.title = element_text(size = 10),
    axis.text = element_text(size = 8),
    plot.margin = margin(10, 10, 10, 10)
  )
# -----
# H-index vs Predicted H-index
r2_pred <- countsDf %>%
  group_by(zone) %>%
  summarise(r2 = summary(lm(predicted_h_index ~ h_index))$r.squared)
```

```
p2 <- ggplot(countsDf, aes(x = h_index, y = predicted_h_index, color = zone)) +
  geom_point(size = 3, alpha = 0.5) +
  geom_smooth(method = "lm", formula = y ~ x, se = FALSE, linewidth = 1.2) +
  geom_text(data = r2_pred, aes(x = 0.05, y = 0.95 - as.numeric(factor(zone)) * 0.05,
                               label = paste0("R^2 = ", round(r2, 2)), color = zone),
           hjust = 0, size = 3.5, inherit.aes = FALSE) +
  theme_minimal() +
  labs(x = "H-index (Ace2)", y = "Predicted H-index (from SDM)", title = "Predicted H-index", color = "
  xlim(0, 1) +
 ylim(0, 1) +
  scale_color_manual(values = zone_colors)
# H-index vs Latitude
# -----
r2_lat <- countsDf %>%
  group_by(zone) %>%
  summarise(r2 = summary(lm(latitude ~ h_index))$r.squared)
p1 <- ggplot(countsDf, aes(x = h_index, y = latitude, color = zone)) +
  geom_point(size = 3, alpha = 0.5) +
  geom_smooth(method = "lm", formula = y ~ x, se = FALSE, linewidth = 1.2) +
  geom_text(data = r2_lat, aes(x = 0.05, y = max(countsDf$latitude, na.rm = TRUE) - as.numeric(factor(z
                              label = paste0("R^2 = ", round(r2, 2)), color = zone),
           hjust = 0, size = 3.5, inherit.aes = FALSE) +
  theme minimal() +
  labs(x = "H-index (Ace2)", y = "Latitude", title = "Latitude", color = "Zone:") +
 xlim(0, 1) +
  scale_color_manual(values = zone_colors)
# Combine plots with unified legend at bottom
# Define the plot object
final_plot <- (p2 | p1) + plot_layout(guides = 'collect') & theme(legend.position = "bottom")
final_plot
```



Save as PDF with specific width and height (in inches)
#ggsave("../figs/h_index_lm_This_by_zone.pdf", plot = final_plot, width = 7, height = 4, units = "in")