

ELISA assignment Analysis

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```
# import data with the raw reading-----
patient1 <- import("Appendix 2.xlsx", sheet = "Patient 1") %>%
  # add a patient column
  mutate(patient = "Patient 1") %>%
  rename(naive_sera_control = 11) %>%
  select(11, patient) %>%
  mutate( group = c("ab1", "ab1", "ab2", "ab2", "ab3", "ab3", "ab4", "ab4")) %>%
  group_by(group, patient) %>%
  summarise(mean_ab_naive_s = mean(naive_sera_control, na.rm = T))
```

'summarise()' has grouped output by 'group'. You can override using the
'.groups' argument.

```
patient2 <- import("Appendix 2.xlsx", sheet = "Patient 2") %>%
  # add a patient column
  mutate(patient = "Patient 2") %>%
  rename(naive_sera_control = 11) %>%
  select(11, patient) %>%
  mutate( group = c("ab1", "ab1", "ab2", "ab2", "ab3", "ab3", "ab4", "ab4")) %>%
  group_by(group, patient) %>%
  summarise(mean_ab_naive_s = mean(naive_sera_control, na.rm = T))
```

'summarise()' has grouped output by 'group'. You can override using the
'.groups' argument.

```
patient3 <- import("Appendix 2.xlsx", sheet = "Patient 3") %>%
  # add a patient column
  mutate(patient = "Patient 3") %>%
  rename(naive_sera_control = 11) %>%
  select(11, patient) %>%
  mutate( group = c("ab1", "ab1", "ab2", "ab2", "ab3", "ab3", "ab4", "ab4")) %>%
  group_by(group, patient) %>%
  summarise(mean_ab_naive_s = mean(naive_sera_control, na.rm = T))
```

'summarise()' has grouped output by 'group'. You can override using the
'.groups' argument.

```
patient4 <- import("Appendix 2.xlsx", sheet = "Patient 4") %>%
  # add a patient column
```

```

mutate(patient = "Patient 4") %>%
rename(naive_sera_control = 11) %>%
select(11, patient) %>%
mutate( group = c("ab1", "ab1", "ab2", "ab2", "ab3", "ab3", "ab4", "ab4")) %>%
group_by(group, patient) %>%
summarise(mean_ab_naive_s = mean(naive_sera_control, na.rm = T))

```

'summarise()' has grouped output by 'group'. You can override using the
'.groups' argument.

```

patient5 <- import("Appendix 2.xlsx", sheet = "Patient 5") %>%
# add a patient column
mutate(patient = "Patient 5") %>%
rename(naive_sera_control = 11) %>%
select(11, patient) %>%
mutate( group = c("ab1", "ab1", "ab2", "ab2", "ab3", "ab3", "ab4", "ab4")) %>%
group_by(group, patient) %>%
summarise(mean_ab_naive_s = mean(naive_sera_control, na.rm = T))

```

'summarise()' has grouped output by 'group'. You can override using the
'.groups' argument.

```

# bind the rows

patients_naive_sera <- bind_rows(patient1, patient2, patient3, patient4, patient5) %>%
  rename(patient = patient,
         Antibody = group) %>%
  mutate(Antibody = recode(Antibody,
                           "ab1" = "Antibody 1",
                           "ab2" = "Antibody 2",
                           "ab3" = "Antibody 3",
                           "ab4" = "Antibody 4")) %>%
  mutate(mean_ab_naive_s = log(mean_ab_naive_s))

```

```

patient_1_raw <- import("Appendix 2.xlsx", sheet = "Patient 1") %>%

# add a patient column
mutate(patient = "Patient 1") %>%

# add the dilution column
mutate(dilution = c(0.10, 0.02, 0.004, 0.0008, 0.00016, 0.000032, 0.0000064, 0)) %>%

# rename the columns
rename(Ab1_R1 = 3, Ab1_R2 = 4, Ab2_R1 = 5, Ab2_R2 = 6,
       Ab3_R1 = 7, Ab3_R2 = 8, Ab4_R1 = 9,
       Ab4_R2 = 10, `PBS (-ve control)` = 2,
       plate = 1) %>%

# group row wise
rowwise() %>%

# calculate the average

```

```

mutate(
  Average_Ab1 = mean(Ab1_R1,Ab1_R2, na.rm = T),
  Average_Ab2 = mean(Ab2_R1,Ab2_R2, na.rm = T),
  Average_Ab3 = mean(Ab3_R1,Ab3_R2, na.rm = T),
  Average_Ab4 = mean(Ab4_R1,Ab4_R2, na.rm = T)) %>%
select(Average_Ab1,
       Average_Ab2,
       Average_Ab3,
       Average_Ab4,
       `PBS (-ve control)`,
       dilution,
       patient
)

patient_2_raw <- import(("Appendix 2.xlsx"), sheet = "Patient 2") %>%

mutate(patient = "Patient 2") %>%
mutate(dilution = c(0.10, 0.02, 0.004, 0.0008, 0.00016,0.000032,0.0000064,0)) %>%
rename(Ab1_R1 = 3, Ab1_R2 = 4, Ab2_R1 = 5, Ab2_R2 = 6,
       Ab3_R1 = 7, Ab3_R2 = 8, Ab4_R1 = 9,
       Ab4_R2 = 10, `PBS (-ve control)` = 2,
       plate = 1) %>%
rowwise() %>%
mutate(
  Average_Ab1 = mean(Ab1_R1,Ab1_R2, na.rm = T),
  Average_Ab2 = mean(Ab2_R1,Ab2_R2, na.rm = T),
  Average_Ab3 = mean(Ab3_R1,Ab3_R2, na.rm = T),
  Average_Ab4 = mean(Ab4_R1,Ab4_R2, na.rm = T)) %>%
select(Average_Ab1,
       Average_Ab2,
       Average_Ab3,
       Average_Ab4,
       `PBS (-ve control)`,
       dilution,
       patient
)

patient_3_raw <- import(("Appendix 2.xlsx"), sheet = "Patient 3") %>%
mutate(patient = "Patient 3") %>%
mutate(dilution = c(0.10, 0.02, 0.004, 0.0008, 0.00016,0.000032,0.0000064,0)) %>%
rename(Ab1_R1 = 3, Ab1_R2 = 4, Ab2_R1 = 5, Ab2_R2 = 6,
       Ab3_R1 = 7, Ab3_R2 = 8, Ab4_R1 = 9,
       Ab4_R2 = 10, `PBS (-ve control)` = 2,
       plate = 1) %>%
rowwise() %>%
mutate(
  Average_Ab1 = mean(Ab1_R1,Ab1_R2, na.rm = T),
  Average_Ab2 = mean(Ab2_R1,Ab2_R2, na.rm = T),
  Average_Ab3 = mean(Ab3_R1,Ab3_R2, na.rm = T),
  Average_Ab4 = mean(Ab4_R1,Ab4_R2, na.rm = T)) %>%
select(Average_Ab1,

```

```

    Average_Ab2,
    Average_Ab3,
    Average_Ab4,
    `PBS (-ve control)`,
    dilution,
    patient
  )
)

patient_4_raw <- import("Appendix 2.xlsx", sheet = "Patient 4") %>%
  mutate(patient = "Patient 4") %>%
  mutate(dilution = c(0.10, 0.02, 0.004, 0.0008, 0.00016, 0.000032, 0.0000064, 0)) %>%
  rename(Ab1_R1 = 3, Ab1_R2 = 4, Ab2_R1 = 5, Ab2_R2 = 6,
    Ab3_R1 = 7, Ab3_R2 = 8, Ab4_R1 = 9,
    Ab4_R2 = 10, `PBS (-ve control)` = 2,
    plate = 1) %>%
  rowwise() %>%
  mutate(
    Average_Ab1 = mean(Ab1_R1, Ab1_R2, na.rm = T),
    Average_Ab2 = mean(Ab2_R1, Ab2_R2, na.rm = T),
    Average_Ab3 = mean(Ab3_R1, Ab3_R2, na.rm = T),
    Average_Ab4 = mean(Ab4_R1, Ab4_R2, na.rm = T)) %>%
  select(Average_Ab1,
    Average_Ab2,
    Average_Ab3,
    Average_Ab4,
    `PBS (-ve control)`,
    dilution,
    patient
  )
)

patient_5_raw <- import("Appendix 2.xlsx", sheet = "Patient 5") %>%
  mutate(patient = "Patient 5") %>%
  mutate(dilution = c(0.10, 0.02, 0.004, 0.0008, 0.00016, 0.000032, 0.0000064, 0)) %>%
  rename(Ab1_R1 = 3, Ab1_R2 = 4, Ab2_R1 = 5, Ab2_R2 = 6,
    Ab3_R1 = 7, Ab3_R2 = 8, Ab4_R1 = 9,
    Ab4_R2 = 10, `PBS (-ve control)` = 2,
    plate = 1) %>%
  rowwise() %>%
  mutate(
    Average_Ab1 = mean(Ab1_R1, Ab1_R2, na.rm = T),
    Average_Ab2 = mean(Ab2_R1, Ab2_R2, na.rm = T),
    Average_Ab3 = mean(Ab3_R1, Ab3_R2, na.rm = T),
    Average_Ab4 = mean(Ab4_R1, Ab4_R2, na.rm = T)) %>%
  select(Average_Ab1,
    Average_Ab2,
    Average_Ab3,
    Average_Ab4,
    `PBS (-ve control)`,
    dilution,
    patient
  )
)

```

patient_1_raw

```
## # A tibble: 8 x 7
## # Rowwise:
##   Average_Ab1 Average_Ab2 Average_Ab3 Average_Ab4 'PBS (-ve control)' dilution
##   <dbl>      <dbl>      <dbl>      <dbl>      <dbl>      <dbl>
## 1     0.471     0.463     0.978     0.238     0.199 0.1
## 2     0.293     0.253     0.886     0.215     0.204 0.02
## 3     0.207     0.200     0.693     0.213     0.197 0.004
## 4     0.224     0.208     0.456     0.224     0.202 0.0008
## 5     0.218     0.213     0.399     0.212     0.201 0.00016
## 6     0.211     0.205     0.238     0.214     0.200 0.000032
## 7     0.211     0.2      0.214     0.216     0.2   0.0000064
## 8     0.211     0.203     0.250     0.214     0.198 0
## # i 1 more variable: patient <chr>
```

patient_2_raw

```
## # A tibble: 8 x 7
## # Rowwise:
##   Average_Ab1 Average_Ab2 Average_Ab3 Average_Ab4 'PBS (-ve control)' dilution
##   <dbl>      <dbl>      <dbl>      <dbl>      <dbl>      <dbl>
## 1     0.631     0.517     1.29      0.426     0.195 0.1
## 2     0.452     0.328     1.15      0.363     0.211 0.02
## 3     0.447     0.384     0.852     0.258     0.2   0.004
## 4     0.377     0.288     0.501     0.354     0.206 0.0008
## 5     0.372     0.287     0.402     0.445     0.205 0.00016
## 6     0.239     0.259     0.403     0.302     0.202 0.000032
## 7     0.259     0.262     0.397     0.226     0.194 0.0000064
## 8     0.260     0.277     0.441     0.264     0.195 0
## # i 1 more variable: patient <chr>
```

patient_3_raw

```
## # A tibble: 8 x 7
## # Rowwise:
##   Average_Ab1 Average_Ab2 Average_Ab3 Average_Ab4 'PBS (-ve control)' dilution
##   <dbl>      <dbl>      <dbl>      <dbl>      <dbl>      <dbl>
## 1     0.289     0.268     0.509     0.237     0.196 0.1
## 2     0.233     0.250     0.370     0.216     0.199 0.02
## 3     0.211     0.222     0.299     0.213     0.199 0.004
## 4     0.218     0.222     0.241     0.208     0.195 0.0008
## 5     0.209     0.212     0.226     0.21     0.196 0.00016
## 6     0.208     0.219     0.216     0.202     0.197 0.000032
## 7     0.206     0.211     0.207     0.205     0.194 0.0000064
## 8     0.204     0.210     0.206     0.206     0.194 0
## # i 1 more variable: patient <chr>
```

patient_4_raw

```
## # A tibble: 8 x 7
```

```
## # Rowwise:
##   Average_Ab1 Average_Ab2 Average_Ab3 Average_Ab4 'PBS (-ve control)' dilution
##   <dbl>      <dbl>      <dbl>      <dbl>      <dbl>      <dbl>
## 1      1.43      1.32      1.60      0.668      0.183 0.1
## 2      1.09      0.881     1.51      0.340      0.175 0.02
## 3      0.681     0.382     1.12      0.230      0.182 0.004
## 4      0.316     0.252     0.551     0.21       0.180 0.0008
## 5      0.22      0.214     0.277     0.212      0.187 0.00016
## 6      0.21      0.199     0.218     0.207      0.186 0.000032
## 7      0.211     0.208     0.205     0.215      0.194 0.0000064
## 8      0.218     0.200     0.200     0.200      0.181 0
## # i 1 more variable: patient <chr>
```

```
patient_5_raw
```

```
## # A tibble: 8 x 7
## # Rowwise:
##   Average_Ab1 Average_Ab2 Average_Ab3 Average_Ab4 'PBS (-ve control)' dilution
##   <dbl>      <dbl>      <dbl>      <dbl>      <dbl>      <dbl>
## 1      0.463     0.428     0.633     0.256      0.193 0.1
## 2      0.346     0.327     0.475     0.210      0.167 0.02
## 3      0.282     0.255     0.302     0.203      0.156 0.004
## 4      0.238     0.230     0.253     0.213      0.146 0.0008
## 5      0.232     0.223     0.218     0.206      0.146 0.00016
## 6      0.215     0.225     0.212     0.192      0.142 0.000032
## 7      0.204     0.217     0.199     0.219      0.152 0.0000064
## 8      0.241     0.195     0.212     0.202      0.178 0
## # i 1 more variable: patient <chr>
```

```
p1_mean_diff <- patient_1_raw %>%
rowwise() %>%
  mutate(redu_av_Ab1 = 0.2112 - Average_Ab1,
         redu_av_Ab2 = 0.2034 - Average_Ab2,
         redu_av_Ab3 = 0.2502 - Average_Ab3,
         redu_av_Ab4 = 0.2142 - Average_Ab4) %>%
  select(redu_av_Ab1, redu_av_Ab2, redu_av_Ab3, redu_av_Ab4, patient, dilution, `PBS (-ve control)`)

p2_mean_diff <- patient_2_raw %>%
rowwise() %>%
  mutate(redu_av_Ab1 = 0.2605 - Average_Ab1,
         redu_av_Ab2 = 0.2769 - Average_Ab2,
         redu_av_Ab3 = 0.4411 - Average_Ab3,
         redu_av_Ab4 = 0.2638 - Average_Ab4) %>%
  select(redu_av_Ab1, redu_av_Ab2, redu_av_Ab3, redu_av_Ab4, patient, dilution, `PBS (-ve control)`)

p3_mean_diff <- patient_3_raw %>%
rowwise() %>%
  mutate(redu_av_Ab1 = 0.2036 - Average_Ab1,
         redu_av_Ab2 = 0.2104 - Average_Ab2,
         redu_av_Ab3 = 0.2058 - Average_Ab3,
         redu_av_Ab4 = 0.2059 - Average_Ab4) %>%
```

```

select(redu_av_Ab1, redu_av_Ab2, redu_av_Ab3, redu_av_Ab4, patient, dilution, `PBS (-ve control)`)

p4_mean_diff <- patient_4_raw %>%
rowwise() %>%
  mutate(redu_av_Ab1 = 0.2180 - Average_Ab1,
         redu_av_Ab2 = 0.2003 - Average_Ab2,
         redu_av_Ab3 = 0.1999 - Average_Ab3,
         redu_av_Ab4 = 0.1996 - Average_Ab4) %>%
select(redu_av_Ab1, redu_av_Ab2, redu_av_Ab3, redu_av_Ab4, patient, dilution, `PBS (-ve control)`)

p5_mean_diff <- patient_5_raw %>%
rowwise() %>%
  mutate(redu_av_Ab1 = 0.2407 - Average_Ab1,
         redu_av_Ab2 = 0.1949 - Average_Ab2,
         redu_av_Ab3 = 0.2124 - Average_Ab3,
         redu_av_Ab4 = 0.2025 - Average_Ab4) %>%
select(redu_av_Ab1, redu_av_Ab2, redu_av_Ab3, redu_av_Ab4, patient, dilution, `PBS (-ve control)`)

p1_mean_diff

```

```

## # A tibble: 8 x 7
## # Rowwise:
##   redu_av_Ab1 redu_av_Ab2 redu_av_Ab3 redu_av_Ab4 patient dilution
##   <dbl>      <dbl>      <dbl>      <dbl> <chr>      <dbl>
## 1   -0.260      -0.259      -0.728     -0.0233 Patient 1 0.1
## 2   -0.0822     -0.05       -0.635     -0.00120 Patient 1 0.02
## 3    0.00440     0.00330     -0.443      0.00100 Patient 1 0.004
## 4   -0.0130     -0.00450     -0.206     -0.0103 Patient 1 0.0008
## 5   -0.00710     -0.00970     -0.149      0.00230 Patient 1 0.00016
## 6    0.000600    -0.00170      0.0122      0.000300 Patient 1 0.000032
## 7    0.000200     0.00340      0.0364     -0.00190 Patient 1 0.0000064
## 8      0         0           0           0 Patient 1 0
## # i 1 more variable: `PBS (-ve control)` <dbl>

```

```
p2_mean_diff
```

```

## # A tibble: 8 x 7
## # Rowwise:
##   redu_av_Ab1 redu_av_Ab2 redu_av_Ab3 redu_av_Ab4 patient dilution
##   <dbl>      <dbl>      <dbl>      <dbl> <chr>      <dbl>
## 1   -0.371      -0.24       -0.844     -0.163 Patient 2 0.1
## 2   -0.191      -0.0510     -0.708     -0.0995 Patient 2 0.02
## 3   -0.187      -0.107      -0.411      0.00590 Patient 2 0.004
## 4   -0.117      -0.0107     -0.0601     -0.0907 Patient 2 0.0008
## 5   -0.111      -0.0105      0.0396     -0.181 Patient 2 0.00016
## 6    0.0214      0.0177      0.0383     -0.0380 Patient 2 0.000032
## 7    0.00170      0.0151      0.0441      0.0379 Patient 2 0.0000064
## 8      0         0           0           0 Patient 2 0
## # i 1 more variable: `PBS (-ve control)` <dbl>

```

```
p3_mean_diff
```

```
## # A tibble: 8 x 7
## # Rowwise:
##   redu_av_Ab1 redu_av_Ab2 redu_av_Ab3 redu_av_Ab4 patient dilution
##   <dbl>      <dbl>      <dbl>      <dbl> <chr>      <dbl>
## 1   -0.085    -0.0571    -0.303    -0.0308 Patient 3 0.1
## 2   -0.0293   -0.0391    -0.164    -0.00970 Patient 3 0.02
## 3   -0.00760  -0.0116    -0.093    -0.00750 Patient 3 0.004
## 4   -0.0144   -0.0113    -0.0351   -0.00170 Patient 3 0.0008
## 5   -0.00570  -0.00170   -0.0198   -0.00410 Patient 3 0.00016
## 6   -0.00390  -0.00890   -0.0101    0.00340 Patient 3 0.000032
## 7   -0.00210  -0.000900  -0.000900  0.000900 Patient 3 0.0000064
## 8     0       0       0       0 Patient 3 0
## # i 1 more variable: 'PBS (-ve control)' <dbl>
```

```
p5_mean_diff
```

```
## # A tibble: 8 x 7
## # Rowwise:
##   redu_av_Ab1 redu_av_Ab2 redu_av_Ab3 redu_av_Ab4 patient dilution
##   <dbl>      <dbl>      <dbl>      <dbl> <chr>      <dbl>
## 1   -0.222    -0.234    -0.420    -0.0531 Patient 5 0.1
## 2   -0.105    -0.132    -0.262    -0.00770 Patient 5 0.02
## 3   -0.041    -0.0602   -0.0891   -0.000500 Patient 5 0.004
## 4    0.00270  -0.0355   -0.0402   -0.0107 Patient 5 0.0008
## 5    0.0088   -0.0277   -0.00570  -0.00400 Patient 5 0.00016
## 6    0.0261   -0.0299    0.000900  0.0107 Patient 5 0.000032
## 7    0.0369   -0.0218    0.0135   -0.0164 Patient 5 0.0000064
## 8     0       0       0       0 Patient 5 0
## # i 1 more variable: 'PBS (-ve control)' <dbl>
```

```
p5_mean_diff
```

```
## # A tibble: 8 x 7
## # Rowwise:
##   redu_av_Ab1 redu_av_Ab2 redu_av_Ab3 redu_av_Ab4 patient dilution
##   <dbl>      <dbl>      <dbl>      <dbl> <chr>      <dbl>
## 1   -0.222    -0.234    -0.420    -0.0531 Patient 5 0.1
## 2   -0.105    -0.132    -0.262    -0.00770 Patient 5 0.02
## 3   -0.041    -0.0602   -0.0891   -0.000500 Patient 5 0.004
## 4    0.00270  -0.0355   -0.0402   -0.0107 Patient 5 0.0008
## 5    0.0088   -0.0277   -0.00570  -0.00400 Patient 5 0.00016
## 6    0.0261   -0.0299    0.000900  0.0107 Patient 5 0.000032
## 7    0.0369   -0.0218    0.0135   -0.0164 Patient 5 0.0000064
## 8     0       0       0       0 Patient 5 0
## # i 1 more variable: 'PBS (-ve control)' <dbl>
```

```
elisa_red_av <- bind_rows(p1_mean_diff,
                           p2_mean_diff,
                           p3_mean_diff,
```



```

        p4_mean_diff,
        p5_mean_diff) %>%
mutate(dilution = format(dilution,scientific = FALSE)) %>%
mutate(dilution = as.numeric(dilution)) %>%
select(redu_av_Ab1,
       redu_av_Ab2,
       redu_av_Ab3,
       redu_av_Ab4,
       `PBS (-ve control)`,
       dilution,
       patient
)
elisa_red_av

```

```

## # A tibble: 40 x 7
## # Rowwise:
##   redu_av_Ab1 redu_av_Ab2 redu_av_Ab3 redu_av_Ab4 `PBS (-ve control)` dilution
##   <dbl>      <dbl>      <dbl>      <dbl>      <dbl>      <dbl>
## 1  -0.260    -0.259    -0.728    -0.0233      0.199 0.1
## 2  -0.0822   -0.05     -0.635    -0.00120     0.204 0.02
## 3   0.00440   0.00330   -0.443     0.00100     0.197 0.004
## 4  -0.0130   -0.00450   -0.206    -0.0103     0.202 0.0008
## 5  -0.00710  -0.00970   -0.149     0.00230     0.201 0.00016
## 6   0.000600 -0.00170    0.0122    0.000300     0.200 0.000032
## 7   0.000200  0.00340    0.0364   -0.00190     0.2   0.0000064
## 8    0        0          0         0          0.198 0
## 9  -0.371    -0.24     -0.844    -0.163      0.195 0.1
## 10 -0.191    -0.0510   -0.708    -0.0995     0.211 0.02
## # i 30 more rows
## # i 1 more variable: patient <chr>

```

```

elisa_raw <- bind_rows(patient_1_raw,
                       patient_2_raw,
                       patient_3_raw,
                       patient_4_raw,
                       patient_5_raw) %>%
mutate(dilution = format(dilution,scientific = FALSE)) %>%
mutate(dilution = as.numeric(dilution)) %>%
select(Average_Ab1,
       Average_Ab2,
       Average_Ab3,
       Average_Ab4,
       `PBS (-ve control)`,
       dilution,
       patient
)

elisa_raw

```

```

## # A tibble: 40 x 7
## # Rowwise:
##   Average_Ab1 Average_Ab2 Average_Ab3 Average_Ab4 `PBS (-ve control)` dilution
##   <dbl>      <dbl>      <dbl>      <dbl>      <dbl>      <dbl>

```

```
## 1      0.471      0.463      0.978      0.238      0.199 0.1
## 2      0.293      0.253      0.886      0.215      0.204 0.02
## 3      0.207      0.200      0.693      0.213      0.197 0.004
## 4      0.224      0.208      0.456      0.224      0.202 0.0008
## 5      0.218      0.213      0.399      0.212      0.201 0.00016
## 6      0.211      0.205      0.238      0.214      0.200 0.000032
## 7      0.211      0.2      0.214      0.216      0.2 0.0000064
## 8      0.211      0.203      0.250      0.214      0.198 0
## 9      0.631      0.517      1.29      0.426      0.195 0.1
## 10     0.452      0.328      1.15      0.363      0.211 0.02
## # i 30 more rows
## # i 1 more variable: patient <chr>
```

```
elisa <- elisa_raw %>%
  mutate(dilution = log(dilution)) %>%
  pivot_longer(
    cols = c(Average_Ab1:`PBS (-ve control)`),
    names_to = "Antibody",
    values_to = "Absorbtion"
  ) %>%
  mutate(Absorbtion = log(Absorbtion)) %>%
  mutate(Antibody = recode(Antibody,
    "Average_Ab1" = "Antibody 1",
    "Average_Ab2" = "Antibody 2",
    "Average_Ab3" = "Antibody 3",
    "Average_Ab4" = "Antibody 4"
  )) %>%
  mutate(identity_combined = paste(patient, Antibody))

# create the background noise for the standard (row-H)-----

Background_noise <- elisa %>%
  filter(dilution == -Inf & Antibody != "PBS (-ve control)") %>%
  mutate(identity_combined = paste(patient, Antibody))

elisa
```

```
## # A tibble: 200 x 5
##   dilution patient Antibody Absorbtion identity_combined
##   <dbl> <chr> <chr> <dbl> <chr>
## 1 -2.30 Patient 1 Antibody 1 -0.753 Patient 1 Antibody 1
## 2 -2.30 Patient 1 Antibody 2 -0.770 Patient 1 Antibody 2
## 3 -2.30 Patient 1 Antibody 3 -0.0217 Patient 1 Antibody 3
## 4 -2.30 Patient 1 Antibody 4 -1.44 Patient 1 Antibody 4
## 5 -2.30 Patient 1 PBS (-ve control) -1.61 Patient 1 PBS (-ve control)
## 6 -3.91 Patient 1 Antibody 1 -1.23 Patient 1 Antibody 1
## 7 -3.91 Patient 1 Antibody 2 -1.37 Patient 1 Antibody 2
## 8 -3.91 Patient 1 Antibody 3 -0.121 Patient 1 Antibody 3
## 9 -3.91 Patient 1 Antibody 4 -1.54 Patient 1 Antibody 4
## 10 -3.91 Patient 1 PBS (-ve control) -1.59 Patient 1 PBS (-ve control)
## # i 190 more rows
```

```
# ELISA corrected-----
```

```
elisa_corrected <- elisa_red_av %>%
  mutate(dilution = log10(dilution)) %>%
  pivot_longer(
    cols = c(redu_av_Ab1:`PBS (-ve control)`),
    names_to = "Antibody",
    values_to = "Absorbtion"
  ) %>%
  mutate(Absorbtion = log10(Absorbtion))
```

```
## Warning: There was 1 warning in 'mutate()'.
## i In argument: 'Absorbtion = log10(Absorbtion)'.
## Caused by warning:
## ! NaNs produced
```

```
# mutate(Antibody = recode(Antibody,
#                           "Average_Ab1" = "Antibody 1",
#                           "Average_Ab2" = "Antibody 2",
#                           "Average_Ab3" = "Antibody 3",
#                           "Average_Ab4" = "Antibody 4"
# )) %>%
# mutate(identity_combined = paste(patient, Antibody))
```

```
# Display background noise -----
```

```
elisa %>%
  filter(dilution != -Inf) %>%
  ggplot(aes(x = dilution,
             y = Absorbtion,
             color = Antibody)) +
  geom_point() +
  geom_line(size = 1.0) +

  # set the breaks accordings to the natural log value of the concentartion

  scale_x_continuous(breaks = c(-11.9, -10.3, -8.7, -7.1, -5.5, -3.9, -2.3)) +

  facet_wrap(~patient,
             scales = "free",
             ncol = 2) +
  labs(y = "The log of average spectrophotometer reading for each antibody",
       x = "The log of dilution (from dilution factor 1/156,250 to 1/10)",
       title = "Indirect sandwich ELISA test of four different antibodies to diagnose DENV-3 from patient",
       subtitle = "**Scales are in natural logarithmic scale **not base10**",
       fill = "Antibody",
       caption = "The photospectometer reading and the \n
dilution factor are changed in to base log values \n
from -11.9 (1/156250) to -2.3 (1/10) in the x axis, \n
and from -1.6 (0.1946) to 0.5 (1.5995) in the y axis") +
  theme(axis.title = element_text(size = 12, face = "italic", color = "blue"),
        plot.title = element_text(size = 12, face = "bold", color = "red"),
        plot.subtitle = element_text(size = 12, face = 'italic', color = "blue"),
```

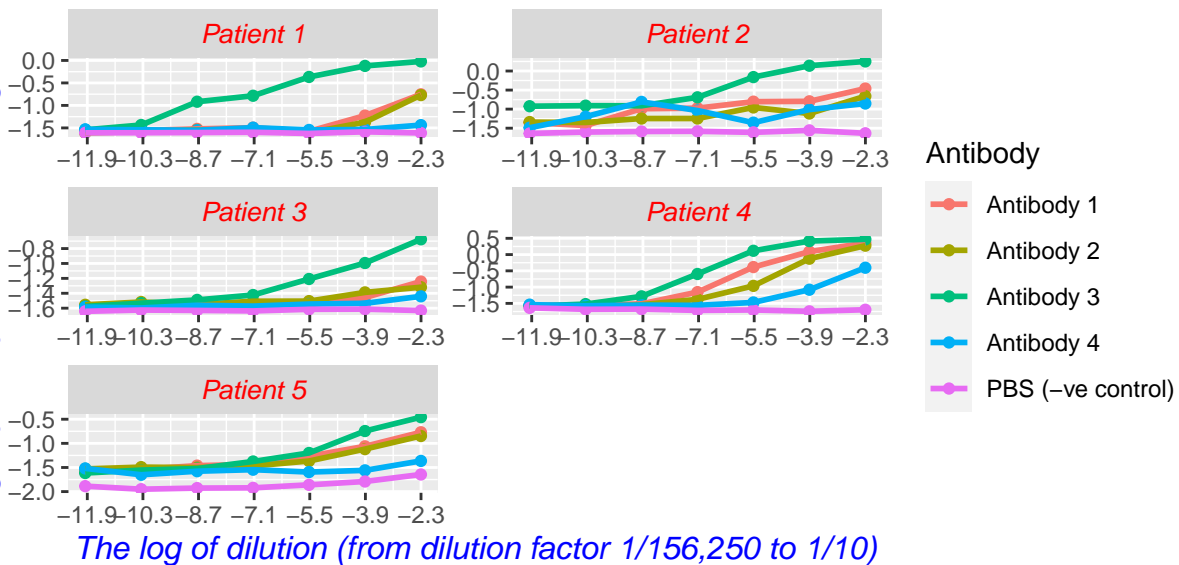
```
strip.placement = "outside",
strip.text = element_text(face = "italic", color = "red", size = 10),
plot.caption = element_text(face = "italic", color = "red"))
```

```
## Warning: Using 'size' aesthetic for lines was deprecated in ggplot2 3.4.0.
## i Please use 'linewidth' instead.
## This warning is displayed once every 8 hours.
## Call 'lifecycle::last_lifecycle_warnings()' to see where this warning was
## generated.
```

The log of average spectrophotometer reading for each ant

Indirect sandwich ELISA test of four different antibodies to diagnose DENV

Scales are in natural logarithmic scale **not base10



The photospectrometer reading and the
dilution factor are changed in to base log values
from -11.9 (1/156250) to -2.3 (1/10) in the x axis,
and from -1.6 (0.1946) to 0.5 (1.5995) in the y axis

```
conrol_join <- left_join(Background_noise, patients_naive_sera, by = c("Antibody", "patient"))
```

```
# Display background noise -----
```

```
elisa %>%
  filter(dilution != -Inf & Antibody != "PBS (-ve control)") %>%
  ggplot(aes(x = dilution,
             y = Absorbtion,
             color = Antibody)) +
  geom_point() +
  geom_line(size = 1.0) +
  geom_hline(data = conrol_join,
            aes(yintercept = Absorbtion, lty = "Average reading for PBS + test Ab + Detection Ab")) +
  geom_hline(data = conrol_join,
```

```

aes(yintercept = mean_ab_naive_s, lty = "Average reading for test Ab + Naiva Sera" )) +
scale_x_continuous(breaks = c(-11.9, -10.3, -8.7, -7.1, -5.5, -3.9, -2.3)) +
facet_grid(Antibody~patient) +
labs(y = "The log of average spectrophotometer reading for each antibody",
x = "The log of dilution (from dilution factor 1/156,250 to 1/10)",
title = "Indirect sandwich ELISA test of four different antibodies to diagnose DENV-3 from patient",
subtitle = "**Scales are in natural logarithmic scale **not base10**",
fill = "Antibody",
caption = "** The horizontal dashed lines are the blank wells reading where there is no antigen added but there is PBS + capture antibody and secondary detection antibodies") +
theme(axis.title = element_text(size = 12, face = "italic", color = "blue"),
plot.title = element_text(size = 12, face = "bold", color = "red"),
plot.subtitle = element_text(size = 12, face = "italic", color = "blue"),
strip.placement = "outside",
strip.text = element_text(face = "italic", color = "red", size = 10),
plot.caption = element_text(face = "italic", color = "red"))

```

