002 - Motivating Examples

EPIB 607

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Case study 1: Safety and immunogenicity of the ChAdOx1 nCoV-19 vaccine against SARS-CoV-2

Case study 2: Comparison of Estimated Rates of Coronavirus Disease 2019 (COVID-19) in Border Counties in Iowa Without a Stay-at-Home Order and Border Counties in Illinois With a Stay-at-Home Order

Early phase COVID-19 vaccine trial[1]

Safety and immunogenicity of the ChAdOx1 nCoV-19 vaccine against SARS-CoV-2: a preliminary report of a phase 1/2, single-blind, randomised controlled trial

Pedro M Folegatti, Katie J Ewer*, Parvinder K Aley, Brian Angus, Stephan Becker, Sandra Belij-Rammerstorfer, Duncan Bellamy, Sagida Bibi, Mustapha Bittaye, Elizabeth A Clutterbuck, Christina Dold, Saul N Faust, Adam Finn, Amy L Flaxman, Bassam Hallis, Paul Heath, Daniel Jenkin, Rajeka Lazarus, Rebecca Makinson, Angela M Minassian, Katrina M Pollock, Maheshi Ramasamy, Hannah Robinson, Matthew Snape, Richard Tarrant, Merryn Voysey, Catherine Green*, Alexander D Douglas*, Adrian V S Hill*, Teresa Lambe*, Sarah C Gilbert*, Andrew J Pollard*, on behalf of the Oxford COVID Vaccine Trial Group†*

Summary

Background The pandemic of severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) might be curtailed by vaccination. We assessed the safety, reactogenicity, and immunogenicity of a viral vectored coronavirus vaccine that expresses the spike protein of SARS-CoV-2.

Methods We did a phase 1/2, single-blind, randomised controlled trial in five trial sites in the UK of a chimpanzee adenovirus-vectored vaccine (ChAdOx1 nCoV-19) expressing the SARS-CoV-2 spike protein compared with a meningococcal conjugate vaccine (MenACWY) as control. Healthy adults aged 18–55 years with no history of laboratory confirmed SARS-CoV-2 infection or of COVID-19-like symptoms were randomly assigned (1:1) to receive ChAdOx1 nCoV-19 at a dose of 5×10^{10} viral particles or MenACWY as a single intramuscular injection. A protocol amendment in two of the five sites allowed prophylactic paracetamol to be administered before vaccination. Ten participants assigned to a non-randomised, unblinded ChAdOx1 nCoV-19 prime-boost group received a two-dose schedule, with the booster vaccine administered 28 days after the first dose. Humoral responses at baseline and following vaccination were assessed using a standardised total IgG ELISA against trimeric SARS-CoV-2 spike protein, a muliplexed immunoassay, three live SARS-CoV-2 neutralisation assays (a 50% plaque reduction neutralisation assay [$PRNT_{50}$]; a microneutralisation assay [MNA_{50}, MNA_{80}, and MNA_{90}]; and Marburg VN), and a pseudovirus neutralisation assay. Cellular responses were assessed using an ex-vivo interferon-γ enzyme-linked immunospot assay. The co-primary outcomes are to assess efficacy, as measured by cases of symptomatic virologically confirmed COVID-19, and safety, as measured by the occurrence of serious adverse events. Analyses were done by group allocation in participants who received the vaccine. Safety was assessed over 28 days after

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[1] https://www.thelancet.com/journals/lancet/article/PIIS0140-6736(20)31604-4/fulltext

Phase 1/2 trial

- The focus in phase 1/2 trials is looking at what the vaccine does to the body and what the body does with the vaccine in *healthy* individuals
- Adults with no history of laboratory confirmed SARS-CoV-2 infection or of COVID-19-like symptoms were randomly assigned (1:1) to receive **ChAdOx1 nCoV-19** or **MenACWY** (Meningococcal) as a single intramuscular injection
- Convalescent plasma samples from adults with PCR-positive SARS-CoV-2 infection were obtained from symptomatic patients admitted to the hospitals to characterize the immunological properties of COVID-19[2]
- The enzyme-linked immunosorbent assay (ELISA) technique was used to detect antibodies (i.e. levels of immunity)

[2] Convalescent plasma is collected from someone who has recovered from a virus. When a person is infected with a virus, their body starts making antibodies to fight it. It is believed these antibodies could be the key ingredient for a treatment to help others with the same virus.

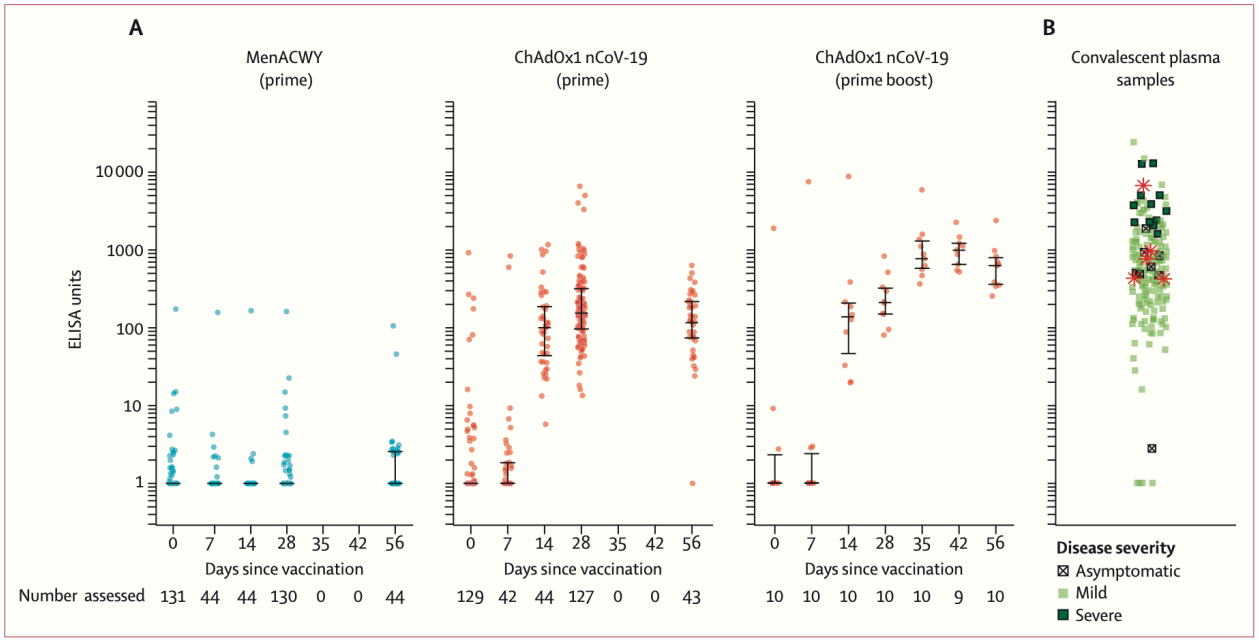

Figure 3: **SARS-CoV-2 IgG response by standardised ELISA to spike protein in trial participants (A) and in 180 convalescent plasma samples from 172 patients with PCR-confirmed COVID-19 and eight asymptomatic health-care workers (B)**

Error bars show median (IQR). Participants in the prime boost group received their second dose at day 28. Lower limit of quantification is 1 ELISA unit. Red stars in panel B show five samples also tested on the Marburg VN assay (see figure 4). MenACWY=meningococcal group A, C, W-135, and Y conjugate vaccine. SARS-CoV-2=severe acute respiratory syndrome coronavirus 2.

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1. What levels of immunity are found in patients who have recovered from COVID-19? (panel B)
2. Relative to these what levels of immunity are found in persons who have received the ChAdOx1 nCoV-19 vaccine? Compare panel A (prime, 28 days) vs panel B.

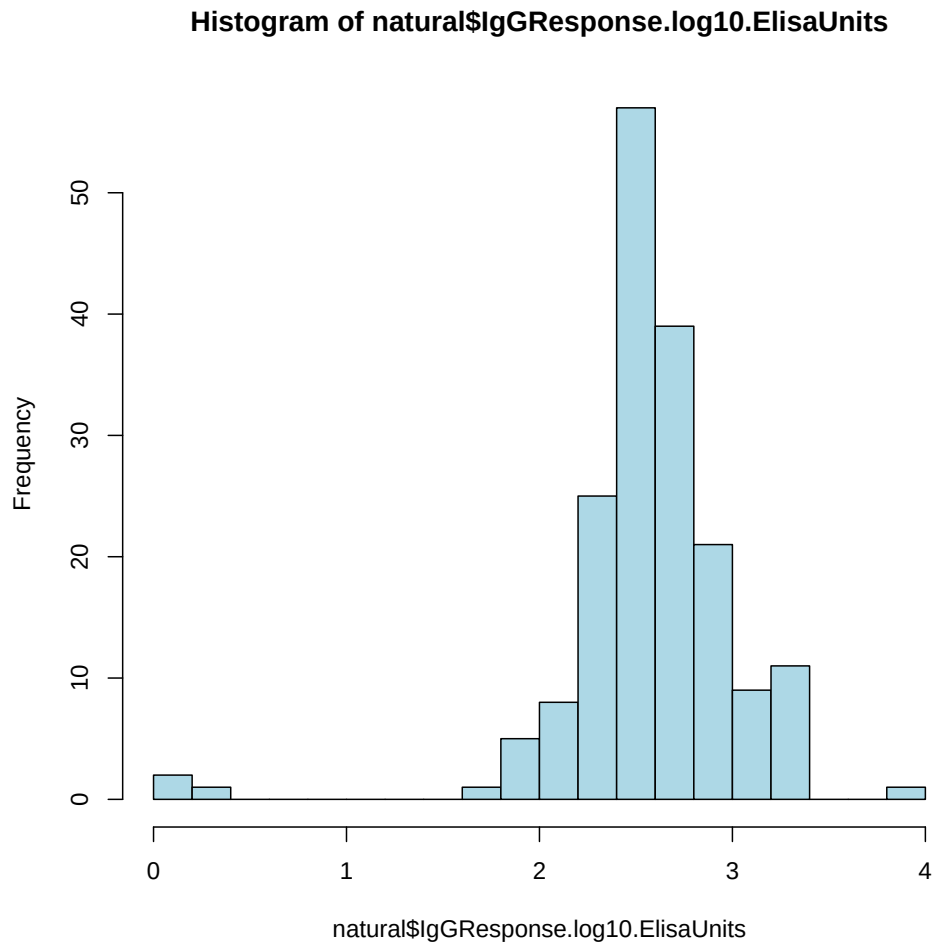
What levels of immunity are found in patients who have recovered from COVID-19?[3]

```
path <-
  "http://www.biostat.mcgill.ca/hanley/statbook/immunogenicityChAdOx1.nCoV-19vaccine.txt"
ds <- read.table(path)
ds$RefIndexCategory <- factor(ds$RefIndexCategory)
head(ds)

##   RefIndexCategory IgGResponse.log10.ElisaUnits
## 1     Convalescent                         2.56
## 2     Convalescent                         2.74
## 3     Convalescent                         2.79
## 4     Convalescent                         3.32
## 5     Convalescent                         3.15
## 6     Convalescent                         2.35

str(ds)

## 'data.frame':^^I307 obs. of  2 variables:
##  $ RefIndexCategory           : Factor w/ 2 levels "Convalescent",..: 1 1 1 1 1 1 1 1 1 1 ...
##  $ IgGResponse.log10.ElisaUnits: num  2.56 2.74 2.79 3.32 3.15 2.35 2.72 2.95 2.42 2.64 ...

levels(ds$RefIndexCategory)

## [1] "Convalescent"             "Day28PostChAdOx1 nCoV-19"
```

[3] Data were (imperfectly) scraped from the Postscript file "behind" the pdf file by Dr. Hanley

What levels of immunity are found in patients who have recovered from COVID-19?

```
natural <- ds[ds$RefIndexCategory=="Convalescent",]
hist(natural$IgGResponse.log10.ElisaUnits,
     breaks = 20, col = "lightblue")
```

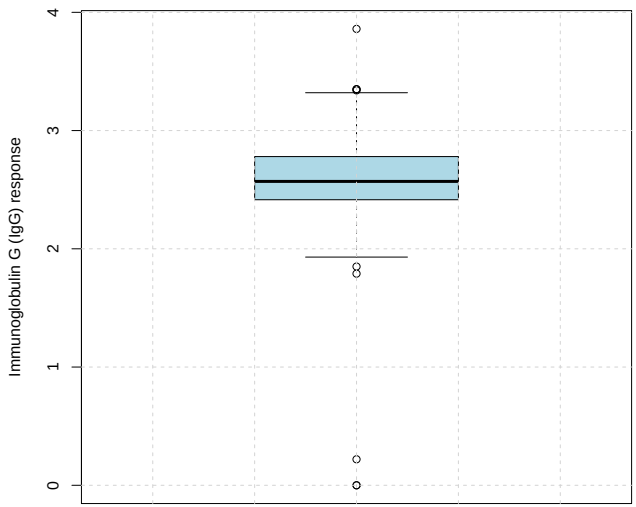


Three different methods of calculating the mean

```r
summary(natural$IgGResponse.log10.ElisaUnits)
```

```
##    Min. 1st Qu.  Median    Mean 3rd Qu.    Max.
##   0.000   2.417   2.570   2.577   2.780   3.860
```

```r
boxplot(natural$IgGResponse.log10.ElisaUnits,
        col = "lightblue",
        ylab = "Immunoglobulin G (IgG) response")
grid(lty = "dashed")
```




```r
t.test(natural$IgGResponse.log10.ElisaUnits)
```

```
## One Sample t-test with natural$IgGResponse.log10.ElisaUnits
## t = 75.0898, df = 179, p-value < 2.2e-16
## alternative hypothesis: true mean is not equal to 0
## 95 percent confidence interval:
##  2.509603 2.645064
## sample estimates:
## mean of x
##  2.577333
```

```r
fit1 <- glm(IgGResponse.log10.ElisaUnits ~ 1, data = natural)
summary(fit1)
```

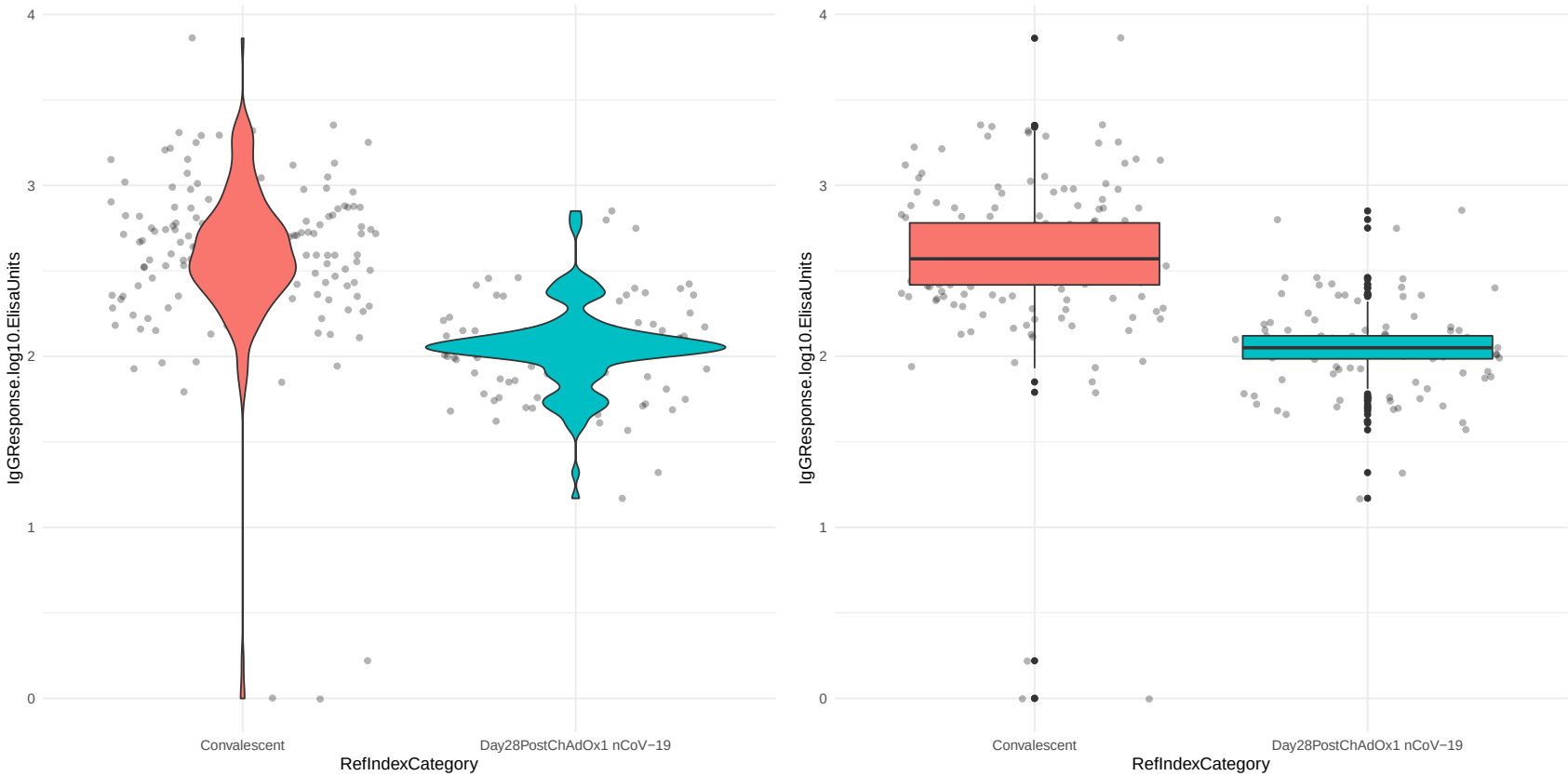
```
##
## Coefficients:
##             Estimate Std. Error t value Pr(>|t|)
## (Intercept)  2.57733    0.03432   75.09   <2e-16 ***
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
##
## (Dispersion parameter for gaussian family taken to be 0.2120565)
##
##     Null deviance: 37.958  on 179  degrees of freedom
## Residual deviance: 37.958  on 179  degrees of freedom
## AIC: 234.65
##
## Number of Fisher Scoring iterations: 2
```

```r
confint(fit1)
```

```
##    2.5 %   97.5 %
## 2.510061 2.644606
```

Naturally vs. vaccine-induced response levels

```
p1 <- ggplot(data = ds, mapping = aes(x = RefIndexCategory, y = IgGResponse.log10.ElisaUnits,
    fill = RefIndexCategory)) + geom_jitter(alpha = 0.3) + theme_minimal() + theme(legend.position = "none")
p1 + geom_violin()
p1 + geom_boxplot()
```



(a) Violin plot

(b) Boxplot

Comparing means using classic methods

1. Numerical summary

```
base::by(ds$IgGResponse.log10.ElisaUnits,ds$RefIndexCategory,summary)

## ds$RefIndexCategory: Convalescent
##    Min. 1st Qu.  Median    Mean 3rd Qu.    Max.
##   0.000   2.417   2.570   2.577   2.780   3.860
## ------------------------------------------------------------
## ds$RefIndexCategory: Day28PostChAdOx1 nCoV-19
##    Min. 1st Qu.  Median    Mean 3rd Qu.    Max.
##   1.170   1.985   2.050   2.047   2.120   2.850
```

2. Another "dot" test

```
stats::t.test(IgGResponse.log10.ElisaUnits ~ RefIndexCategory, data = ds)

## Welch Two Sample t-test with IgGResponse.log10.ElisaUnits by RefIndexCategory
## t = 13.1047, df = 284.781, p-value < 2.2e-16
## alternative hypothesis: true difference in means is not equal to 0
## 95 percent confidence interval:
##  0.4510720 0.6105238
## sample estimates:
##             mean in group Convalescent mean in group Day28PostChAdOx1 nCoV-19
##                               2.577333                               2.046535
```

Comparing means using regression

3. Regression

```
fit2 <- glm(IgGResponse.log10.ElisaUnits ~ RefIndexCategory, data = ds)
print(summary(fit2), signif.star = FALSE)
```

```
##
## Coefficients:
##                                        Estimate Std. Error t value Pr(>|t|)
## (Intercept)                             2.57733    0.02874   89.67   <2e-16
## RefIndexCategoryDay28PostChAdOx1 nCoV-19 -0.53080    0.04469  -11.88   <2e-16
##
## (Dispersion parameter for gaussian family taken to be 0.1487187)
##
##     Null deviance: 66.339  on 306  degrees of freedom
## Residual deviance: 45.359  on 305  degrees of freedom
## AIC: 290.17
##
## Number of Fisher Scoring iterations: 2
```

```
confint(fit2)
```

```
##                                           2.5 %     97.5 %
## (Intercept)                           2.5209962  2.6336704
## RefIndexCategoryDay28PostChAdOx1 nCoV-19 -0.6183894 -0.4432064
```

Fitted regression line

```
plot(ds$RefIndexCategory, ds$IgGResponse.log10.ElisaUnits, pch=19, cex=0.5)
abline(h = seq(0,4,0.5),col = "lightblue")
lines(ds$RefIndexCategory, fit2$fitted.values, col = "red", lwd = 3)
```

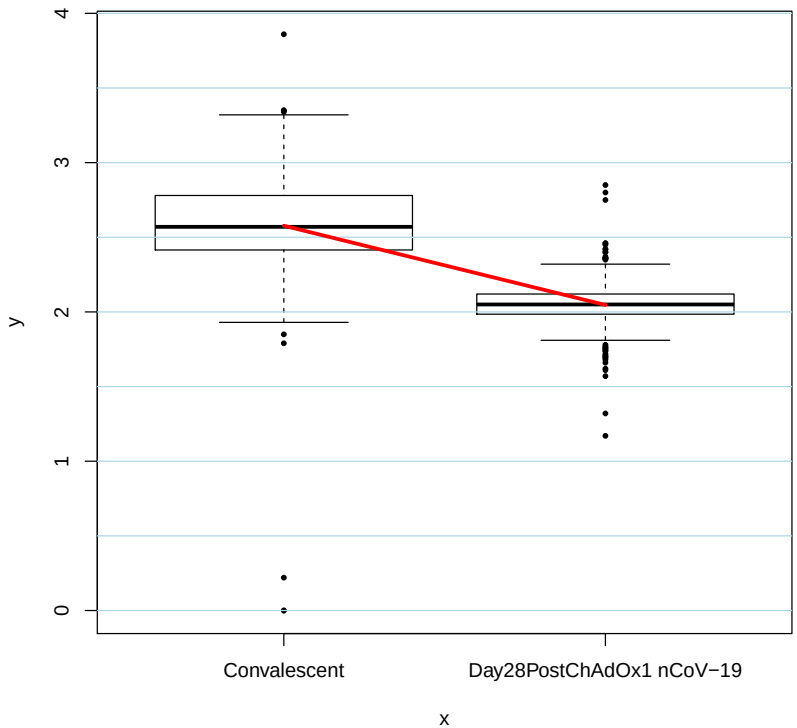


Figure: The red line is the fitted regression from the previous slide.

Case study 1: Safety and immunogenicity of the ChAdOx1 nCoV-19 vaccine against SARS-CoV-2

Case study 2: Comparison of Estimated Rates of Coronavirus Disease 2019 (COVID-19) in Border Counties in Iowa Without a Stay-at-Home Order and Border Counties in Illinois With a Stay-at-Home Order

Comparing Iowa and Illinois Cases[4]

JAMA Network Open

Original Investigation | Public Health

Comparison of Estimated Rates of Coronavirus Disease 2019 (COVID-19) in Border Counties in Iowa Without a Stay-at-Home Order and Border Counties in Illinois With a Stay-at-Home Order

Wei Lyu, MS; George L. Wehby, PhD

Abstract

IMPORTANCE Iowa is 1 of 5 states in the US that have not issued a stay-at-home order during the coronavirus disease 2019 (COVID-19) pandemic. There is no empirical evidence on whether issuing a stay-at-home order in Iowa could have been associated with a reduced rate of COVID-19 infections in the state.

OBJECTIVE To compare COVID-19 cases in border counties in Iowa, which did not issue a stay-at-home order, with cases in border counties in Illinois, which did issue a stay-at-home order.

DESIGN, SETTING, AND PARTICIPANTS This cross-sectional study with a difference-in-differences design compared daily changes in COVID-19 cases per 10 000 residents in 8 Iowa counties bordering Illinois with those in the 7 Illinois counties bordering Iowa before and after Illinois issued a stay-at-home order on March 21, 2020. Additional sensitivity analyses were conducted to account for differences in timing of closing schools and nonessential businesses between the 2 states and differential trends in COVID-19 cases by county population density and poverty rates.

Key Points

Question Was the stay-at-home order in Illinois associated with different rates of coronavirus disease 2019 (COVID-19) compared with Iowa, which did not issue a stay-at-home order?

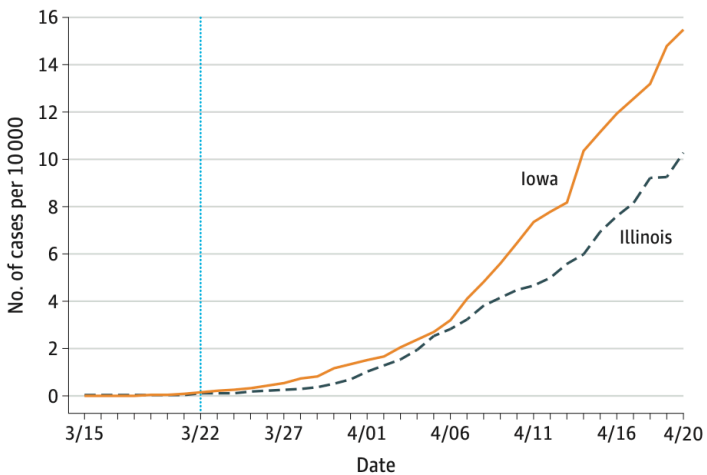
Findings This cross-sectional study of border counties in Iowa and Illinois used difference-in-differences design and found an increase in estimated rates of COVID-19 cases per 10 000 residents in the border counties in Iowa compared with the border counties in Illinois after a stay-at-home order was implemented in Illinois but not in Iowa.

[4] https://jamanetwork.com/journals/jamanetworkopen/fullarticle/2766229

Are the difference in curves real? Or just random variation?

- This study compared COVID-19 cases in border counties in Iowa, which did not issue a stay-at-home order, with cases in border counties in Illinois, which did issue a stay-at-home order.

Figure 1. Cumulative Coronavirus Disease 2019 (COVID-19) Cases per 10 000 Residents in Iowa and Illinois Border Counties



The vertical line represents the date on which the stay-at-home order took effect in Illinois.

JAMA Network Open. 2020;3(5):e2011102. doi:10.1001/jamanetworkopen.2020.11102 May 15, 2020 3/7

Freely available county level data from NYTimes[5]

```
library(covdata) # remotes::install_github("kjhealy/covdata")
library(dplyr); library(tidyr); library(ggplot2); library(readr)

# get population data from https://covid19.census.gov/datasets/
pop_county <- read_csv("https://opendata.arcgis.com/datasets/21843f238cbb46b08615fc53e19e0daf_1.csv") %>%
              dplyr::rename(fips = GEOID, population = B01001_001E, state = State) %>%
              dplyr::select(state, fips, population)

county_level <- nytcovcounty %>%
                dplyr::left_join(pop_county, by = c("state","fips")) %>%
                dplyr::mutate(cases.per.10k = cases/population * 1e4) %>%
                dplyr::filter(state %in% c("Iowa","Illinois")) %>%
                dplyr::group_by(county) %>%
                dplyr::filter(date >= "2020-03-15") %>%
                dplyr::filter(date <= "2020-09-01")

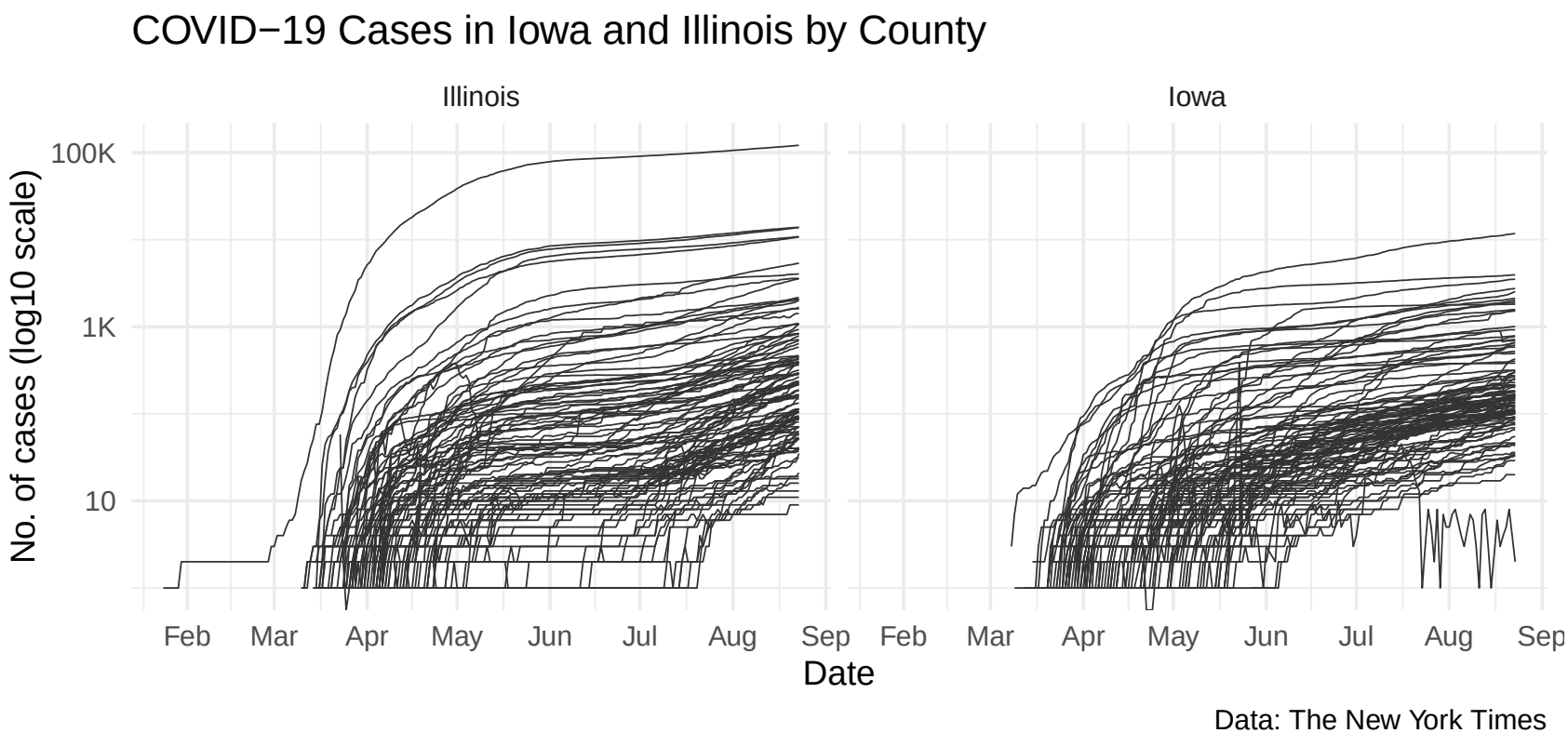
pop_state <- pop_county %>%
             dplyr::group_by(state) %>%
             dplyr::summarise(population = sum(population, na.rm = TRUE))

state_level <- county_level %>%
               dplyr::group_by(state, date) %>%
               dplyr::filter(date >= "2020-03-15") %>%
               dplyr::filter(date <= "2020-09-01") %>%
               dplyr::summarise(cases = sum(cases)) %>%
               dplyr::left_join(pop_state, by = "state") %>%
               dplyr::mutate(cases.per.10k = cases / population * 1e4, state = factor(state),
                             time = as.numeric(date - min(date)) + 1)
head(state_level)

## # A tibble: 6 x 6
## # Groups:   state [1]
##   state    date       cases population cases.per.10k  time
##   <fct>    <date>     <dbl>      <dbl>         <dbl> <dbl>
## 1 Illinois 2020-03-15    94   12821497        0.0733     1
## 2 Illinois 2020-03-16   104   12821497        0.0811     2
## 3 Illinois 2020-03-17   159   12821497        0.124      3
## 4 Illinois 2020-03-18   286   12821497        0.223      4
## 5 Illinois 2020-03-19   420   12821497        0.328      5
## 6 Illinois 2020-03-20   583   12821497        0.455      6
```

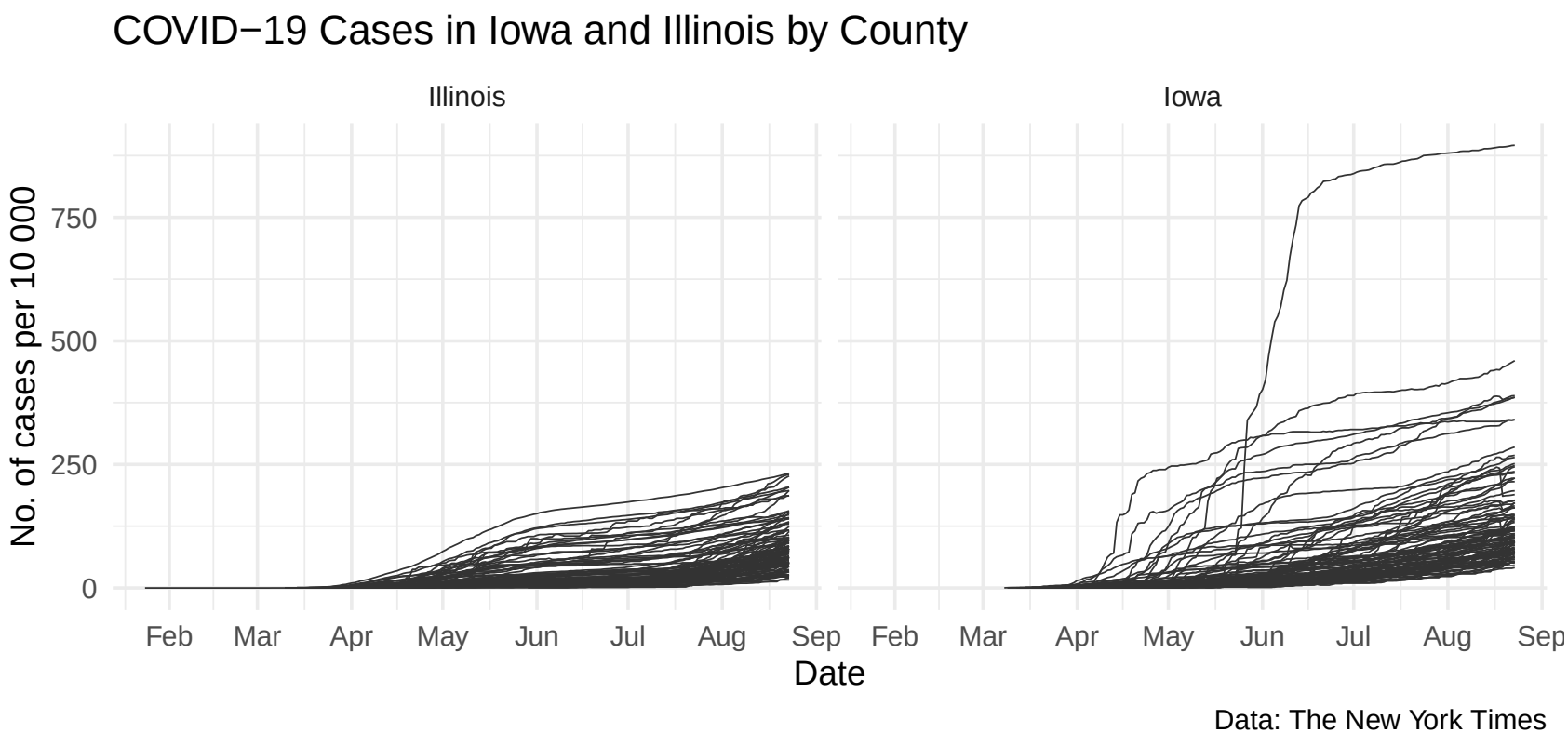
County level cases for Iowa and Illinois - log10 scale

```
ggplot(data = county_level, mapping = aes(x = date, y = cases, group = county)) +
  geom_line(size = 0.25, color = "gray20") +
  scale_x_date(date_breaks = "1 month", date_labels = "%b")+
  scale_y_log10(labels = scales::label_number_si()) +
  guides(color = FALSE) + facet_wrap(~ state, ncol = 2) +
  labs(title = "COVID-19 Cases in Iowa and Illinois by County",
       x = "Date", y = "No. of cases (log10 scale)", caption = "Data: The New York Times") +
  theme_minimal()
```



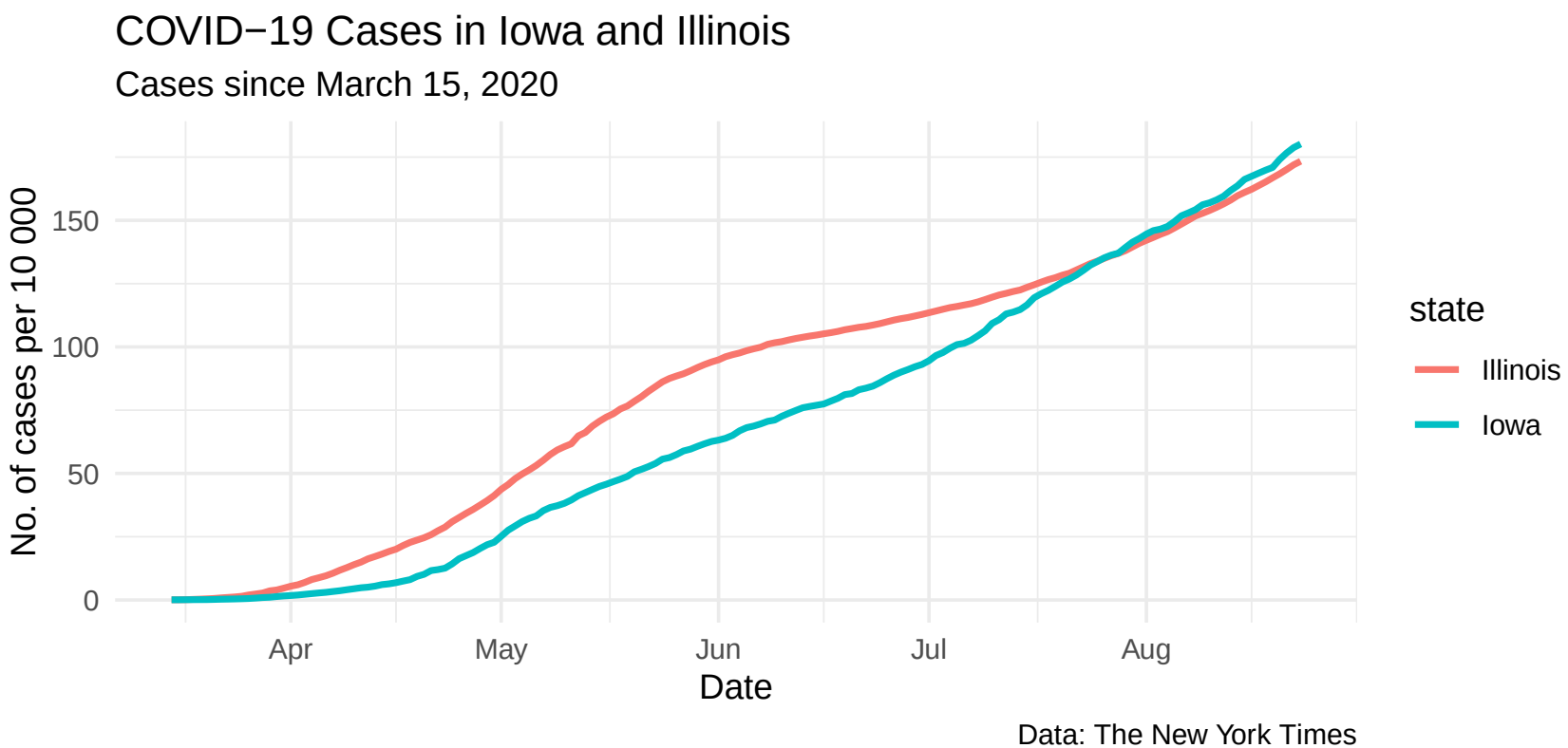
County level cases for Iowa and Illinois - per capita

```
ggplot(data = county_level, mapping = aes(x = date, y = cases.per.10k, group = county)) +
  geom_line(size = 0.25, color = "gray20") +
  scale_x_date(date_breaks = "1 month", date_labels = "%b")+
  scale_y_continuous(labels = scales::label_number_si()) +
  guides(color = FALSE) + facet_wrap(~ state, ncol = 2) +
  labs(title = "COVID-19 Cases in Iowa and Illinois by County",
       x = "Date", y = "No. of cases per 10 000", caption = "Data: The New York Times") +
  theme_minimal()
```



State level cases for Iowa and Illinois - per capita

```
ggplot(data = state_level, mapping = aes(x = date, y = cases.per.10k, color = state)) +
  geom_line(size = 1) +
  scale_x_date(date_breaks = "1 month", date_labels = "%b")+
  scale_y_continuous(labels = scales::label_number_si()) +
  labs(title = "COVID-19 Cases in Iowa and Illinois",
       subtitle = "Cases since March 15, 2020",
       x = "Date", y = "No. of cases per 10 000", caption = "Data: The New York Times") +
  theme_minimal()
```



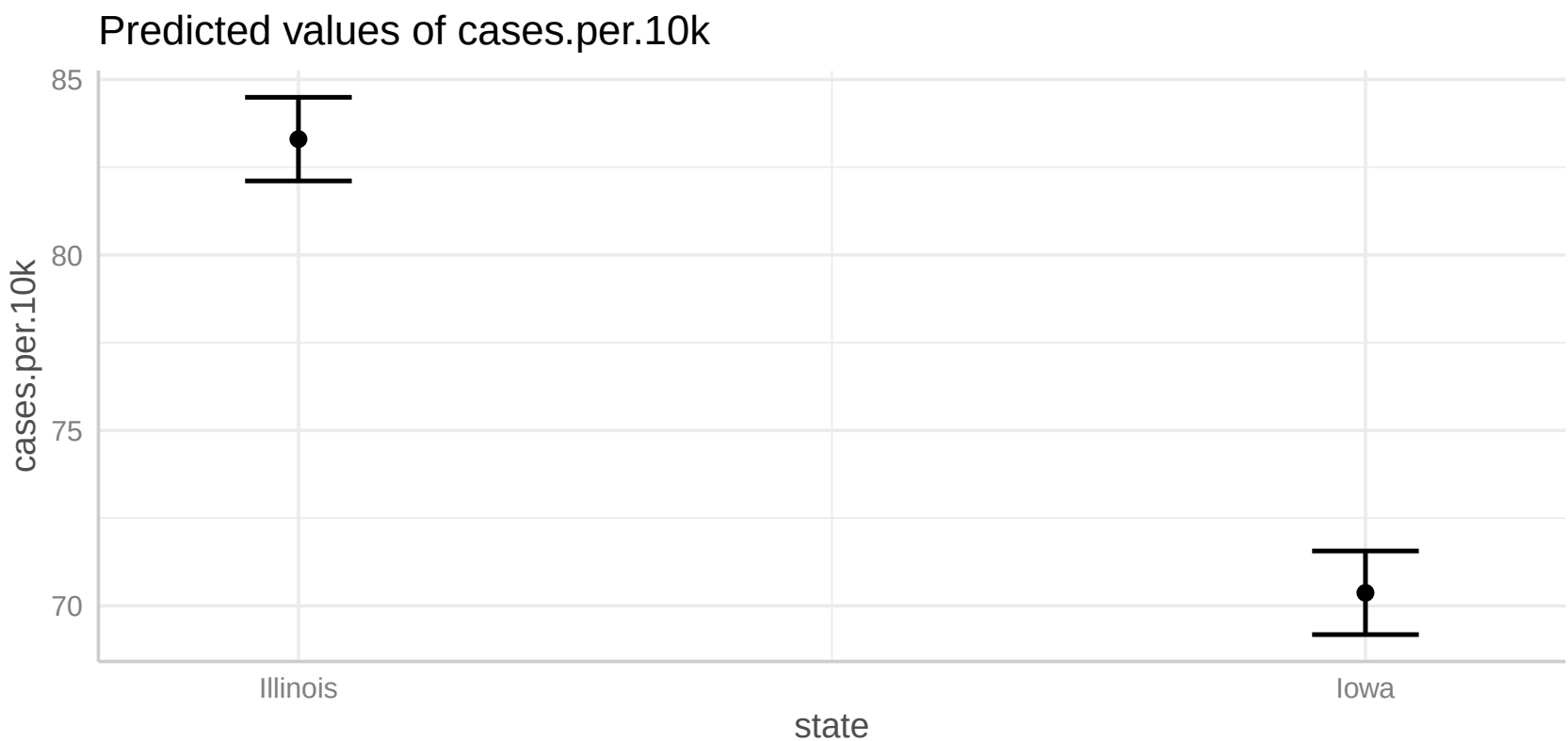
Are the findings in the paper reproducible?

```
fit3 <- glm(cases.per.10k ~ state*time, data = state_level)
summary(fit3)
```

```
##
## Coefficients:
##                 Estimate Std. Error t value Pr(>|t|)
## (Intercept)     -7.33863    1.32302  -5.547 5.88e-08 ***
## stateIowa      -20.32020    1.87103 -10.860  < 2e-16 ***
## time             1.11358    0.01334  83.462  < 2e-16 ***
## stateIowa:time   0.10422    0.01887   5.523 6.65e-08 ***
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
##
## (Dispersion parameter for gaussian family taken to be 74.17442)
##
##     Null deviance: 1170732  on 341  degrees of freedom
## Residual deviance:   25071  on 338  degrees of freedom
## AIC: 2449.3
##
## Number of Fisher Scoring iterations: 2
```

Model-based predictions

```
pacman::p_load(ggeffects)
ggeffects::ggpredict(fit3, terms = "state") %>%
  plot()
```



Session Info

```
R version 4.0.2 (2020-06-22)
Platform: x86_64-pc-linux-gnu (64-bit)
Running under: Pop!_OS 20.10

Matrix products: default
BLAS:   /usr/lib/x86_64-linux-gnu/openblas-pthread/libblas.so.3
LAPACK: /usr/lib/x86_64-linux-gnu/openblas-pthread/libopenblasp-r0.3.10.so

attached base packages:
[1] tools     stats     graphics  grDevices utils     datasets  methods
[8] base

other attached packages:
 [1] ggeffects_0.16.0 covdata_0.8      here_0.1         NCStats_0.4.7
 [5] FSA_0.8.30       forcats_0.5.1    stringr_1.4.0    dplyr_1.0.7
 [9] purrr_0.3.4      readr_1.4.0      tidyr_1.1.3      tibble_3.1.3
[13] ggplot2_3.3.5    tidyverse_1.3.0  knitr_1.33

loaded via a namespace (and not attached):
 [1] fs_1.5.0           lubridate_1.7.9    insight_0.9.6      httr_1.4.2
 [5] rprojroot_2.0.2    backports_1.2.1    sjlabelled_1.1.7   utf8_1.2.2
 [9] R6_2.5.1           DBI_1.1.1          colorspace_2.0-2   withr_2.4.2
[13] tidyselect_1.1.1   gridExtra_2.3      leaflet_2.0.3      curl_4.3.2
[17] compiler_4.0.2     cli_3.0.1          rvest_1.0.0        formatR_1.8
[21] pacman_0.5.1       xml2_1.3.2         ggdendro_0.1.22    labeling_0.4.2
[25] mosaicCore_0.8.0   scales_1.1.1       digest_0.6.27      ggformula_0.9.4
[29] foreign_0.8-80     rio_0.5.16         pkgconfig_2.0.3    htmltools_0.5.1.1
[33] highr_0.9          dbplyr_1.4.4       htmlwidgets_1.5.3  rlang_0.4.11
[37] readxl_1.3.1       rstudioapi_0.13    farver_2.1.0       generics_0.1.0
[41] jsonlite_1.7.2     crosstalk_1.1.1    zip_2.2.0          car_3.0-9
[45] magrittr_2.0.1     mosaicData_0.20.1  Matrix_1.2-18      Rcpp_1.0.7
[49] munsell_0.5.0      fansi_0.5.0        abind_1.4-5        lifecycle_1.0.0
[53] stringi_1.7.3      snakecase_0.11.0   carData_3.0-4      MASS_7.3-53
[57] plyr_1.8.6         ggstance_0.3.4     grid_4.0.2         blob_1.2.1
[61] ggrepel_0.8.2      crayon_1.4.1       lattice_0.20-41    haven_2.3.1
[65] splines_4.0.2      hms_1.0.0          pillar_1.6.2       reprex_0.3.0
[69] glue_1.4.2         evaluate_0.14      data.table_1.14.0  modelr_0.1.8
[73] vctrs_0.3.8        tweenr_1.0.1       cellranger_1.1.0   gtable_0.3.0
[77] polyclip_1.10-0    assertthat_0.2.1   TeachingDemos_2.12 xfun_0.25
[81] ggforce_0.3.2      openxlsx_4.1.5     broom_0.7.2        mosaic_1.7.0
[85] ellipsis_0.3.2
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