

Pooled testing with penalized regression models

Christopher R. Bilder
University of Nebraska–Lincoln
Department of Statistics
chris@chrisbilder.com

Research is supported by NIH grant R01 AI121351

Joint work with
Pranta Das at University of Nebraska-Lincoln,
Joshua M. Tebbs at University of South Carolina, and
Christopher S. McMahan at Clemson University

- Infectious disease testing

- Timely
- Efficient

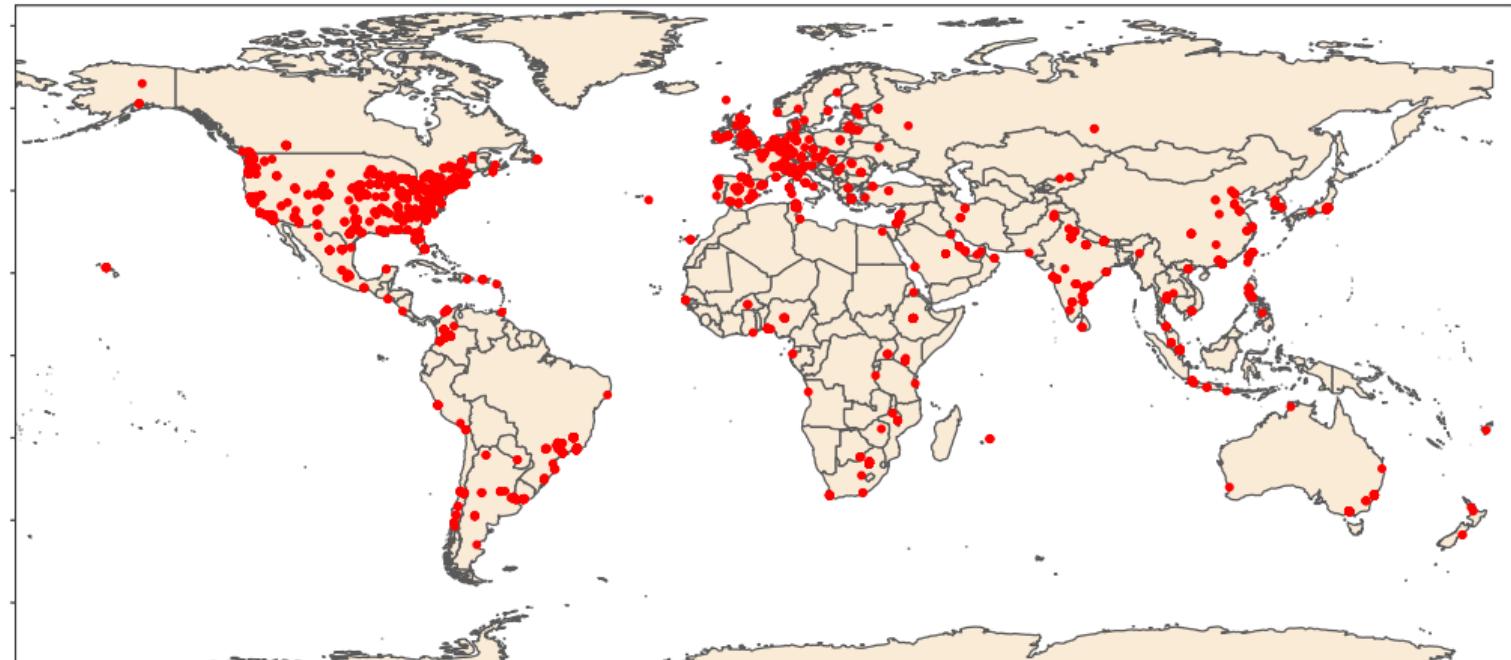
- Infectious disease testing
 - Timely
 - Efficient
- COVID-19 pandemic

- Infectious disease testing
 - Timely
 - Efficient
- COVID-19 pandemic
- Pooled testing
 - Also known as “group testing”
 - Process in Nebraska (Abdalhamid et al. 2020)

- Infectious disease testing
 - Timely
 - Efficient
- COVID-19 pandemic
- Pooled testing
 - Also known as “group testing”
 - Process in Nebraska (Abdalhamid et al. 2020)
 - Combine together portions of 5 specimens from different individuals into a “pool”
 - Test as if it were a single specimen
 - Pool tests negative: All 5 individuals are negative
 - Pool tests positive: Retest each individual separately to determine who is positive or negative

- Infectious disease testing
 - Timely
 - Efficient
- COVID-19 pandemic
- Pooled testing
 - Also known as “group testing”
 - Process in Nebraska (Abdalhamid et al. 2020)
 - Combine together portions of 5 specimens from different individuals into a “pool”
 - Test as if it were a single specimen
 - Pool tests negative: All 5 individuals are negative
 - Pool tests positive: Retest each individual separately to determine who is positive or negative
 - Decrease number of tests, increase testing capacity

- Widely used during pandemic
 - A Shiny App for Pooled Testing
 - 91 countries during 6 months of 2020



- Australia

- App: 73 separate visits from Sydney!



- Australia

- App: 73 separate visits from Sydney!



- Department of Health, Disability and Ageing: "Revised testing framework for COVID-19 in Australia, March 2022"

- Australia

- App: 73 separate visits from Sydney!



- Department of Health, Disability and Ageing: "Revised testing framework for COVID-19 in Australia, March 2022"
 - Microbiological Diagnostic Unit Public Health Lab at U. of Melbourne (Chong et al. 2020)

- Australia

- App: 73 separate visits from Sydney!



- Department of Health, Disability and Ageing: "Revised testing framework for COVID-19 in Australia, March 2022"
 - Microbiological Diagnostic Unit Public Health Lab at U. of Melbourne (Chong et al. 2020)
- Widely used elsewhere: Blood donations, sexually transmitted infections, congenital infections, animal infections, food safety surveillance, computer networks assessments, flower infection levels

- Algorithmic process

- Dorfman testing for previous example: Stage 1 test in pool, Stage 2 test separately (if needed)

• Algorithmic process

- Dorfman testing for previous example: Stage 1 test in pool, Stage 2 test separately (if needed)
- Different pool sizes
- Other algorithms exist

• Algorithmic process

- Dorfman testing for previous example: Stage 1 test in pool, Stage 2 test separately (if needed)
- Different pool sizes
- Other algorithms exist
- Statistician involvement: Efficiency (expected number of tests per individual) is comparison metric

- Algorithmic process

- Dorfman testing for previous example: Stage 1 test in pool, Stage 2 test separately (if needed)
- Different pool sizes
- Other algorithms exist
- Statistician involvement: Efficiency (expected number of tests per individual) is comparison metric

- During COVID-19 pandemic

- Over 1 GB of papers published on pooled testing during the first two years of pandemic!

- Algorithmic process

- Dorfman testing for previous example: Stage 1 test in pool, Stage 2 test separately (if needed)
- Different pool sizes
- Other algorithms exist
- Statistician involvement: Efficiency (expected number of tests per individual) is comparison metric

- During COVID-19 pandemic

- Over 1 GB of papers published on pooled testing during the first two years of pandemic!
- Two new innovative algorithms developed
 - Shental et al. (2020), Ghosh et al. (2021), Zismanov et al. (2024)
 - Non-statistical journal papers

- Algorithmic process

- Dorfman testing for previous example: Stage 1 test in pool, Stage 2 test separately (if needed)
- Different pool sizes
- Other algorithms exist
- Statistician involvement: Efficiency (expected number of tests per individual) is comparison metric

- During COVID-19 pandemic

- Over 1 GB of papers published on pooled testing during the first two years of pandemic!
- Two new innovative algorithms developed
 - Shental et al. (2020), Ghosh et al. (2021), Zismanov et al. (2024)
 - Non-statistical journal papers
- Use viral load responses rather than binary (positive/negative) responses
- Use linear model to predict positive/negative

- Algorithmic process

- Dorfman testing for previous example: Stage 1 test in pool, Stage 2 test separately (if needed)
- Different pool sizes
- Other algorithms exist
- Statistician involvement: Efficiency (expected number of tests per individual) is comparison metric

- During COVID-19 pandemic

- Over 1 GB of papers published on pooled testing during the first two years of pandemic!
- Two new innovative algorithms developed
 - Shental et al. (2020), Ghosh et al. (2021), Zismanov et al. (2024)
 - Non-statistical journal papers
- Use viral load responses rather than binary (positive/negative) responses
- Use linear model to predict positive/negative
- Want to avoid retesting in a second stage

- Algorithmic process

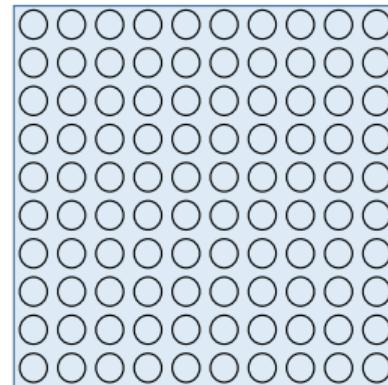
- Dorfman testing for previous example: Stage 1 test in pool, Stage 2 test separately (if needed)
- Different pool sizes
- Other algorithms exist
- Statistician involvement: Efficiency (expected number of tests per individual) is comparison metric

- During COVID-19 pandemic

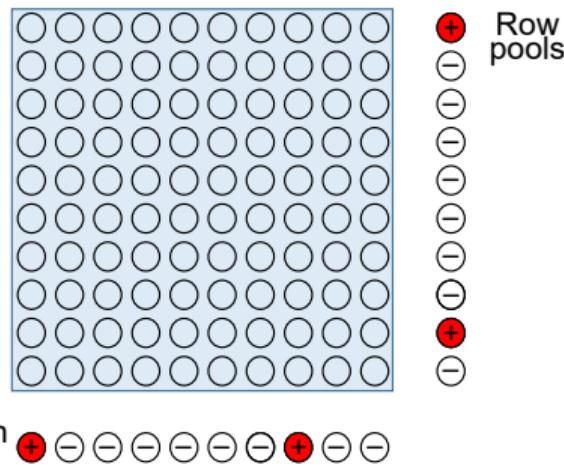
- Over 1 GB of papers published on pooled testing during the first two years of pandemic!
- Two new innovative algorithms developed
 - Shental et al. (2020), Ghosh et al. (2021), Zismanov et al. (2024)
 - Non-statistical journal papers
- Use viral load responses rather than binary (positive/negative) responses
- Use linear model to predict positive/negative
- Want to avoid retesting in a second stage

- Purpose: Examine use of viral load response and linear model prediction with “array testing” algorithm

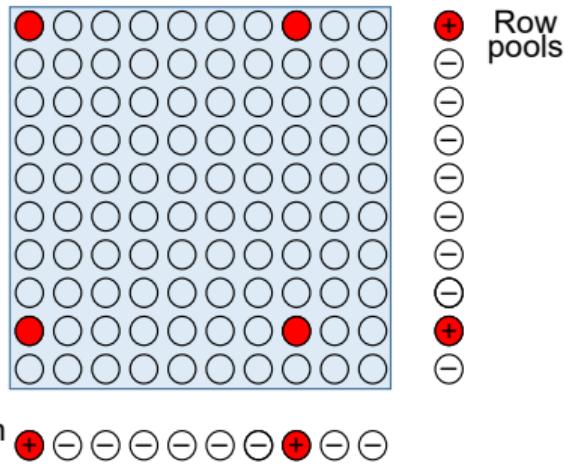
● Array testing



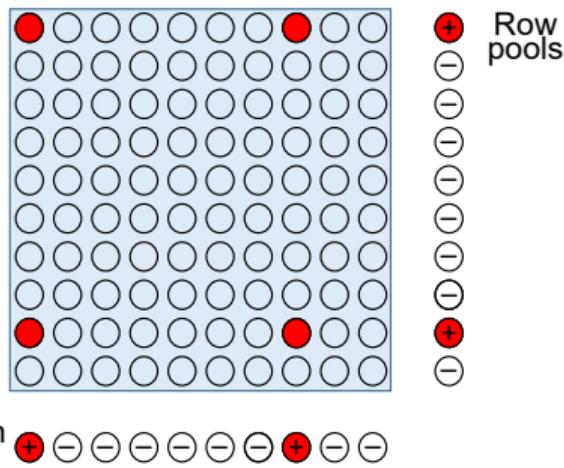
• Array testing



• Array testing



- Array testing



- **Key aspect:** Test in multiple pools (groups) during first stage to reduce the number of retests in a second stage

- A 3×3 array

	Column 1	Column 2	Column 3
Row 1	1	2	3
Row 2	4	5	6
Row 3	7	8	9

- A 3×3 array

	Column 1	Column 2	Column 3
Row 1	1	2	3
Row 2	4	5	6
Row 3	7	8	9

- Alternative form

Pools	Specimens									Regular Array
	1	2	3	4	5	6	7	8	9	
1	1	1	1	0	0	0	0	0	0	Row 1
2	0	0	0	1	1	1	0	0	0	Row 2
3	0	0	0	0	0	0	1	1	1	Row 3
4	1	0	0	1	0	0	1	0	0	Col. 1
5	0	1	0	0	1	0	0	1	0	Col. 2
6	0	0	1	0	0	1	0	0	1	Col. 3

- A 3×3 array

	Column 1	Column 2	Column 3
Row 1	1	2	3
Row 2	4	5	6
Row 3	7	8	9

- Alternative form

Pools	Specimens									Regular Array
	1	2	3	4	5	6	7	8	9	
1	1	1	1	0	0	0	0	0	0	Row 1
2	0	0	0	1	1	1	0	0	0	Row 2
3	0	0	0	0	0	0	1	1	1	Row 3
4	1	0	0	1	0	0	1	0	0	Col. 1
5	0	1	0	0	1	0	0	1	0	Col. 2
6	0	0	1	0	0	1	0	0	1	Col. 3

- Pooling matrix: $X_{6 \times 9}$ is a matrix of 0's and 1's

- Could we use a linear model to predict positives/negatives rather than going to a second stage?

- Could we use a linear model to predict positives/negatives rather than going to a second stage?
- Define
 - R = Number of rows of array, C = Number of columns of array
 - $\mathbf{Y} = (Y_1, \dots, Y_{RC})'$, a vector of viral loads for the pools; observable
 - $\boldsymbol{\beta} = (\beta_1, \dots, \beta_{RC})'$, a vector of true individual viral loads; not observable

- Could we use a linear model to predict positives/negatives rather than going to a second stage?
- Define
 - R = Number of rows of array, C = Number of columns of array
 - $\mathbf{Y} = (Y_1, \dots, Y_{RC})'$, a vector of viral loads for the pools; observable
 - $\boldsymbol{\beta} = (\beta_1, \dots, \beta_{RC})'$, a vector of true individual viral loads; not observable
- $E(\mathbf{Y}) = \mathbf{X}\boldsymbol{\beta}$
 - A linear model!
 - Pool viral loads are sums of individual viral loads

- Could we use a linear model to predict positives/negatives rather than going to a second stage?
- Define
 - R = Number of rows of array, C = Number of columns of array
 - $\mathbf{Y} = (Y_1, \dots, Y_{RC})'$, a vector of viral loads for the pools; observable
 - $\boldsymbol{\beta} = (\beta_1, \dots, \beta_{RC})'$, a vector of true individual viral loads; not observable
- $E(\mathbf{Y}) = \mathbf{X}\boldsymbol{\beta}$
 - A linear model!
 - Pool viral loads are sums of individual viral loads
 - β_i
 - Equal 0: Specimen has no virus, negative individual
 - Greater than 0: Specimen has virus, positive individual

- Could we use a linear model to predict positives/negatives rather than going to a second stage?
- Define
 - R = Number of rows of array, C = Number of columns of array
 - $\mathbf{Y} = (Y_1, \dots, Y_{RC})'$, a vector of viral loads for the pools; observable
 - $\boldsymbol{\beta} = (\beta_1, \dots, \beta_{RC})'$, a vector of true individual viral loads; not observable
- $E(\mathbf{Y}) = \mathbf{X}\boldsymbol{\beta}$
 - A linear model!
 - Pool viral loads are sums of individual viral loads
 - β_i
 - Equal 0: Specimen has no virus, negative individual
 - Greater than 0: Specimen has virus, positive individual
- Fit model to estimate $\boldsymbol{\beta}$
 - Assume $MVN(0, \sigma_y^2 \mathbf{I})$ for \mathbf{Y}

- Could we use a linear model to predict positives/negatives rather than going to a second stage?
- Define
 - R = Number of rows of array, C = Number of columns of array
 - $\mathbf{Y} = (Y_1, \dots, Y_{RC})'$, a vector of viral loads for the pools; observable
 - $\boldsymbol{\beta} = (\beta_1, \dots, \beta_{RC})'$, a vector of true individual viral loads; not observable
- $E(\mathbf{Y}) = \mathbf{X}\boldsymbol{\beta}$
 - A linear model!
 - Pool viral loads are sums of individual viral loads
 - β_i
 - Equal 0: Specimen has no virus, negative individual
 - Greater than 0: Specimen has virus, positive individual
- Fit model to estimate $\boldsymbol{\beta}$
 - Assume $MVN(0, \sigma_y^2 \mathbf{I})$ for \mathbf{Y}
 - RC columns of \mathbf{X} (# of specimens) $> R + C$ rows of \mathbf{X} (# of pools)

- Could we use a linear model to predict positives/negatives rather than going to a second stage?
- Define
 - R = Number of rows of array, C = Number of columns of array
 - $\mathbf{Y} = (Y_1, \dots, Y_{RC})'$, a vector of viral loads for the pools; observable
 - $\boldsymbol{\beta} = (\beta_1, \dots, \beta_{RC})'$, a vector of true individual viral loads; not observable
- $E(\mathbf{Y}) = \mathbf{X}\boldsymbol{\beta}$
 - A linear model!
 - Pool viral loads are sums of individual viral loads
 - β_i
 - Equal 0: Specimen has no virus, negative individual
 - Greater than 0: Specimen has virus, positive individual
- Fit model to estimate $\boldsymbol{\beta}$
 - Assume $MVN(0, \sigma_y^2 \mathbf{I})$ for \mathbf{Y}
 - RC columns of \mathbf{X} (# of specimens) $> R + C$ rows of \mathbf{X} (# of pools)
 - Penalized regression model: non-negative LASSO

- Options based on threshold $c > 0$ (determined by assay manufacturer)

- Options based on threshold $c > 0$ (determined by assay manufacturer)
 - #1: Estimated viral load for specimen, $\hat{\beta}_i$
 - Equal to or larger than threshold declare positive
 - Less than a threshold declare negative

- Options based on threshold $c > 0$ (determined by assay manufacturer)

- #1: Estimated viral load for specimen, $\hat{\beta}_i$
 - Equal to or larger than threshold declare positive
 - Less than a threshold declare negative
 - #2: Same as #1 but
 - Retest specimens in a second stage for estimates in an indeterminate range
 - Indeterminate range: $0 < \hat{\beta}_i < c$

• Algorithms investigated

- Array testing with linear model, no retests (option #1)
- Array testing with linear model, potential retests (option #2)
- Array testing
- Dorfman testing

- Algorithms investigated

- Array testing with linear model, no retests (option #1)
- Array testing with linear model, potential retests (option #2)
- Array testing
- Dorfman testing

- Comparison metric: Efficiency

- Expected number of tests per individual
- “Best” algorithm has the lowest value

- Algorithms investigated
 - Array testing with linear model, no retests (option #1)
 - Array testing with linear model, potential retests (option #2)
 - Array testing
 - Dorfman testing
- Comparison metric: Efficiency
 - Expected number of tests per individual
 - “Best” algorithm has the lowest value
- Comparison metric: Positive percentage agreement (PPA)
 - Probability of declaring positive given the individual would test as positive
 - Like a sensitivity

- Algorithms investigated
 - Array testing with linear model, no retests (option #1)
 - Array testing with linear model, potential retests (option #2)
 - Array testing
 - Dorfman testing
- Comparison metric: Efficiency
 - Expected number of tests per individual
 - “Best” algorithm has the lowest value
- Comparison metric: Positive percentage agreement (PPA)
 - Probability of declaring positive given the individual would test as positive
 - Like a sensitivity
- Estimate efficiency and PPA
 - No closed form expressions for linear model-based algorithms
 - Use Monte Carlo simulation

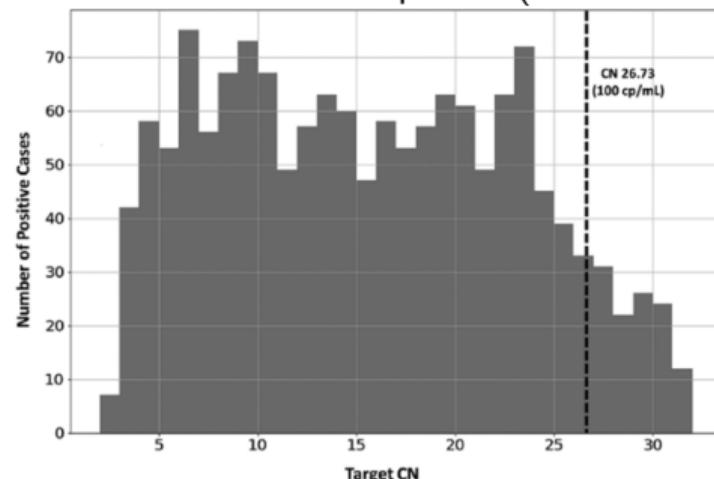
- Monte Carlo simulation summary
 - Simulate individual positive/negative status with Bernoulli(p), p is infection prevalence

• Monte Carlo simulation summary

- Simulate individual positive/negative status with Bernoulli(p), p is infection prevalence
- Simulate reverse transcription polymerase chain reaction (RT-PCR) assay testing process

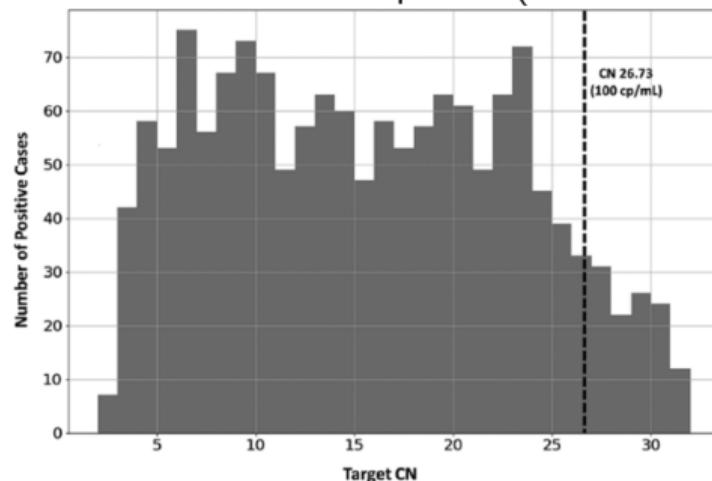
• Monte Carlo simulation summary

- Simulate individual positive/negative status with Bernoulli(p), p is infection prevalence
- Simulate reverse transcription polymerase chain reaction (RT-PCR) assay testing process
 - Emulate Abbott Realtime SARS-CoV-2 Assay (Hirschhorn et al. 2021)
 - Individual test result: Sample CN (also known as CT) and convert to viral load



• Monte Carlo simulation summary

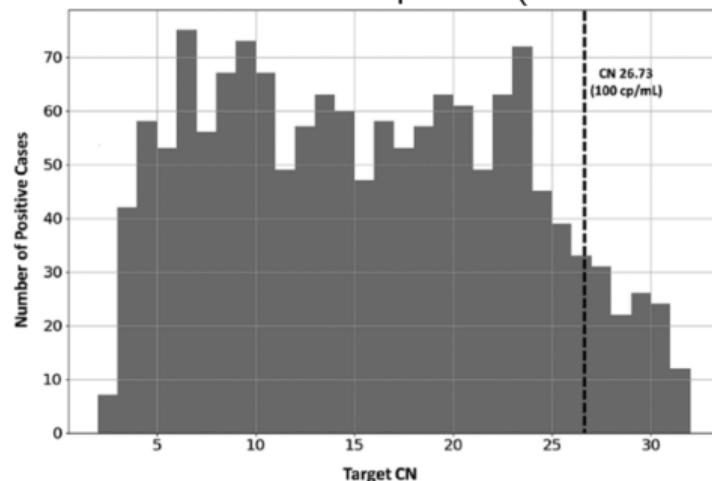
- Simulate individual positive/negative status with Bernoulli(p), p is infection prevalence
- Simulate reverse transcription polymerase chain reaction (RT-PCR) assay testing process
 - Emulate Abbott Realtime SARS-CoV-2 Assay (Hirschhorn et al. 2021)
 - Individual test result: Sample CN (also known as CT) and convert to viral load



- Emulate pool test results using Tan et al. (2020) and Arnout et al. (2021)

• Monte Carlo simulation summary

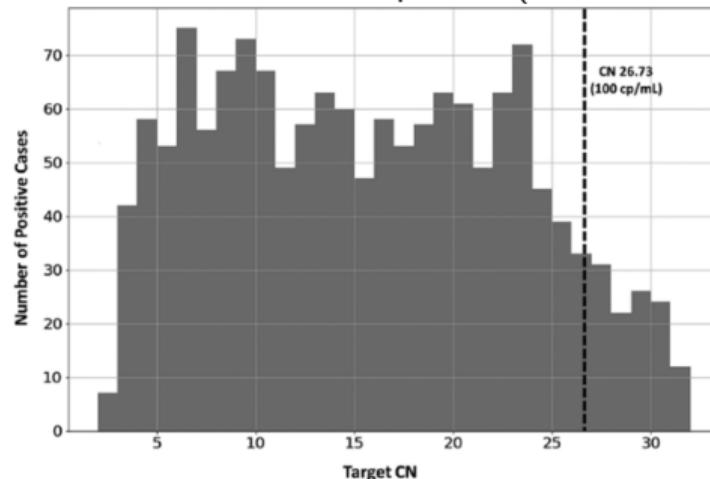
- Simulate individual positive/negative status with Bernoulli(p), p is infection prevalence
- Simulate reverse transcription polymerase chain reaction (RT-PCR) assay testing process
 - Emulate Abbott Realtime SARS-CoV-2 Assay (Hirschhorn et al. 2021)
 - Individual test result: Sample CN (also known as CT) and convert to viral load



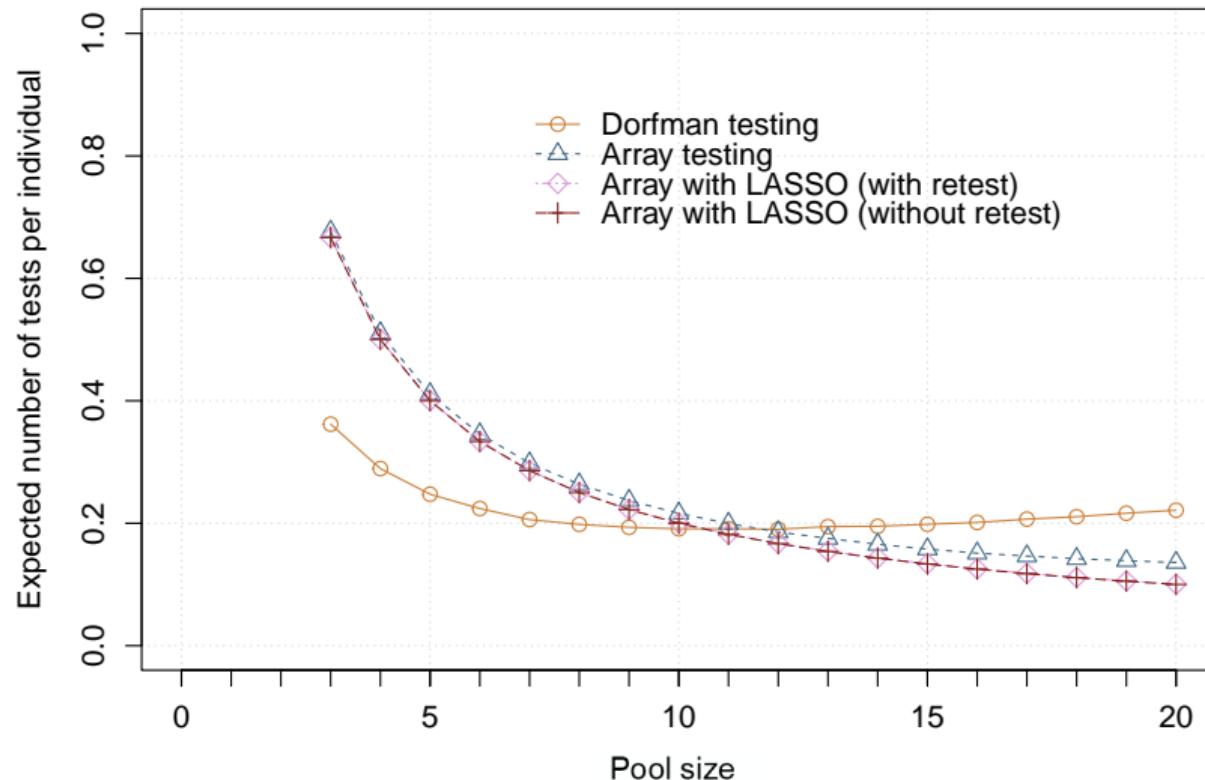
- Emulate pool test results using Tan et al. (2020) and Arnout et al. (2021)
- Repeat data simulation process 10,000 times

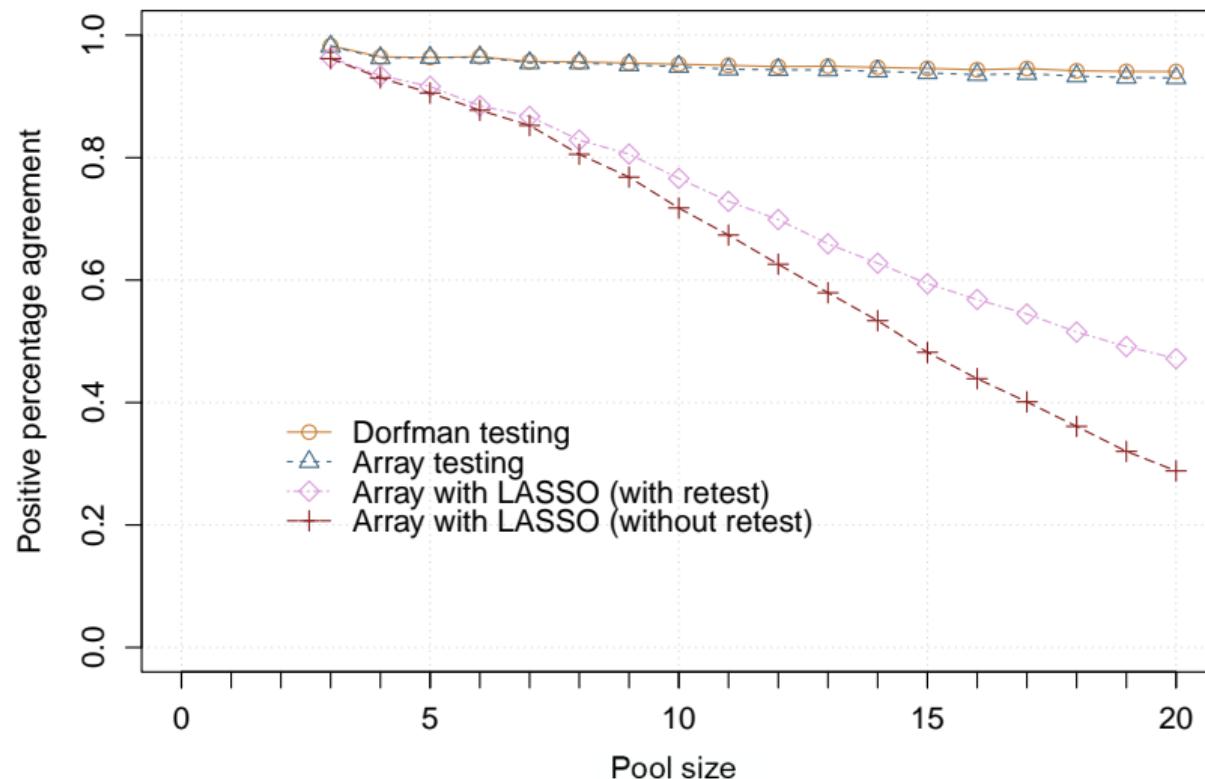
• Monte Carlo simulation summary

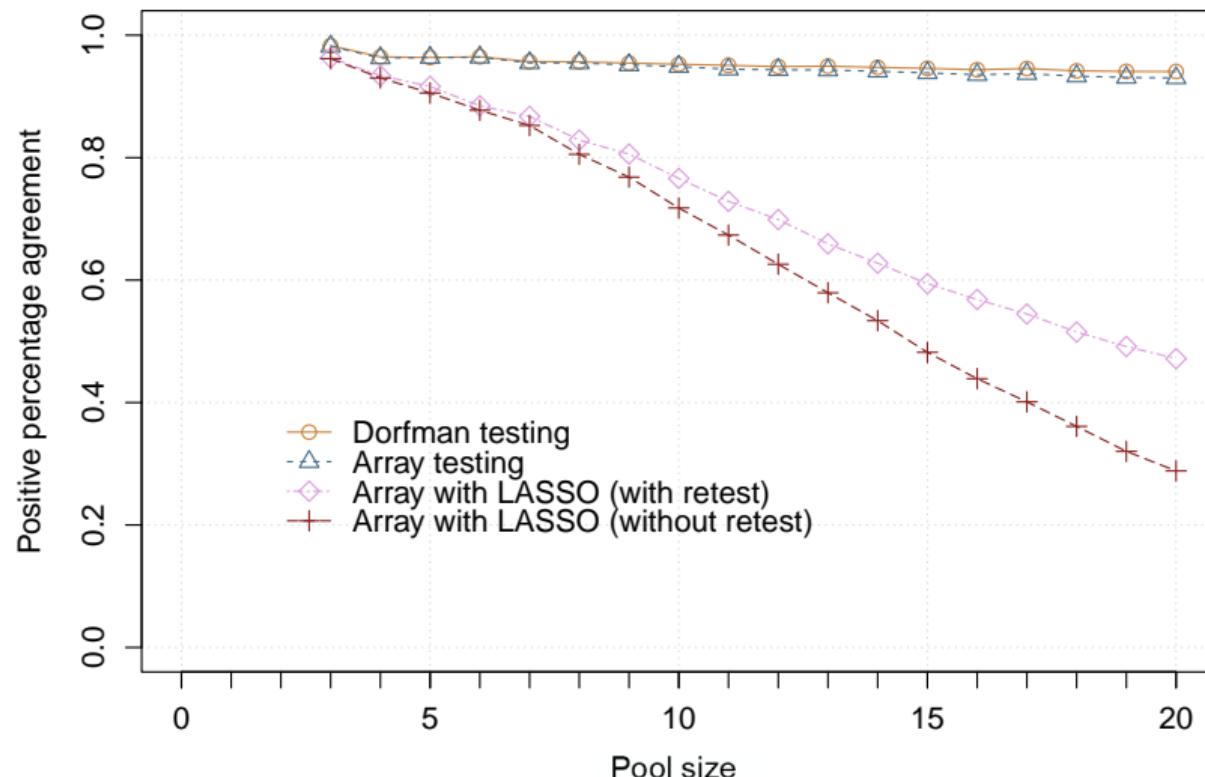
- Simulate individual positive/negative status with Bernoulli(p), p is infection prevalence
- Simulate reverse transcription polymerase chain reaction (RT-PCR) assay testing process
 - Emulate Abbott Realtime SARS-CoV-2 Assay (Hirschhorn et al. 2021)
 - Individual test result: Sample CN (also known as CT) and convert to viral load



- Emulate pool test results using Tan et al. (2020) and Arnout et al. (2021)
- Repeat data simulation process 10,000 times
- One setting: $p = 0.01$, pool sizes 3 to 20







- Why do the linear model-based algorithms perform poorly?

- Linear model-based algorithms with array testing: Not ready for labs yet!

- Linear model-based algorithms with array testing: Not ready for labs yet!
- Other investigations
 - Different p
 - Each individual put into more groups; e.g., 3D array testing
 - Adaptive LASSO
 - Pooling matrices from other authors

- Linear model-based algorithms with array testing: Not ready for labs yet!
- Other investigations
 - Different p
 - Each individual put into more groups; e.g., 3D array testing
 - Adaptive LASSO
 - Pooling matrices from other authors
- Incorporate statistical inference - $H_0 : \beta_i = 0$ vs. $H_a : \beta_i > 0$

Pooled testing with penalized regression models

Christopher R. Bilder
University of Nebraska–Lincoln
Department of Statistics
chris@chrisbilder.com

Research is supported by NIH grant R01 AI121351

Joint work with
Pranta Das at University of Nebraska-Lincoln,
Joshua M. Tebbs at University of South Carolina, and
Christopher S. McMahan at Clemson University