

COVID-19 METHYLATION RISK (COMER) SCORE

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Presenter: BSc. Tran Ba Thien

Date: Nov 14, 2025

About me



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 Research Assistant (Bioinformatics)

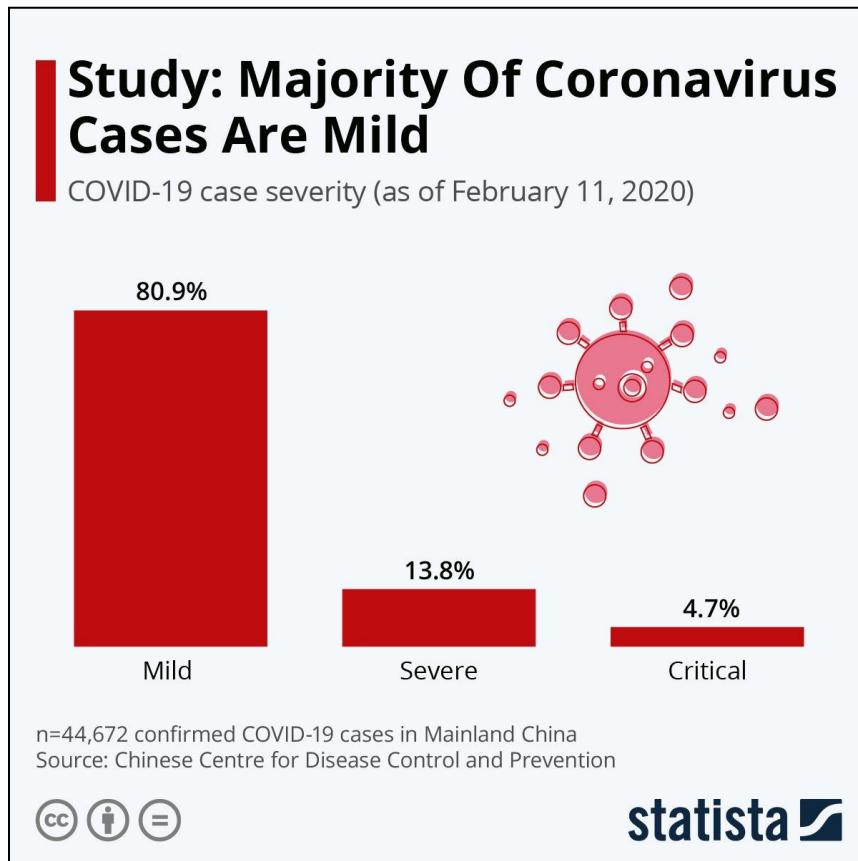
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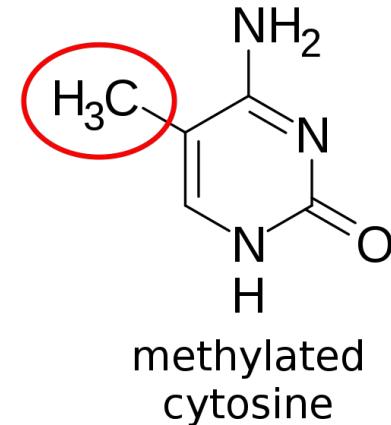
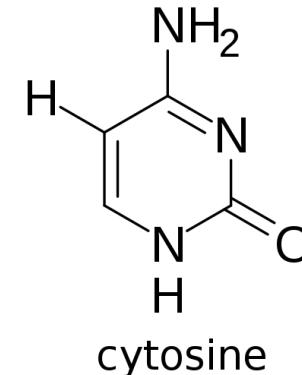
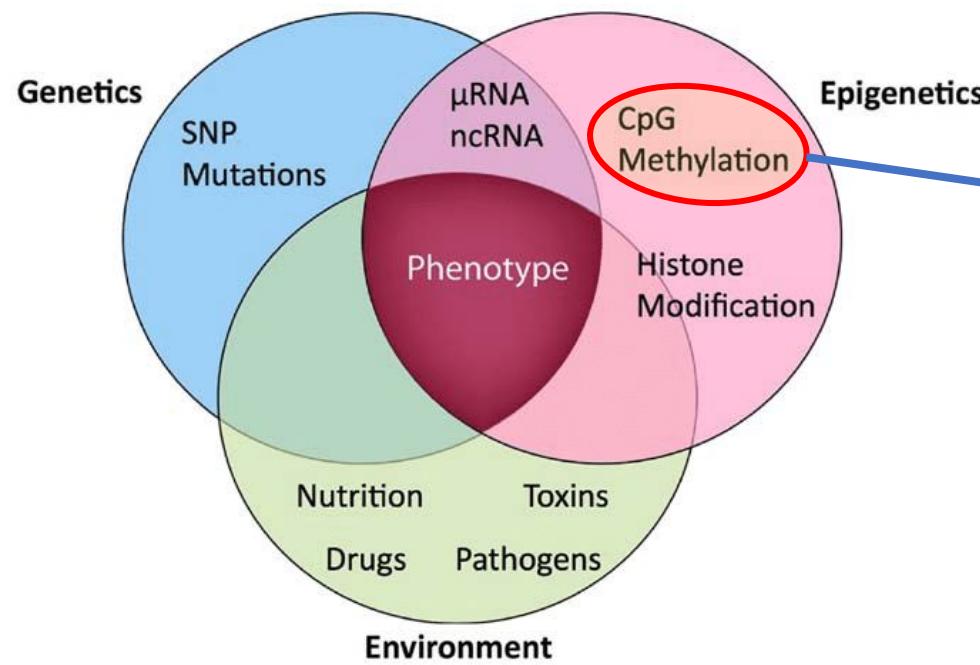
COVID-19 Disease



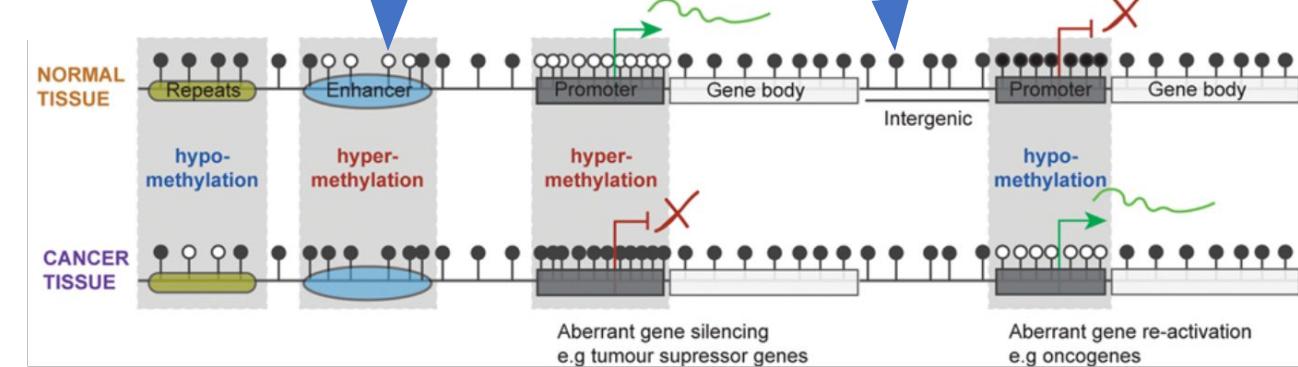
Developed COVID-19 biomarkers:

- SARS-CoV-2 RNA level (rRT-PCR)
- Antigen test (Rapid test kit)
- Inflammatory markers: cytokines, chemokines
- Risk score from clinical markers (Liang et al., 2020)
- ACE2 and TMPRSS2 genes (Barash et al., 2020)
- Epigenetic markers: 145 DMPs (Balnis et al., 2021) and 44 DMPs (Castro de Moura et al., 2021)

Epigenetics & DNA Methylation



- CpG (CG): Cytosine phosphate Guanine
- DMP: Differentially methylated position
- DMR: Differentially methylated region



Objective

1. Developing epigenetic markers for COVID-19 severity prediction
2. Establishing and evaluating COMER score
3. Exploring the relationship between COMER and biological characteristics
4. Launching the COMER score online tool

Data & Method

Data Resource & Sampling Criteria

Data collected from



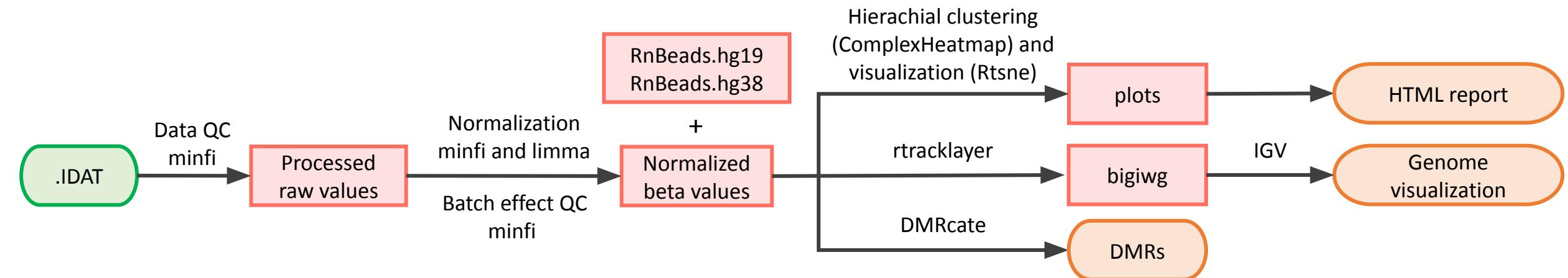
Searching keywords:

- COVID-19
- SARS-CoV-2
- Severity
- Epigenetics
- DNA Methylation
- Infinium
- MethylationEPIC
- Methylation450K
- Bisulfite Sequencing

Sampling criteria:

- Related to COVID-19 disease
- Announced since 2019
- Analyzed of blood samples
- Included clinical information

Data Preprocessing & Normalization Pipeline



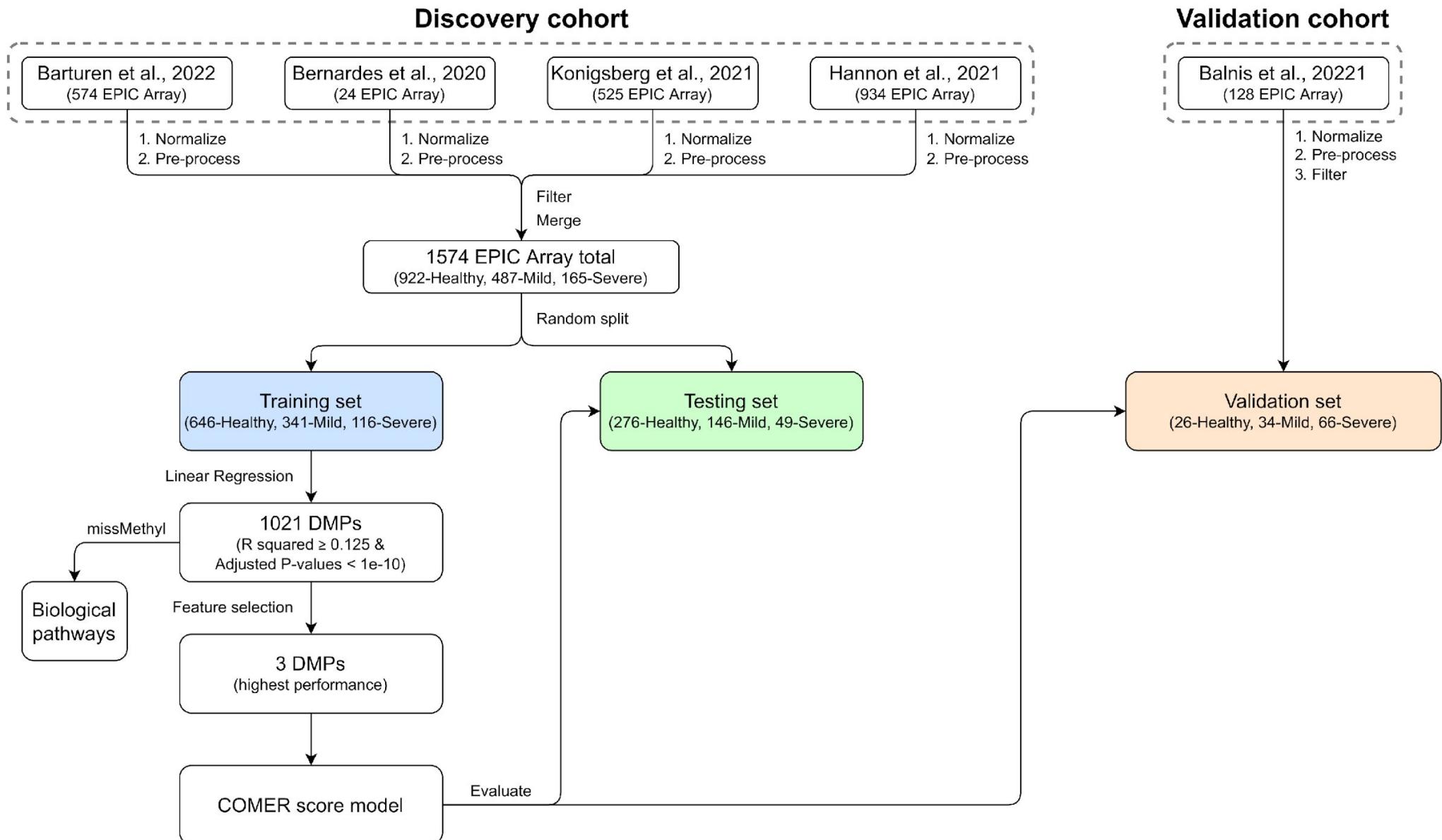
Package	Version	Link
minfi	1.34.0	http://bioconductor.org/packages/release/bioc/html/minfi.html
limma	3.44.3	https://bioconductor.org/packages/release/bioc/html/limma.html
RnBeads.hg19	1.20.0	https://bioconductor.org/packages/release/data/experiment/html/RnBeads.hg19.html
RnBeads.hg38	1.20.0	https://bioconductor.org/packages/release/data/experiment/html/RnBeads.hg38.html
Complex-Heatmap	2.4.3	https://www.bioconductor.org/packages/release/bioc/html/ComplexHeatmap.html
Rtsne	0.15	https://cran.r-project.org/web/packages/Rtsne/index.html
rtracklayer	1.48.0	https://bioconductor.org/packages/release/bioc/html/rtracklayer.html
DMRcate	2.2.3	https://bioconductor.org/packages/release/bioc/html/DMRcate.html

Software & Techniques

- Software: Bash programming & R programming version 4.2
- Techniques

Descriptive statistic	Median, interquartile range, Pearson correlation
Statistical hypothesis test	Wilcoxon rank sum, Kruskal-Wallis H test
DMR/DMP calling	DMRcate package in R
Gene ontology analysis	missMethyl package in R
Feature selection	LASSO regression, MUVR & BMA package in R
Machine learning	Linear regression, Random forest, Gradient boosting & Support vector machine
Visualization	ggplot2, ComplexHeatmap & Gviz package in R

Study Design



Result & Discussion

Quality Controls

Filtering

Study	Full Sample size	Sample included in study	Healthy controls		Mild patients		Severe patients	
			Age (median (IQR))	%Male	Age (median (IQR))	%Male	Age (median (IQR))	%Male
Balnis <i>et al.</i> , 2021	128	126	65 (53-75)	50.00	51.5 (40-64)	52.94	67 (55-78)	68.18
Barturen <i>et al.</i> , 2022	574	574	70 (48-81)	46.53	65.5 (54-77)	48.33	76 (69-85)	58.41
Bernardes <i>et al.</i> , 2020	24	21	56 (53-59)	50.00	70 (60-80)	66.67	70 (58-80)	50.00
Hannon <i>et al.</i> , 2021	934	519	37 (27-50)	47.01	-	-	-	-
Konigsberg <i>et al.</i> , 2021	525	460	59 (41-70)	51.01	49.5 (37-62)	53.39	50 (38-62)	65.22
Total	2185	1700	47 (32-61)	48.31	62 (49-75)	50.10	71 (56-80)	62.34

>850.000 CGs/sample

Preprocessing
Normalization

690.285 CGs/sample

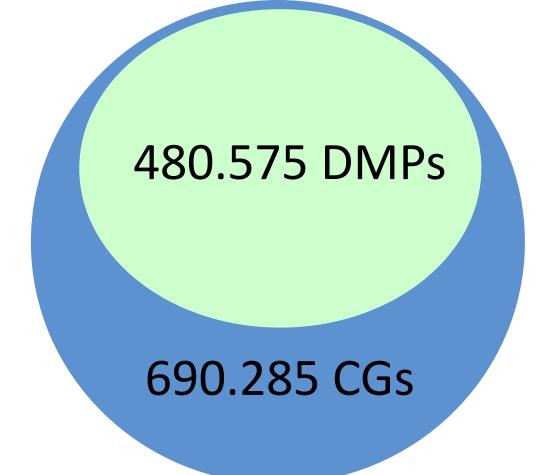
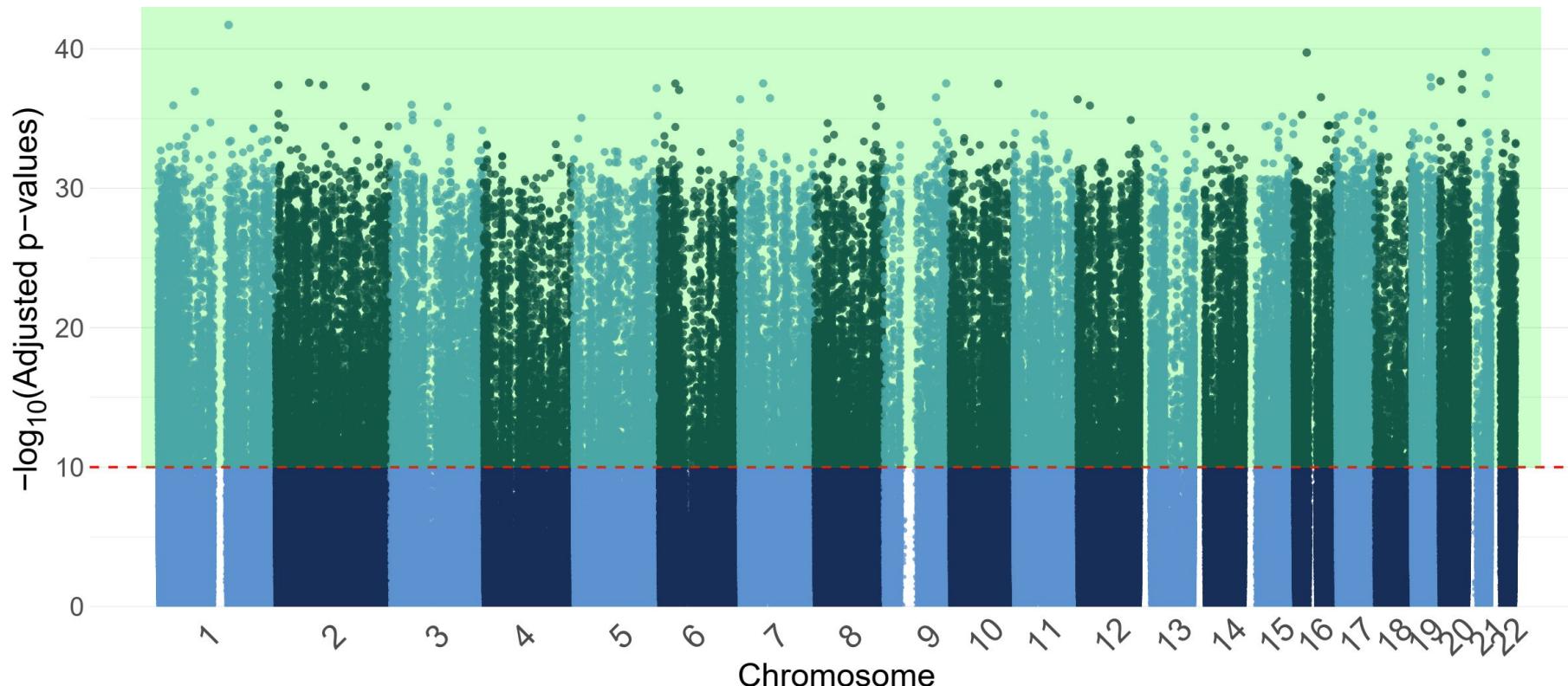
Association of DNA Methylation & COVID-19

Disease

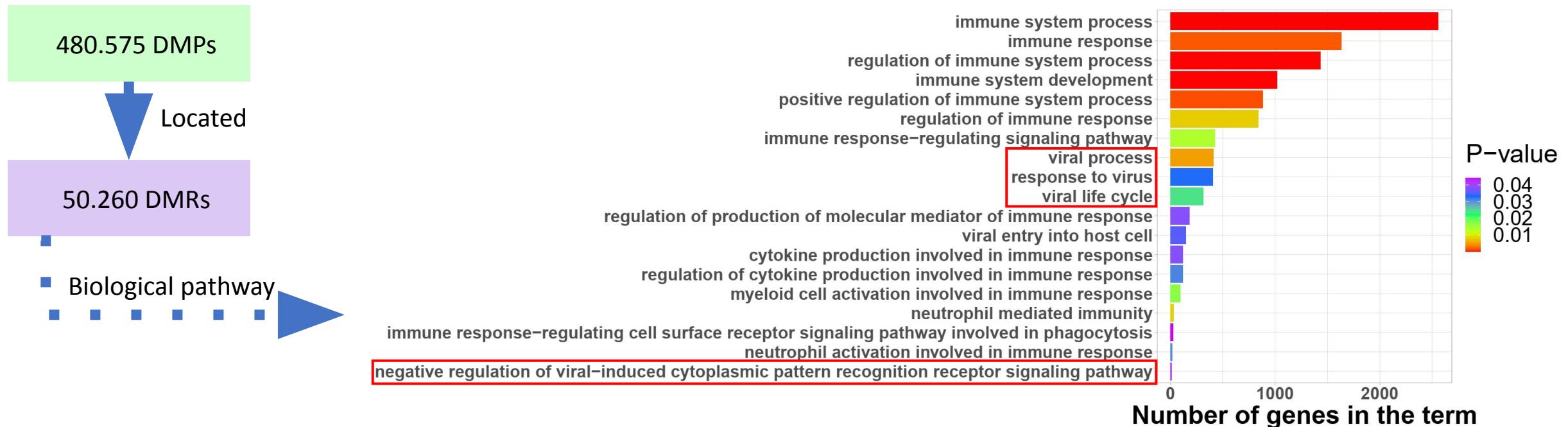
P-value estimated from liner regression:

Disease progression(0, 1, 10) ~ Each CpG mehylated value + Age + Gender

Benjamini-Hochberg adjusted p-value < 10^{-10}



Association of DNA Methylation & Immune System

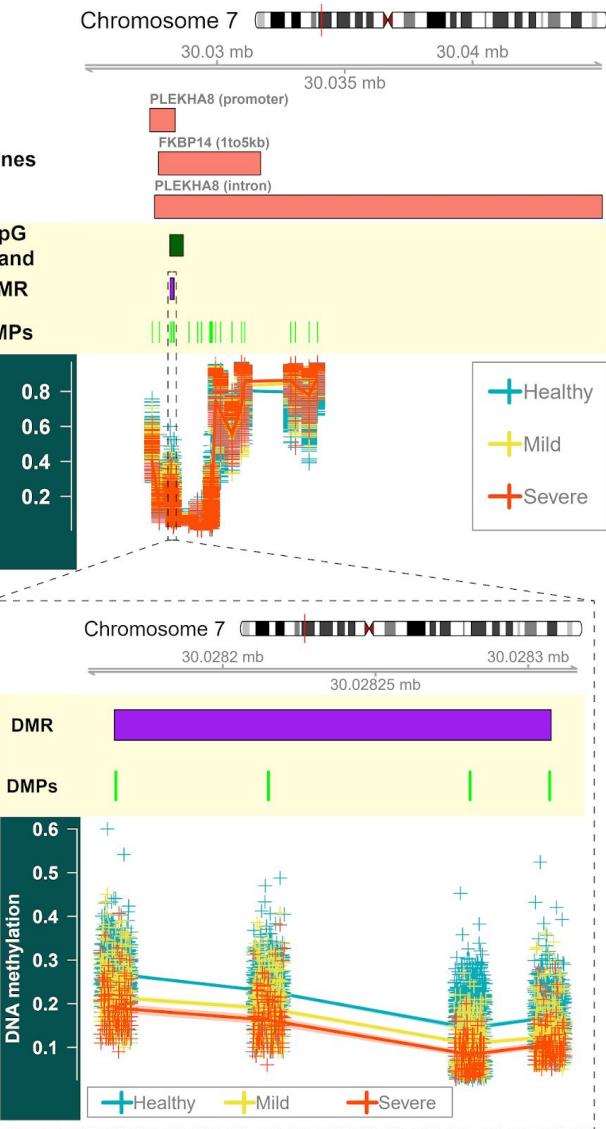
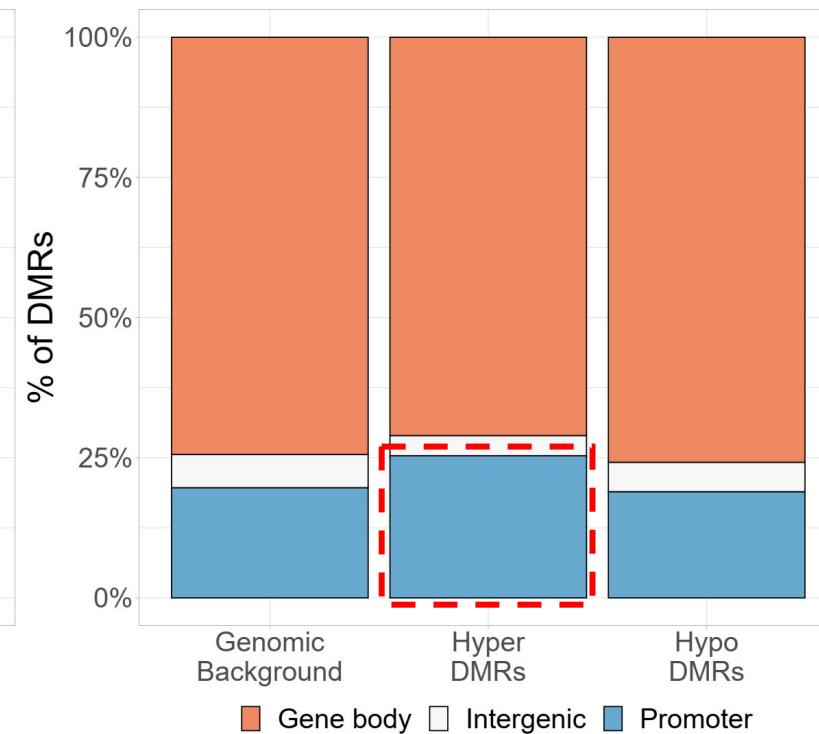
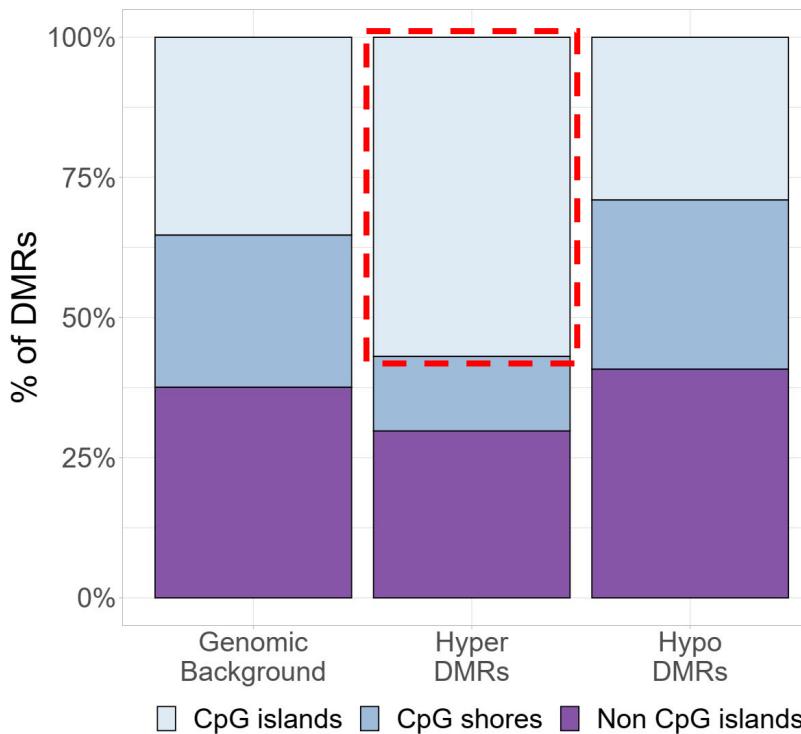


- Changes in DNA methylation associated with immune response to virus

Hyper-DMRs Enrich in CG Islands & Promoters

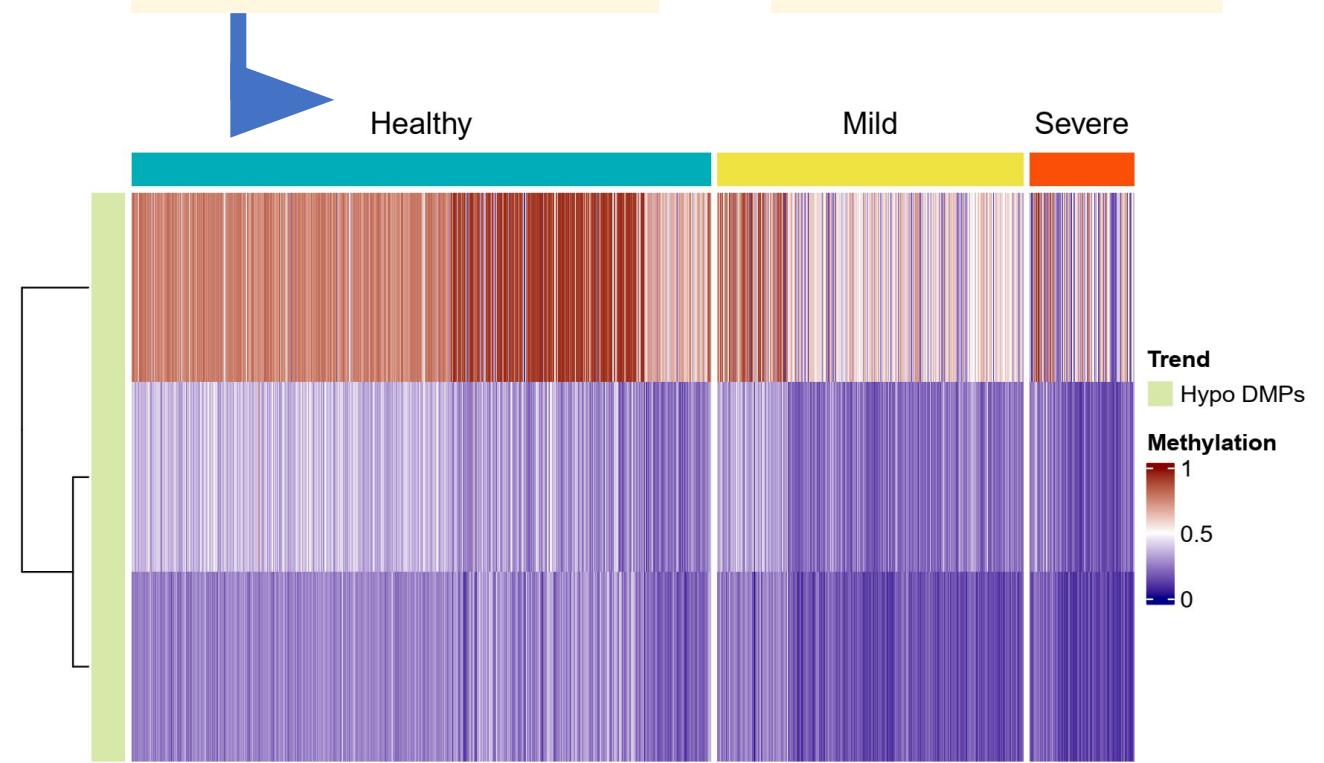
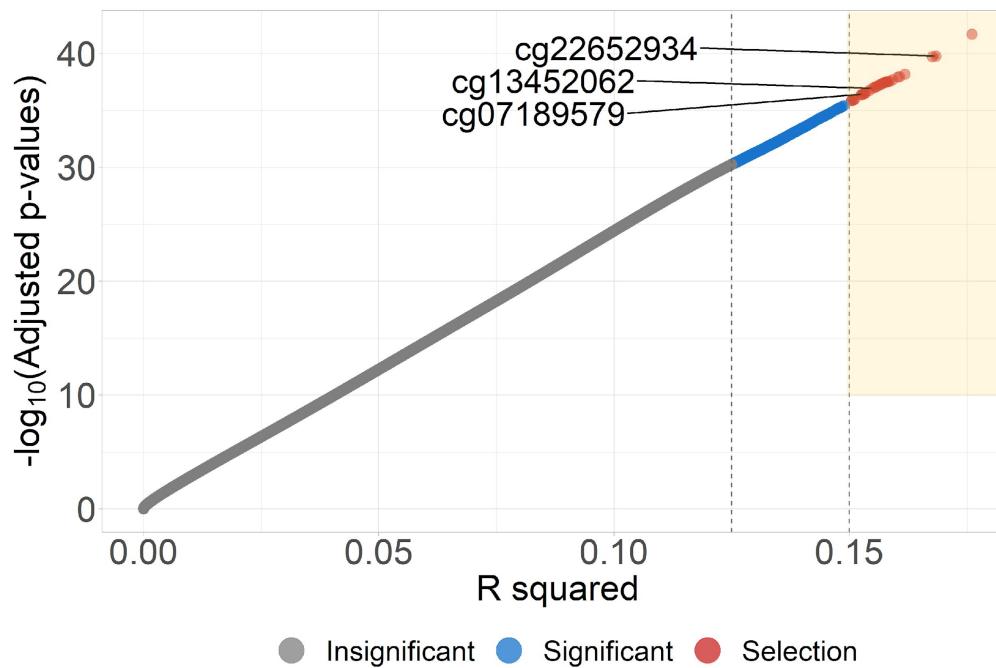
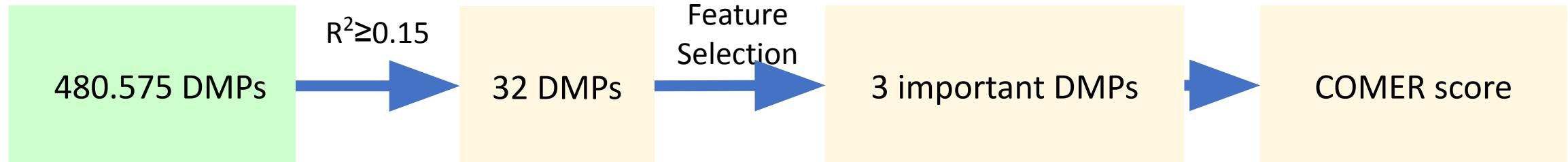
50.260 DMRs

5456 ↑ hyper-DMR
44804 ↓ hypo-DMRs



□ Methylation changes in specific genes may contribute to the severe COVID-19 cases

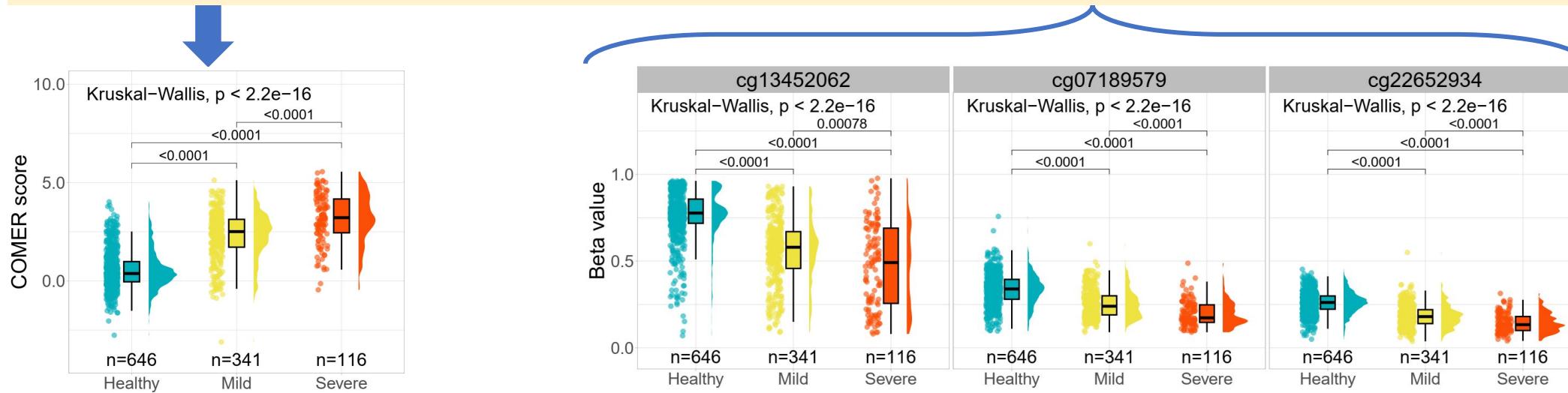
Narrow Down Significant DMPs



COMER Score Equation Establishment

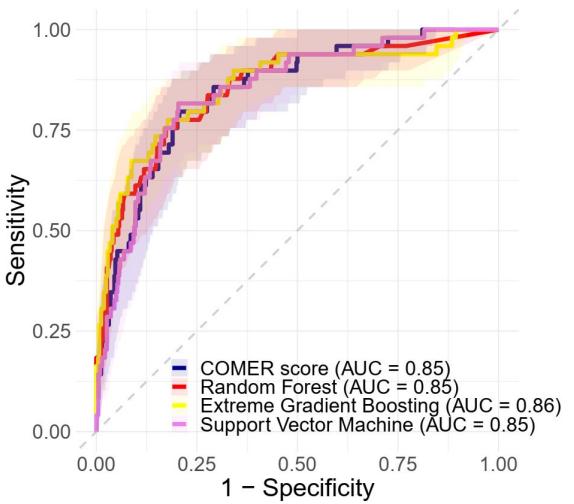
DMPS	CHR	START	END	GENE NAME	UCSC REFGENE		TXDB		MEAN DELTA	
					RELATION TO ISLAND	GROUP	RELATION TO ISLAND	GROUP	SEVERE VS. NON SEVERE	INFECTION VS. HEALTHY
cg13452062	chr1	78622873	78622875	IFI44L	Open Sea	5'UTR	CpG inter	Introns 1to5kb	-0.22	-0.23
cg07189579	chr8	133246200	133246202	NDRG1	Open Sea	Body	CpG inter	Introns 1to5kb	-0.11	-0.10
cg22652934	chr21	34807737	34807739	RUNX1	Open Sea	Body	CpG inter	Introns	-0.09	-0.09

$$\text{COMER score} = 6.9642 - 3.2855 \times \beta_1 - 6.7090 \times \beta_2 - 6.3302 \times \beta_3$$

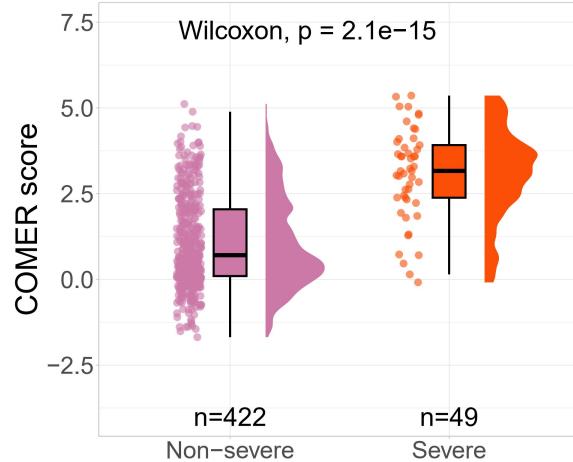
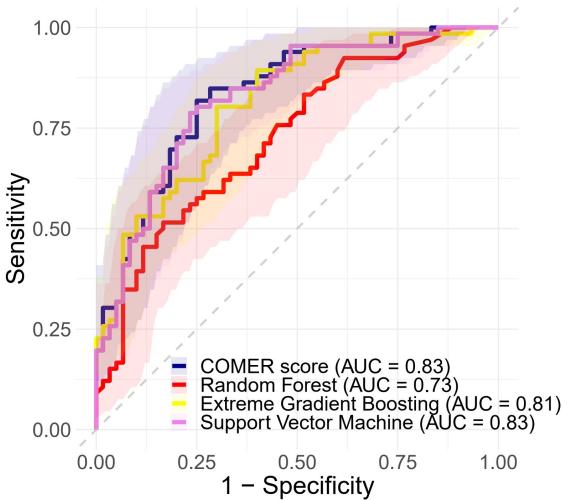


COMER Score Evaluation

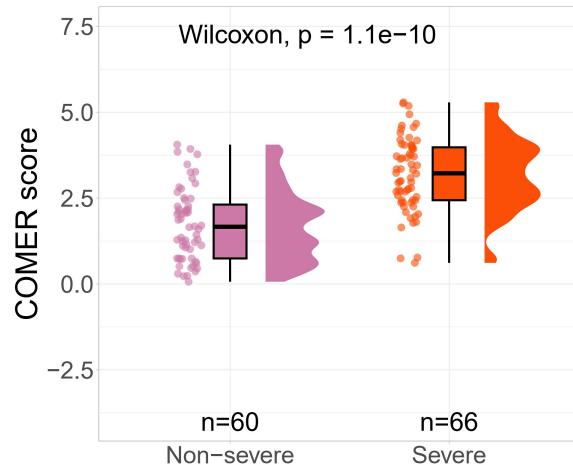
Testing set



Validation set
(Independently)



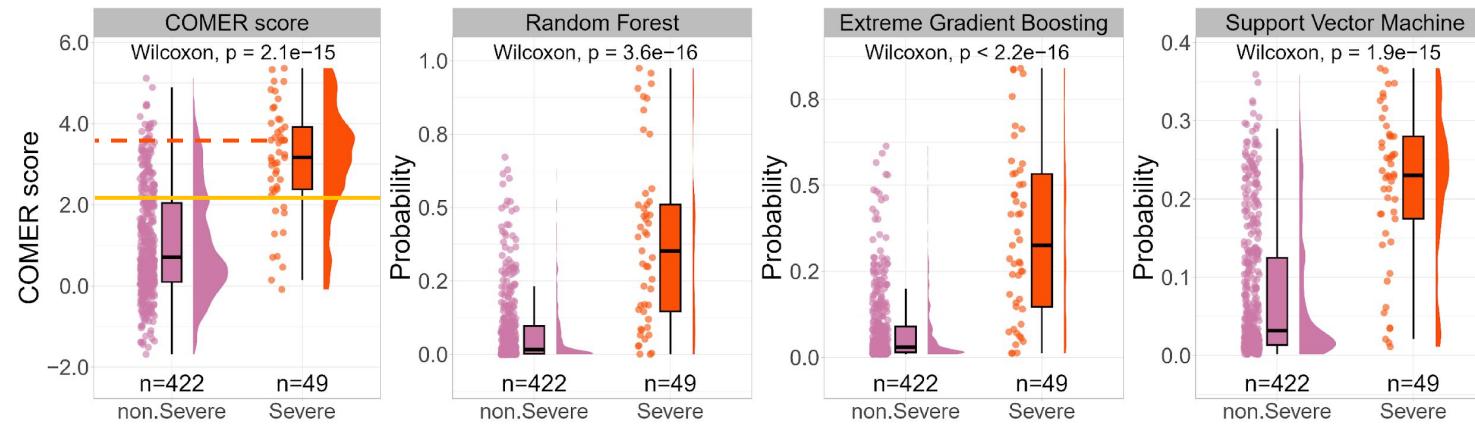
		Actual	
		Non-severe	Severe
Prediction	Non-severe	343	14
	Severe	79	35



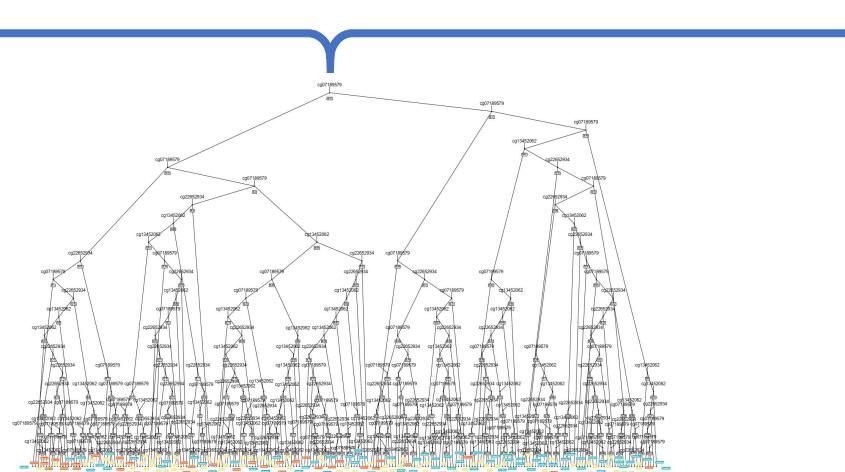
		Actual	
		Non-severe	Severe
Prediction	Non-severe	45	15
	Severe	15	51

□ The COMER score is a good discriminator for severe cases

COMER Score Compare to Other ML Approaches

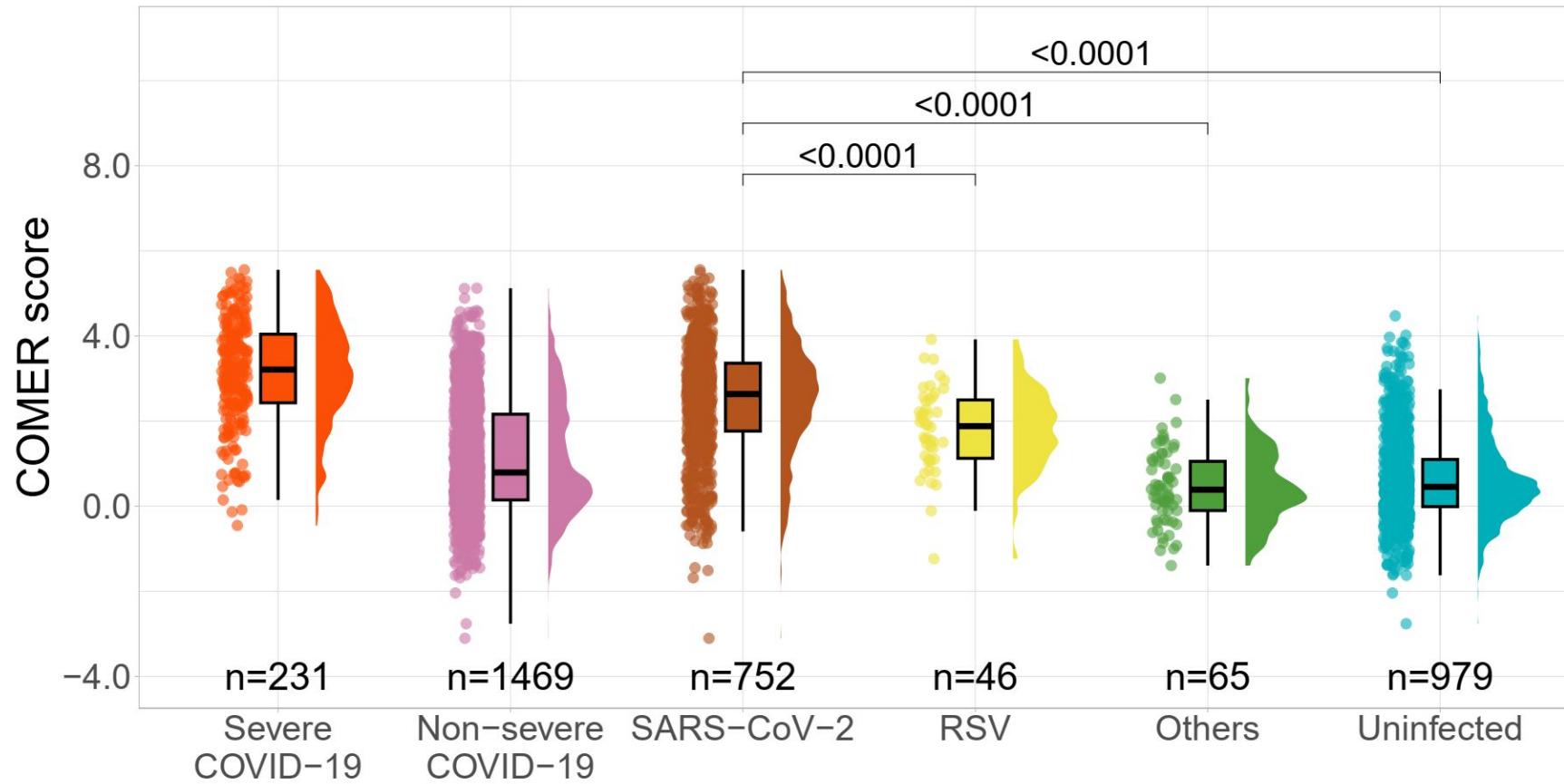


Severe:
COMER > 2.39
Non-severe:
COMER ≤ 2.39
 Interpretable



- COMER score is **consistent, interpretable** and **simple** to use compare to other ML approaches

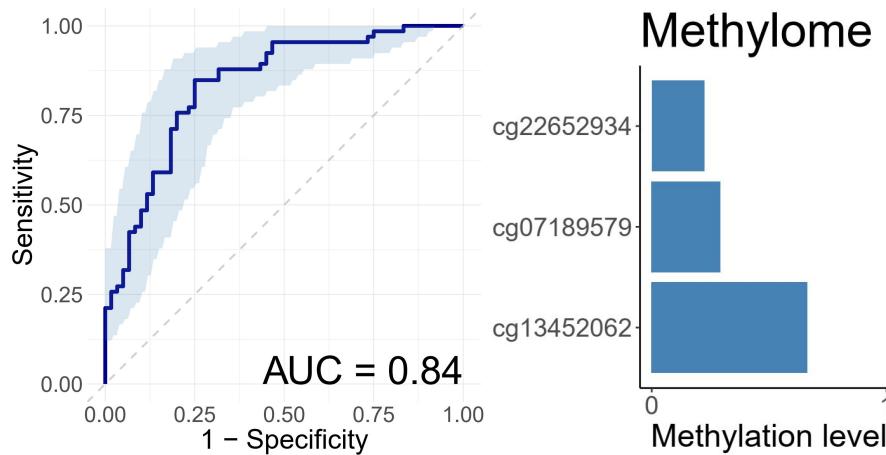
COMER Score is Specific for SARS-CoV-2



- COMER score is highly specific for SARS-CoV-2 and not affected by viral co-infection

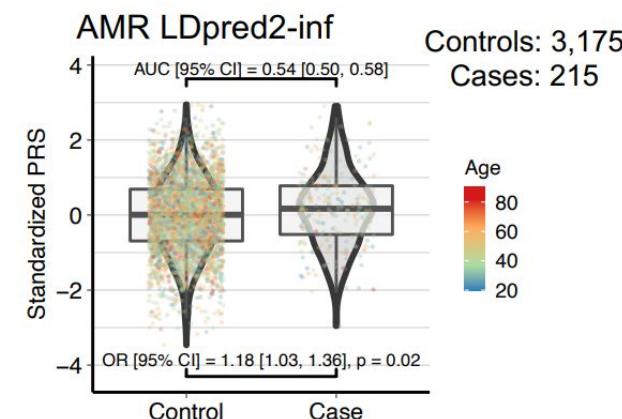
COMER Score Compare to Other Scoring Systems

COMER score



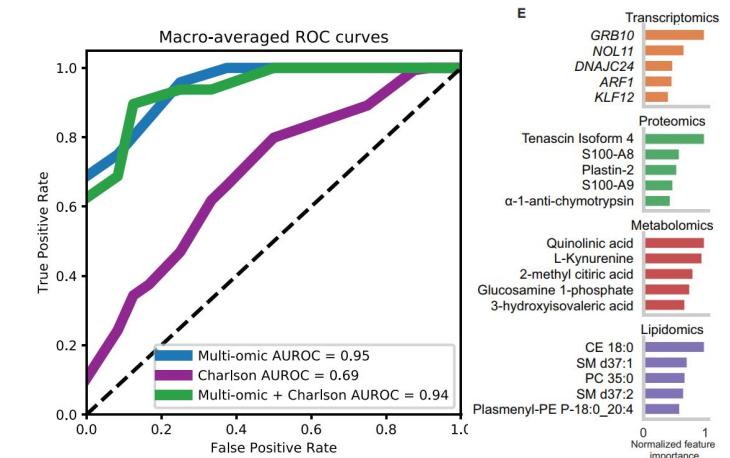
Polygenic Risk Score COVID-19 severity for Admixed Americans

(Christopher et al., 2020)



Large-Scale Multi-omic of COVID-19 Severity ExtraTrees classifier

(Overmyer et al., 2021)



- COMER score is a comparable scoring system that requires simple input

COMER Score is Highly Positive Correlated with Clinical Scores

Description	WHO Ordinal Scale*
ICU, mechanically ventilated with signs of ARDS	7
State with severe signs of a systemic inflammatory response, ICU, high-flow oxygen, intubation readiness	6
Supplemental oxygen, significant signs of systemic inflammation	5
Intermittent supplemental oxygen, minor signs of inflammation	4
No supplemental oxygen, absent inflammation markers	3
Where clinical symptoms and inflammatory markers were increasing, ICU or non-icu	2
At least two weeks after hospital discharge	1
PCR -	0

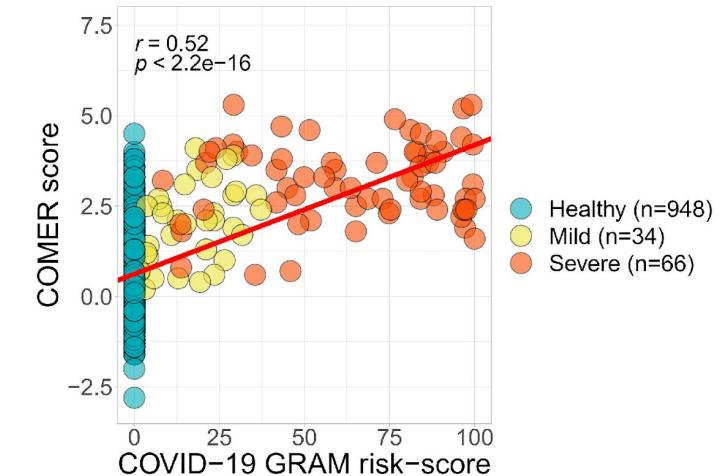
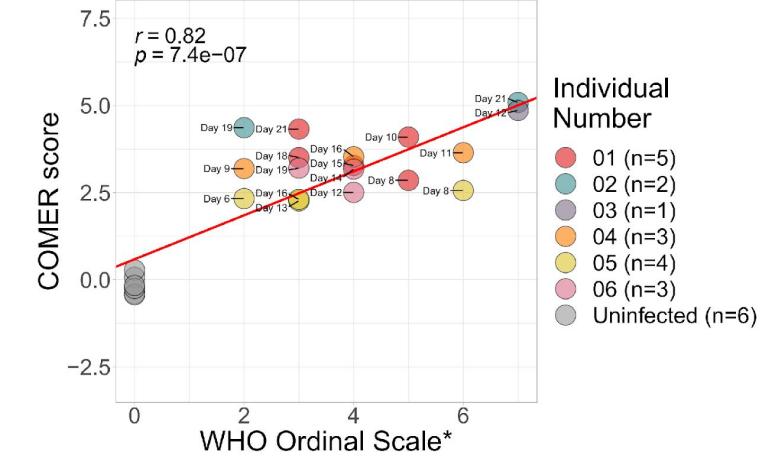
JAMA Internal Medicine | Original Investigation

Development and Validation of a Clinical Risk Score to Predict the Occurrence of Critical Illness in Hospitalized Patients With COVID-19

Wenhua Liang, MD; Hengrui Liang, MD; Limin Ou, MD; Binfeng Chen, MD; Ailan Chen, MD; Caichen Li, MD; Yimin Li, MD; Weijie Guan, MD; Ling Sang, MD; Jitao Lu, MD; Yuanda Xu, MD; Guoqiang Chen, MD; Haiyan Guo, MD; Jun Guo, MD; Zisheng Chen, MD; Yi Zhao, MD; Shiyue Li, MD; Nuofu Zhang, MD; Nanshan Zhong, MD; Jianxing He, MD; for the China Medical Treatment Expert Group for COVID-19

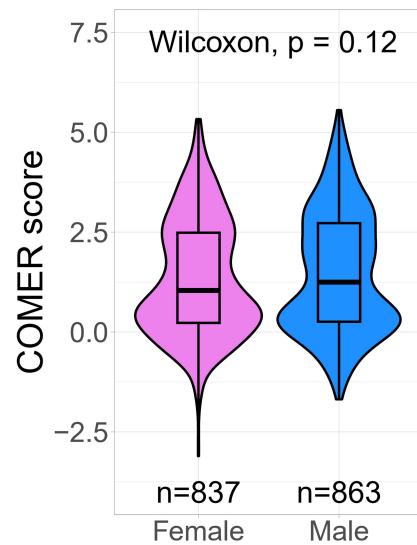
- The COMER score has the potential to early predict patient outcomes and is a reliable alternative to a complex scoring system.

- Based on **10 clinical factors**
- Chest radiographic abnormality
 - Age
 - Hemoptysis
 - Dyspnea
 - Unconsciousness
 - Number of comorbidities
 - Cancer history
 - Neutrophil-to-lymphocyte ratio
 - Lactate dehydrogenase
 - Direct bilirubin
- GRAM risk score AUC = 0.88

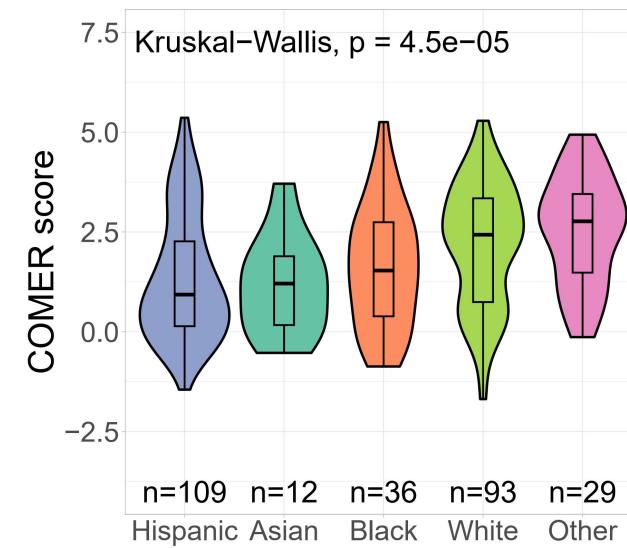


Association of COMER & Other Biological Characteristics

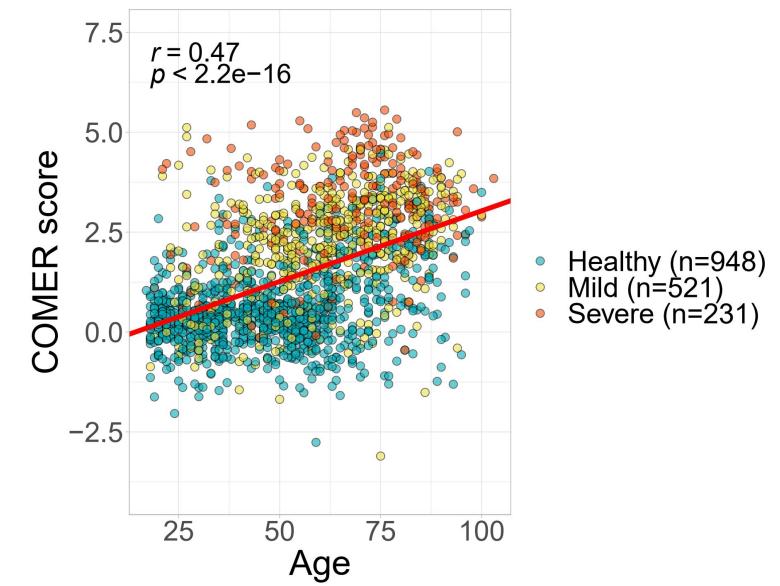
Gender



Race



Age



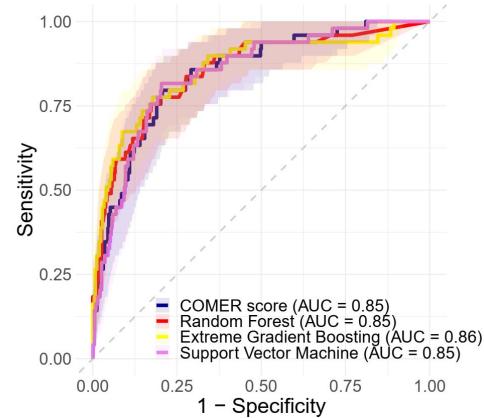
Extensive differences in DNA methylation throughout the genome between ethnic subgroups (Elliott *et al.*, 2022; Galanter *et al.*, 2017)

- COMER score is not significantly differed by gender but by race/age

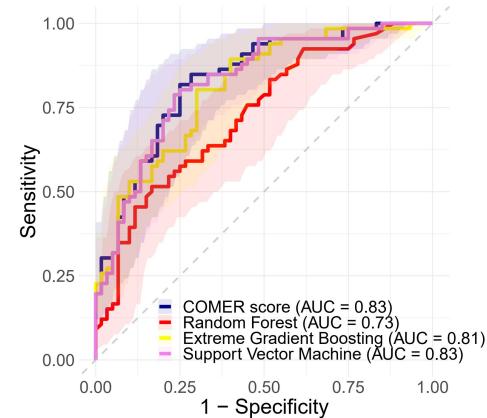
COMER Score Works Well Regardless of Age

Testing set

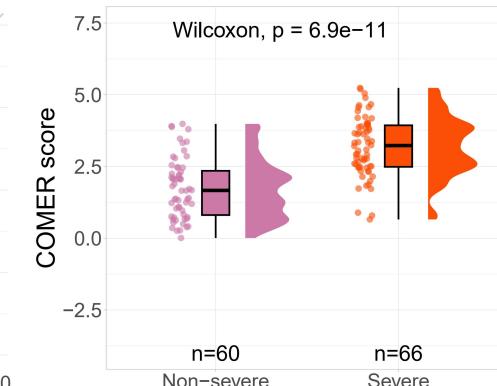
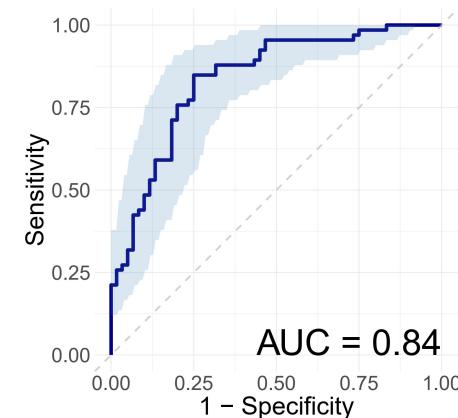
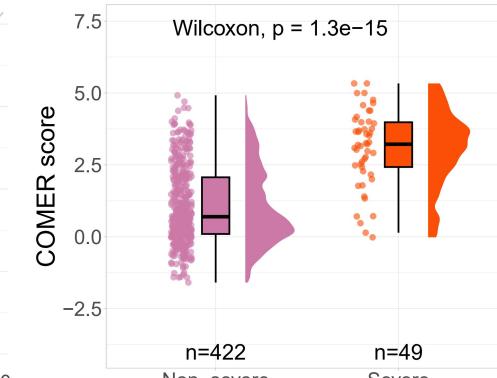
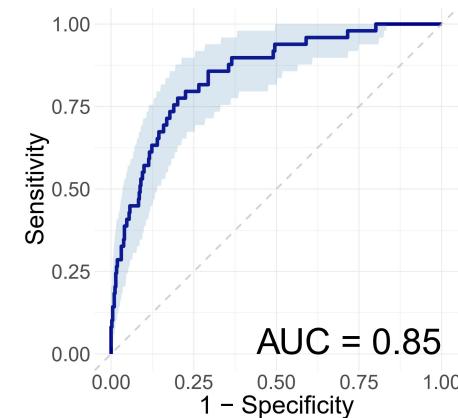
Before age adjustment



Validation set
(Independently)

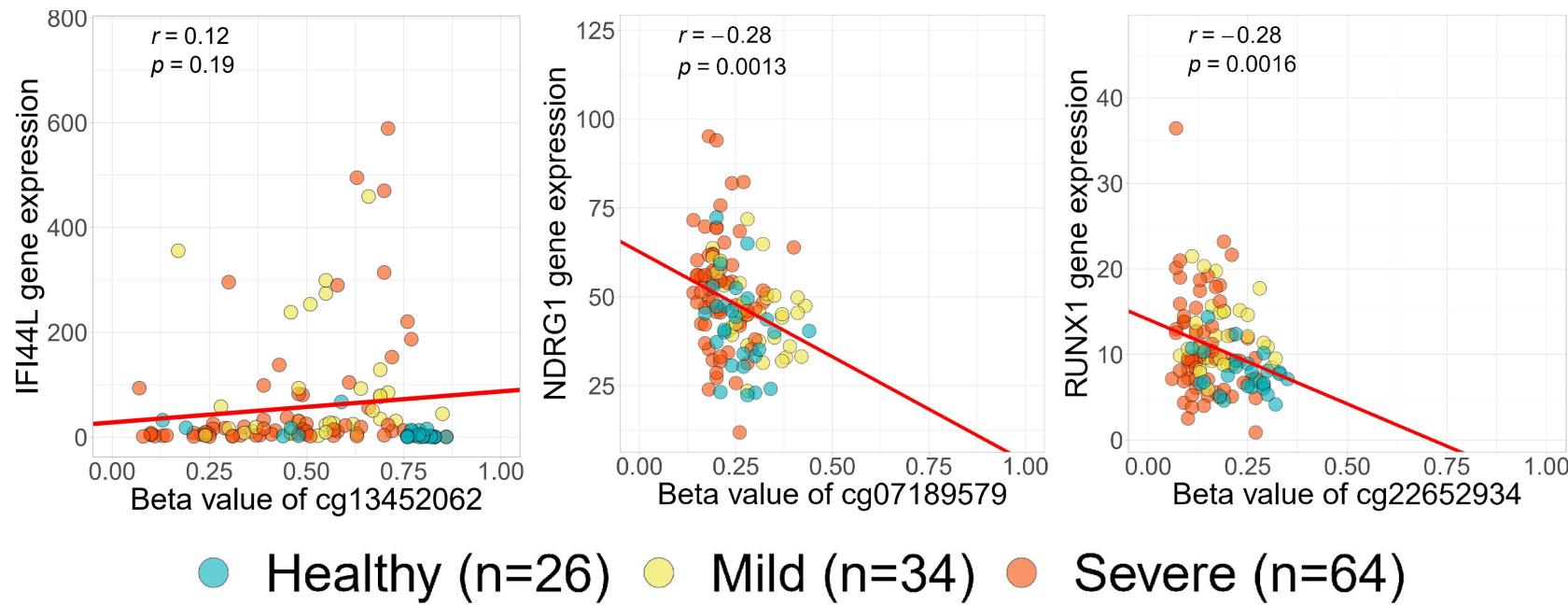


After age adjustment



- The COMER score is effective regardless of age

COMER score component associate to pathogenesis



- Zolfaghari Emameh *et al.* (2020): NDRG1 gene down-regulated result in induction of proinflammatory signals and activation of a cytokine storm
- O'Hare *et al.* (2021): RUNX1 plays a critical role in the pathologic host response to SARS-CoV-2 infection by regulating inflammation and hematopoiesis

□ COMER score components relate to the pathogenesis of COVID-19 infection severity

COMERS Online Tool

[COVID-19 Severity Prediction](#) [Guideline](#) [Multi Prediction](#) [Personal Prediction](#) [About](#)

User guideline

The COMER score shiny app tool provides a simple and user-friendly interface for predicting COVID-19 severity in individuals. The tool includes two prediction tabs: "Multi Prediction" and "Personal Prediction", both of which generate the same COMER score and predicted outcome for the same individual. The main difference between the two tabs is that "Multi Prediction" allows users to observe the distribution of their entire population based on COMER score, while "Personal Prediction" provides a detailed visualization of the value of each CpG site and the corresponding COMER score for the individual in question.

COMER score formulas

Standard formula:
 $\text{COMER score} = 6.9642 - 3.2855 \times \beta_1 - 6.7090 \times \beta_2 - 6.3302 \times \beta_3$

If a patient's COMER score is greater than 2.393785, they will be predicted as Severe.

Age-adjusted formula:
 $\text{COMER score} = 6.500982 + 0.04110 \times \alpha_1 - 3.259439 \times \beta_1 - 6.394061 \times \beta_2 - 6.171600 \times \beta_3$

If a patient's age-adjusted COMER score is greater than 2.508359, they will be predicted as Severe.

More:
 α_1 : age
 β_1 : beta value of cg22652934
 β_2 : beta value of cg13452062
 β_3 : beta value of cg07189579

Multi-prediction:

1. Import data
 Click on the "Browse" button and select your TSV file to import your data. Note that your TSV file must contain at least 5 columns (sample_ID, Age, cg13452062, cg07189579, and cg22652934), with each row representing a sample.

[COVID-19 Severity Prediction](#) [Guideline](#) [Multi Prediction](#) [Personal Prediction](#) [About](#)

Input parameters

Choose TSV File
 Browse... covid3.0_DT_val.tsv
 Upload complete

[Prediction without age](#) [Prediction with age](#)

Show 5 entries Search:

Your samples information

sample_ID	age	cg22652934	cg13452062	cg07189579
1 COVID_01	39	0.18	0.7	0.24
2 COVID_02	63	0.12	0.36	0.23
3 COVID_03	33	0.17	0.17	0.19
4 COVID_04	49	0.21	0.20	0.32
5 COVID_05	49	0.11	0.05	0.24

Showing 1 to 5 of 128 entries Previous 1 2 3 4 5 ... 26 Next

Non-severe Severe

COMER score distribution

Box plot showing COMER scores for Non-severe (pink) and Severe (orange) patients. The y-axis is labeled 'COMER SCORE' and ranges from 0 to 5. The x-axis shows two groups: 'Non-severe' and 'Severe'. The Non-severe group has a median score around 1.5, while the Severe group has a median score around 3.5, with a wider distribution.

Copy CSV Excel PDF Print Show 5 entries

Scores in details and prediction

sample_ID	sex	age	cg22652934	cg13452062	cg07189579
1 COVID_01	M	39	0.18	0.7	0.24
2 COVID_02	M	63	0.12	0.36	0.23
3 COVID_03	M	33	0.17	0.17	0.19
4 COVID_04	M	49	0.21	0.20	0.32
5 COVID_05	M	49	0.11	0.05	0.24

Showing 1 to 5 of 128 entries Previous 1 2 3 4 5 ... 26 Next

Plot without age Plot with age

[COVID-19 Severity Prediction](#) [Guideline](#) [Multi Prediction](#) [Personal Prediction](#) [About](#)

Choose TSV File
 Browse... covid3.0_DT_val.tsv
 Upload complete

Prediction

Show 5 entries Search:

Scores in details and prediction

sample_ID	sex	age	cg22652934	cg13452062	cg07189579
1 COVID_01	M	39	0.18	0.7	0.24
2 COVID_02	M	63	0.12	0.36	0.23
3 COVID_03	M	33	0.17	0.17	0.19
4 COVID_04	M	49	0.21	0.20	0.32
5 COVID_05	M	49	0.11	0.05	0.24

Showing 1 to 5 of 128 entries Previous 1 2 3 4 5 ... 26 Next

Search:

sample_ID sex age cg22652934 cg13452062 cg07189579

1 COVID_01 M 39 0.18 0.7 0.24

2 COVID_02 M 63 0.12 0.36 0.23

3 COVID_03 M 33 0.17 0.17 0.19

4 COVID_04 M 49 0.21 0.20 0.32

5 COVID_05 M 49 0.11 0.05 0.24

Beta value of β_1 : 0.11 COMER score = $6.9642 - 3.2855 \times \beta_1 - 6.7090 \times \beta_2 - 6.3302 \times \beta_3$

Beta value of β_2 : 0.85 COMER score = $6.9642 - 3.2855 \times 0.11 - 6.7090 \times 0.85 - 6.3302 \times 0.24$

Beta value of β_3 : 0.24 COMER score = -0.6191029999999999

Plot without age Plot with age

COMER score distribution

Density plot showing the distribution of COMER scores for Non-severe (pink) and Severe (orange) patients. The x-axis is labeled 'COMER score' and ranges from -2 to 6. The y-axis is labeled 'Density' and ranges from 0.00 to 1.00. Two bell-shaped curves are shown: one centered around 1.5 for Non-severe patients and one centered around 3.5 for Severe patients. A vertical dashed line marks the 'Current patient' at approximately 2.2.



User guideline

The COMER score Shiny app tool provides a simple and user-friendly interface for predicting COVID-19 severity in individuals. The tool includes two prediction tabs: "Multi Prediction" and "Personal Prediction", both of which generate the same COMER score and predicted outcome for the same individual. The main difference between the two tabs is that "Multi Prediction" allows users to observe the distribution of their entire population based on COMER score, while "Personal Prediction" provides a detailed visualization of the value of each CpG site and the corresponding COMER score for the individual in question.

COMER score formulas

Standard formula:

COMER score = 6.9642 - 3.2855×β1 - 6.7090×β2 - 6.3302×β3

If a patient's COMER score is greater than 2.393789, they will be predicted as Severe.

Age adjusted formula:

COMER score = $6.590862 + 0.004110 \times \alpha - 3.250439 \times \beta_1 - 6.394083 \times \beta_2 - 6.171600 \times \beta_3$

If a patient's age-adjusted COMER score is greater than 2.508359, they will be predicted as Severe.

Where:

```
a : age  
β1: beta value of cg13452062  
β2: beta value of cg07189579  
β3: beta value of cg22652934
```

Data requirement

COMER score is generated by using methylation beta value of three CpG sites (cg13452062, cg07189579, and cg22652934), with an optional age addition. So, your TSV format data must contain at least 5 columns (sample ID, age, cg13452062, cg07189579, and cg22652934), with each row representing one sample/individual.

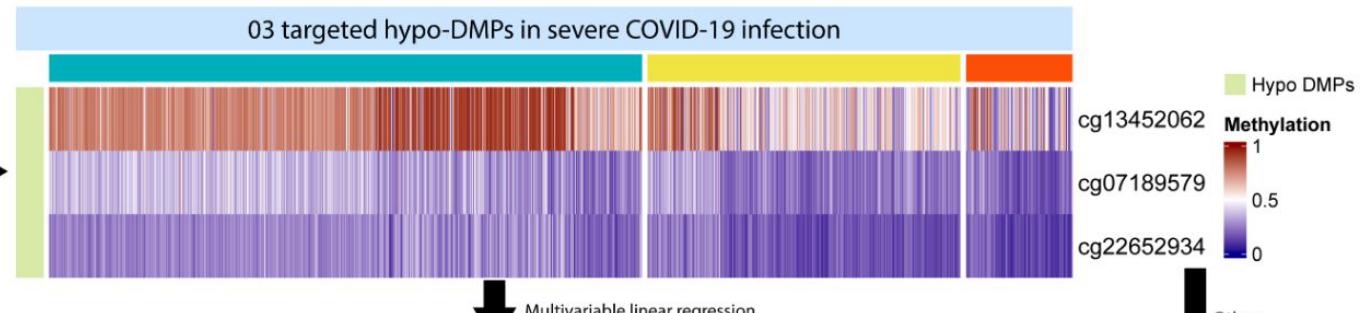
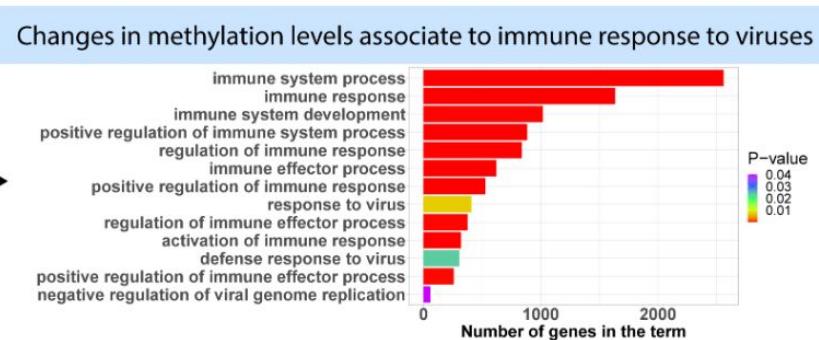
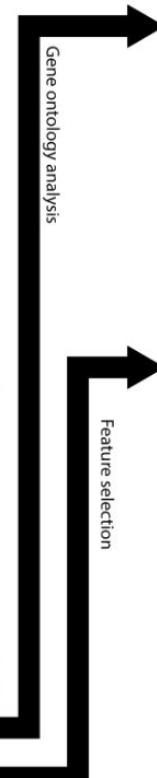
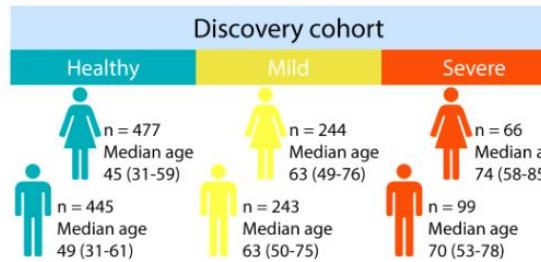
Multi-prediction:

- ## 1. Import data

2 Start prediction

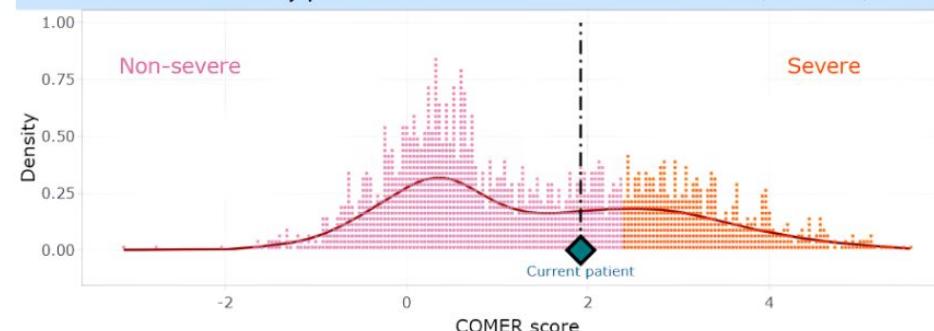
- Click on “Prediction without age” or “Prediction with age” to initiate the prediction

Summary

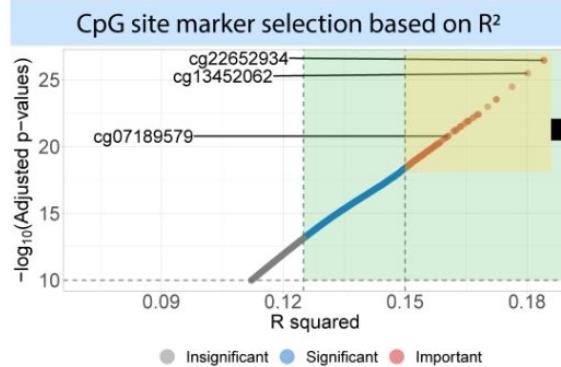
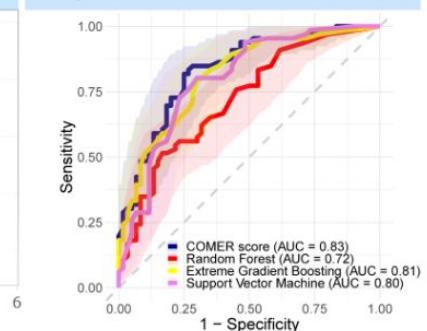


$$\text{COMER score} = 6.9651 - 3.2713 \times \beta_{\text{cg13452062}} - 6.6951 \times \beta_{\text{cg07189579}} - 6.3909 \times \beta_{\text{cg22652934}}$$

COVID-19 severity prediction on COMER score distribution (n = 1700)



Independent validation cohort (n = 126)



Limitation

Lack of available data lead to:

- Some important and qualified studies was not used
- Potential confounders, such as patient co-morbidities, couldn't be explored
- Unreported data regarding the time between blood sampling and occurrence of severe endpoint hindered the analysis of how early the tool can predict severe outcomes

Conclusion

- Three DMPs from public blood DNA methylome datasets were identified to be significantly associated with the severity of COVID-19 infection
- The COVID-19 methylation risk (COMER) score showed good discrimination for severe COVID-19 patients as a simple tool
- Larger cohorts of COVID-19 patients are needed to validate its performance in patients of different races and comorbidities

Acknowledgements



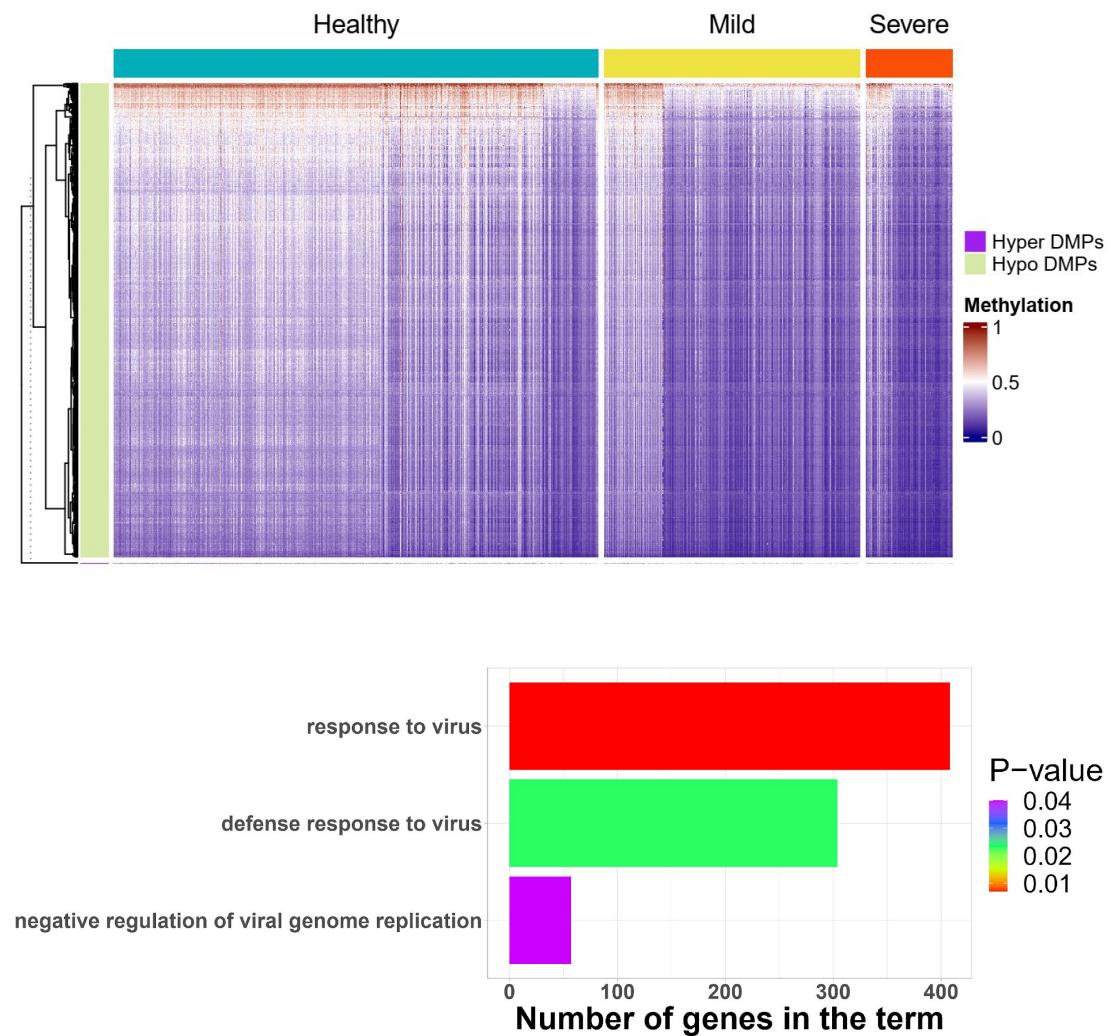
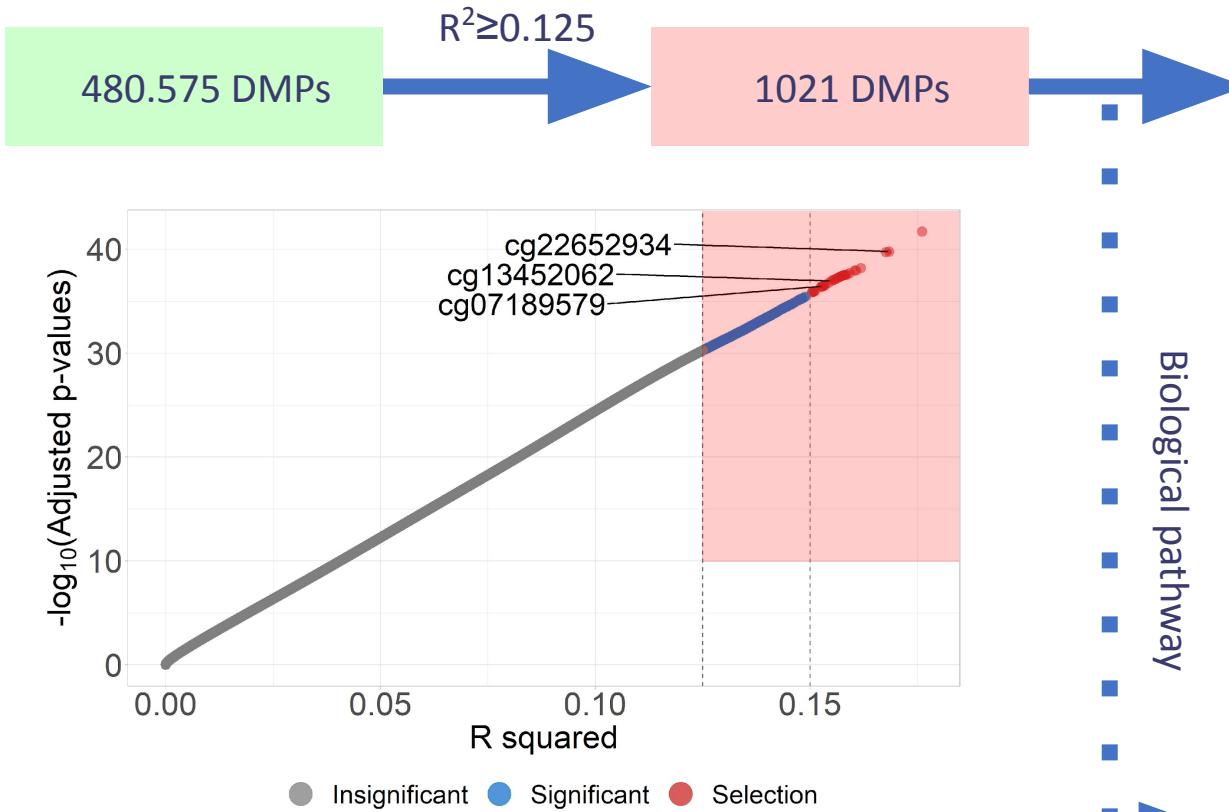
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- All members of Advanced VnPathoinformatics group

All the participants had given their consents in the original studies

THANKS FOR YOUR ATTENTION!

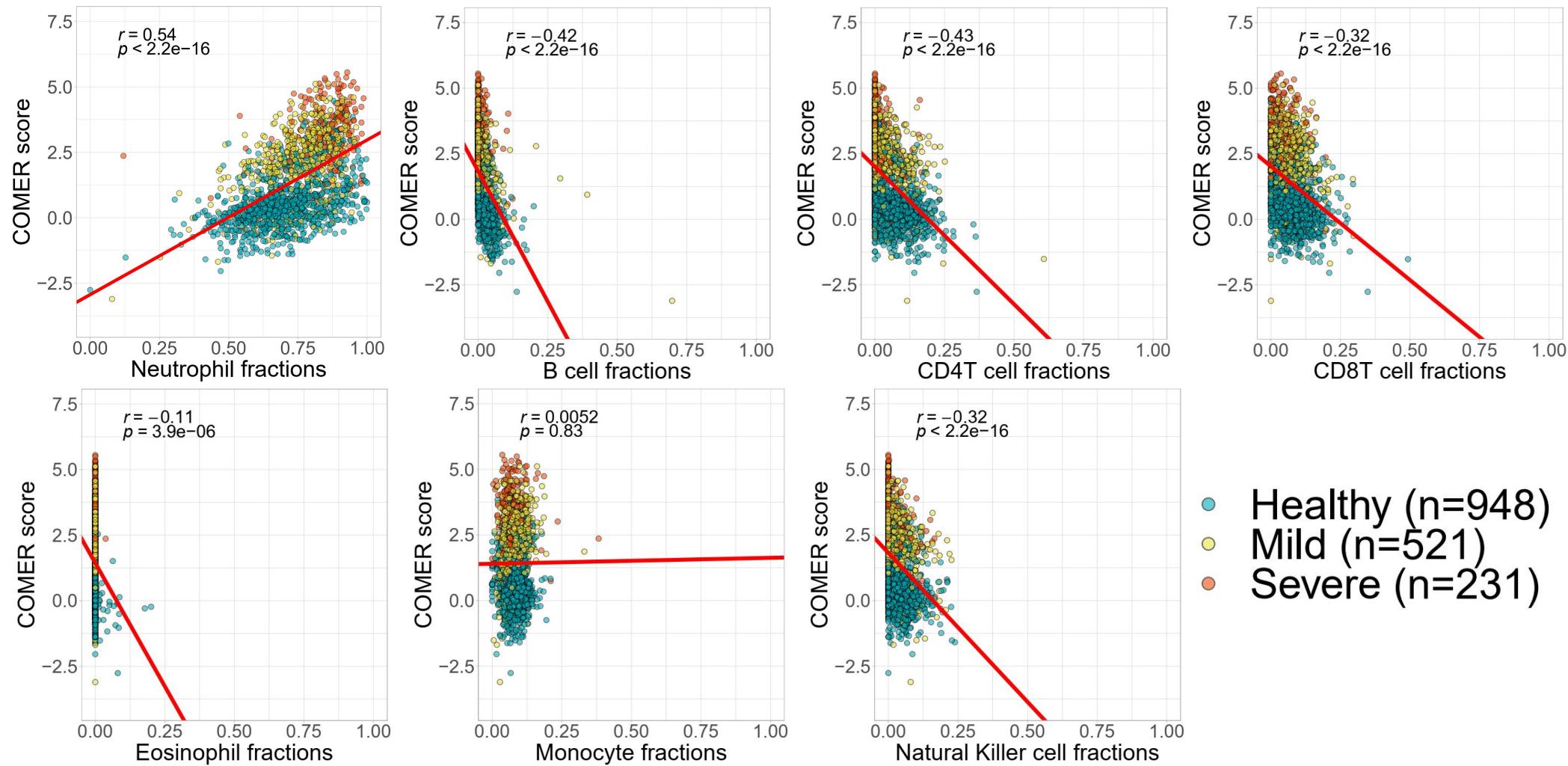
Question time!

NARROW DOWN SIGNIFICANT DMPS



- Reduced DMPs still impact COVID-19 severity, but more DMPs than expected

Association of COMER score and white blood subtypes



COMER score has association with white blood subtypes but Monocyte