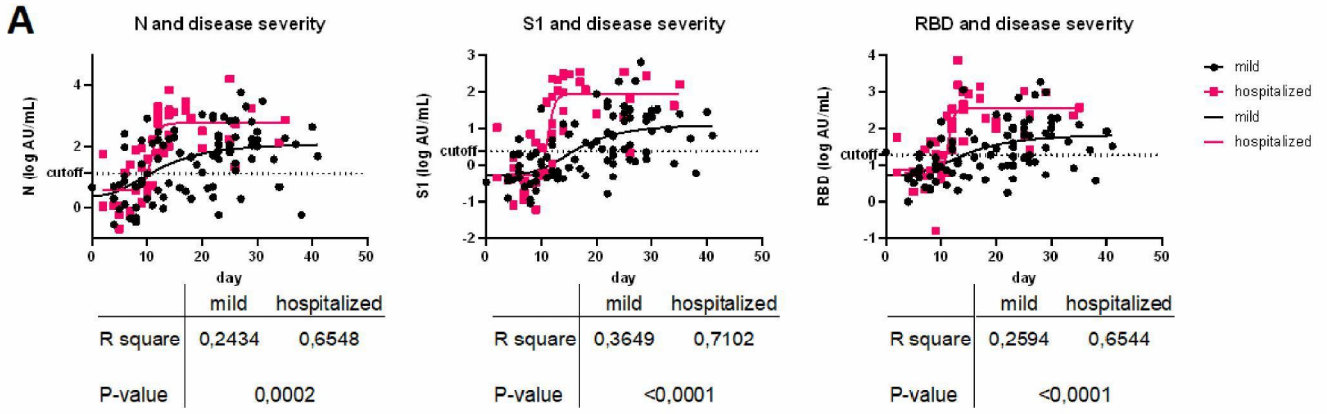


| | | | Specificity-optimized | | Youdens J statistic | |
|------------------|-----------|--------|-----------------------|-------------|---------------------|-------------|
| COVID-19 disease | | | sensitivity | specificity | sensitivity | specificity |
| Single | N | severe | 100 | 98,7 | 100 | 98,2 |
| | | mild | 96,3 | | 85,2 | |
| | S1 | severe | 94,3 | 99,1 | 97,1 | 98,1 |
| | | mild | 76,3 | | 91,3 | |
| | RBD | severe | 94,1 | 98,0 | 97,1 | 94,5 |
| | | mild | 78,3 | | 91,6 | |
| OR | N or S1 | severe | 100 | 97,8 | 100 | 96,3 |
| | | mild | 88,0 | | 92,8 | |
| | N or RBD | severe | 100 | 96,7 | 100 | 92,8 |
| | | mild | 91,7 | | 95,2 | |
| | S1 or RBD | severe | 97,1 | 97,2 | 97,1 | 92,7 |
| | | mild | 90,4 | | 96,4 | |
| N or S1 or RBD | severe | 100 | 95,8 | 100 | 90,9 | |
| | mild | 95,1 | | 98,8 | | |
| AND | N & S1 | severe | 93,5 | 100 | 96,8 | 100 |
| | | mild | 70,5 | | 83,3 | |
| | N & RBD | severe | 93,5 | 100 | 96,8 | 100 |
| | | mild | 69,6 | | 82,3 | |
| | S1 & RBD | severe | 91,2 | 100 | 97,1 | 100 |
| | | mild | 64,6 | | 87,3 | |
| N & S1 & RBD | severe | 90,3 | 100 | 96,8 | 100 | |
| | mild | 59,7 | | 80,5 | | |



B

| | average day of seroconversion | |
|-----|-------------------------------|--------------|
| | mild | hospitalized |
| N | 13,9 | 8,7 |
| S1 | 16,0 | 8,6 |
| RBD | 15,3 | 7,6 |

Latest FFX data 6-5 6pm added
 Add sample/day Susanne Stoof?

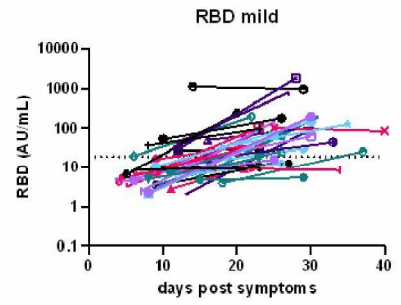
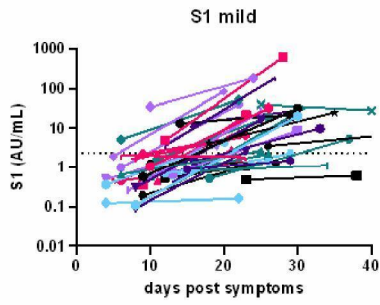
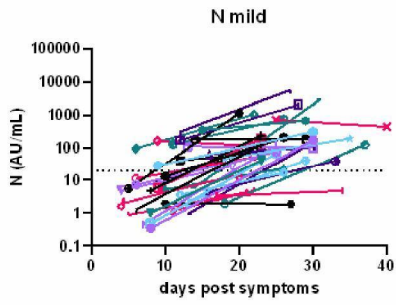
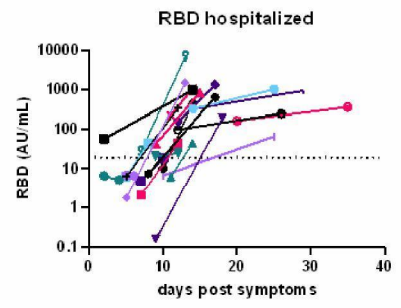
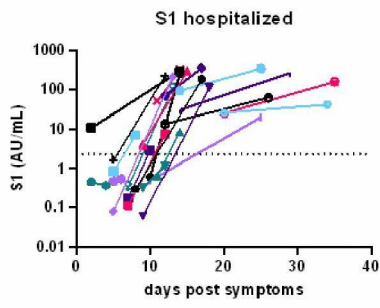
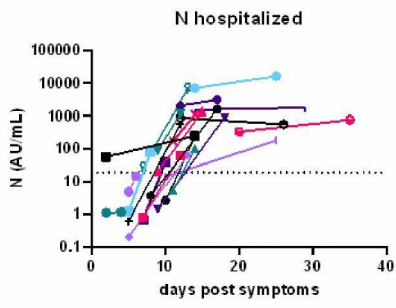
Supplement

Table S1. Prevalence estimated by the test relative to true prevalence and different levels of specificity and sensitivity

| | | true prevalence | | | | | | | relative error | |
|-------------|-----|-----------------|------|------|-----|------|------|------|----------------|----------|
| | | 1 | 3 | 5 | 10 | 20 | 50 | 90 | | |
| sensitivity | 100 | specificity 100 | 1,00 | 3,00 | 5,0 | 10,0 | 20,0 | 50,0 | 90,0 | & /- 5% |
| | | 99 | 1,99 | 3,97 | 6,0 | 10,9 | 20,8 | 50,5 | 90,1 | |
| | | 98 | 2,98 | 4,94 | 6,9 | 11,8 | 21,6 | 51,0 | 90,2 | |
| | | 97 | 3,97 | 5,91 | 7,9 | 12,7 | 22,4 | 51,5 | 90,3 | |
| | | 96 | 4,96 | 6,88 | 8,8 | 13,6 | 23,2 | 52,0 | 90,4 | |
| | | 95 | 5,95 | 7,85 | 9,8 | 14,5 | 24,0 | 52,5 | 90,5 | |
| sensitivity | 98 | specificity 100 | 0,98 | 2,94 | 4,9 | 9,8 | 19,6 | 49,0 | 88,2 | & /- 10% |
| | | 99 | 1,97 | 3,91 | 5,9 | 10,7 | 20,4 | 49,5 | 88,3 | |
| | | 98 | 2,96 | 4,88 | 6,8 | 11,6 | 21,2 | 50,0 | 88,4 | |
| | | 97 | 3,95 | 5,85 | 7,8 | 12,5 | 22,0 | 50,5 | 88,5 | |
| | | 96 | 4,94 | 6,82 | 8,7 | 13,4 | 22,8 | 51,0 | 88,6 | |
| | | 95 | 5,93 | 7,79 | 9,7 | 14,3 | 23,6 | 51,5 | 88,7 | |
| sensitivity | 95 | specificity 100 | 0,95 | 2,85 | 4,8 | 9,5 | 19,0 | 47,5 | 85,5 | & /- 20% |
| | | 99 | 1,94 | 3,82 | 5,7 | 10,4 | 19,8 | 48,0 | 85,6 | |
| | | 98 | 2,93 | 4,79 | 6,7 | 11,3 | 20,6 | 48,5 | 85,7 | |
| | | 97 | 3,92 | 5,76 | 7,6 | 12,2 | 21,4 | 49,0 | 85,8 | |
| | | 96 | 4,91 | 6,73 | 8,6 | 13,1 | 22,2 | 49,5 | 85,9 | |
| | | 95 | 5,90 | 7,70 | 9,5 | 14,0 | 23,0 | 50,0 | 86,0 | |
| sensitivity | 90 | specificity 100 | 0,90 | 2,70 | 4,5 | 9,0 | 18,0 | 45,0 | 81,0 | & /- 5% |
| | | 99 | 1,89 | 3,67 | 5,5 | 9,9 | 18,8 | 45,5 | 81,1 | |
| | | 98 | 2,88 | 4,64 | 6,4 | 10,8 | 19,6 | 46,0 | 81,2 | |
| | | 97 | 3,87 | 5,61 | 7,4 | 11,7 | 20,4 | 46,5 | 81,3 | |
| | | 96 | 4,86 | 6,58 | 8,3 | 12,6 | 21,2 | 47,0 | 81,4 | |
| | | 95 | 5,85 | 7,55 | 9,3 | 13,5 | 22,0 | 47,5 | 81,5 | |
| sensitivity | 85 | specificity 100 | 0,85 | 2,55 | 4,3 | 8,5 | 17,0 | 42,5 | 76,5 | & /- 10% |
| | | 99 | 1,84 | 3,52 | 5,2 | 9,4 | 17,8 | 43,0 | 76,6 | |
| | | 98 | 2,83 | 4,49 | 6,2 | 10,3 | 18,6 | 43,5 | 76,7 | |
| | | 97 | 3,82 | 5,46 | 7,1 | 11,2 | 19,4 | 44,0 | 76,8 | |
| | | 96 | 4,81 | 6,43 | 8,1 | 12,1 | 20,2 | 44,5 | 76,9 | |
| | | 95 | 5,80 | 7,40 | 9,0 | 13,0 | 21,0 | 45,0 | 77,0 | |
| sensitivity | 80 | specificity 100 | 0,80 | 2,40 | 4,0 | 8,0 | 16,0 | 40,0 | 72,0 | & /- 20% |
| | | 99 | 1,79 | 3,37 | 5,0 | 8,9 | 16,8 | 40,5 | 72,1 | |
| | | 98 | 2,78 | 4,34 | 5,9 | 9,8 | 17,6 | 41,0 | 72,2 | |
| | | 97 | 3,77 | 5,31 | 6,9 | 10,7 | 18,4 | 41,5 | 72,3 | |
| | | 96 | 4,76 | 6,28 | 7,8 | 11,6 | 19,2 | 42,0 | 72,4 | |
| | | 95 | 5,75 | 7,25 | 8,8 | 12,5 | 20,0 | 42,5 | 72,5 | |

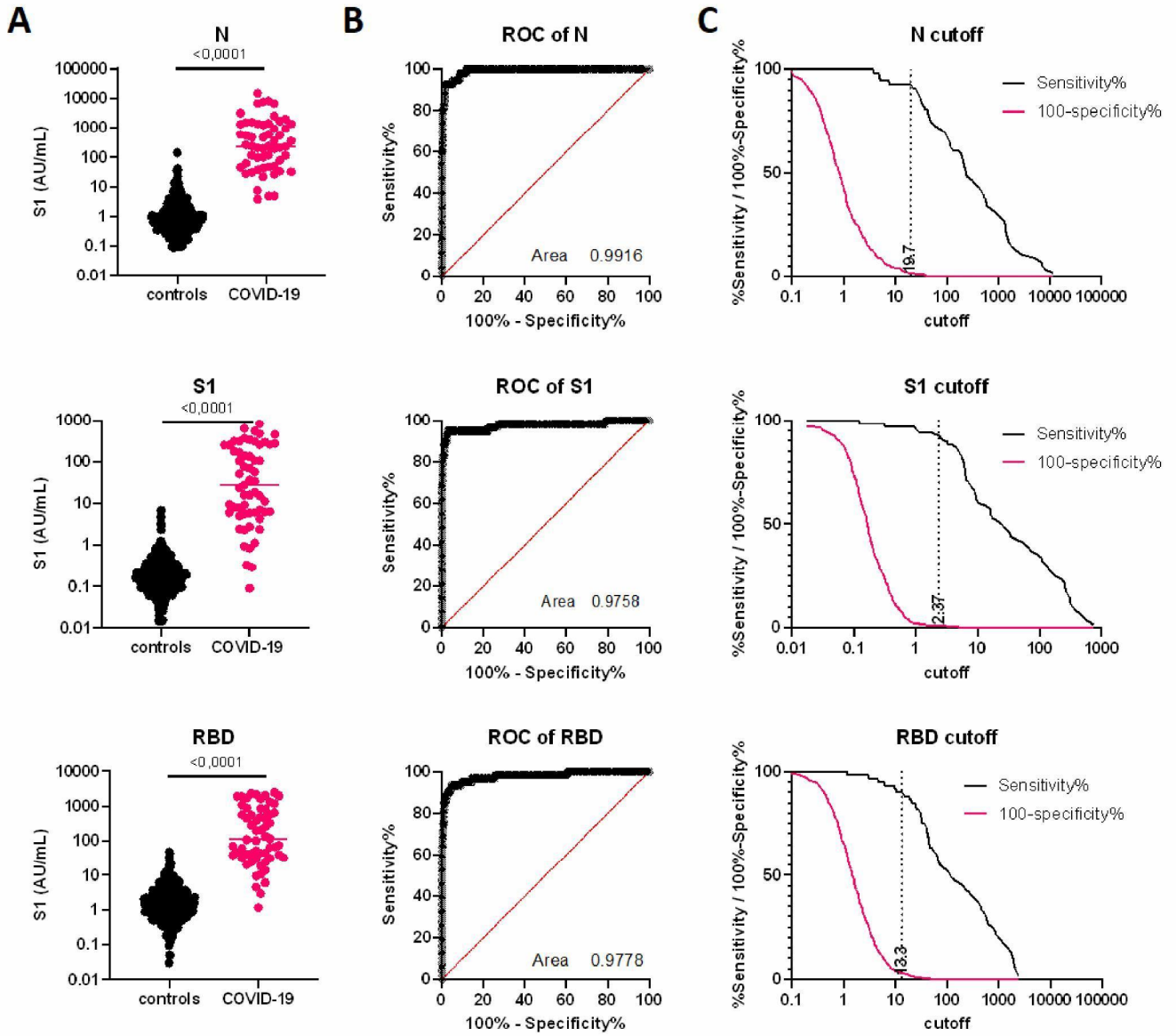
| Supplementary Table. Combining positive concentrations to enhance sensitivity | | | | | | | | | | |
|---|----------------------|--------------|-------------|---------------------|---------------------|----------------------|-------------|-----------|---------------------|---------------------|
| | Specificity cutoff * | | | | | Specificity cutoff * | | | | |
| | %pos | %neg | weight \$ | weighed sensitivity | weighed specificity | %pos | %neg | weight \$ | weighed sensitivity | weighed specificity |
| N or S1 | All controls | 2,717 | 97,3 | | | | 4,9 | 95,1 | | |
| | cross-sectional | 1,63 | 98,4 | 0,7 | | 2,2 | 97,8 | 0,7 | | |
| | ILI non hCoV | 1,333 | 98,7 | 0,15 | | 5,3 | 94,7 | 0,15 | | |
| | ILI hCoV | 5,505 | 94,5 | 0,15 | | 9,2 | 90,8 | 0,15 | | |
| | All COVID-19 | 91,6 | 8,4 | | 91,6 | 95,0 | 5,0 | | 94,9 | |
| | COVID-19 severe | 100,0 | 0,0 | 0,3 | | 100,0 | 0,0 | 0,3 | | |
| COVID-19 mild | 88,0 | 12,0 | 0,7 | | 92,8 | 7,2 | 0,7 | | | |
| N or RBD | All controls | 3,523 | 96,5 | | | 7,6 | 92,4 | | | 92,8 |
| | cross-sectional | 3,261 | 96,7 | 0,7 | | 7,1 | 92,9 | 0,7 | | |
| | ILI non hCoV | 1,333 | 98,7 | 0,15 | | 5,3 | 94,7 | 0,15 | | |
| | ILI hCoV | 5,455 | 94,5 | 0,15 | | 10,0 | 90,0 | 0,15 | | |
| | All COVID-19 | 94,1 | 5,9 | | 94,2 | 96,6 | 3,4 | | 96,7 | |
| | COVID-19 severe | 100,0 | 0,0 | 0,3 | | 100,0 | 0,0 | 0,3 | | |
| COVID-19 mild | 91,7 | 8,3 | 0,7 | | 95,2 | 4,8 | 0,7 | | | |
| S1 or RBD | All controls | 2,981 | 97,0 | | | 8,1 | 91,9 | | | 92,7 |
| | cross-sectional | 2,717 | 97,3 | 0,7 | | 6,0 | 94,0 | 0,7 | | |
| | ILI non hCoV | 2,667 | 97,3 | 0,15 | | 10,7 | 89,3 | 0,15 | | |
| | ILI hCoV | 3,636 | 96,4 | 0,15 | | 10,0 | 90,0 | 0,15 | | |
| | All COVID-19 | 92,4 | 7,6 | | 92,4 | 96,6 | 3,4 | | 96,6 | |
| | COVID-19 severe | 97,1 | 2,9 | 0,3 | | 97,1 | 2,9 | 0,3 | | |
| COVID-19 mild | 90,4 | 9,6 | 0,7 | | 96,4 | 3,6 | 0,7 | | | |
| N or S1 or RBD | All controls | 4,6 | 95,4 | | | 10,3 | 89,7 | | | 90,9 |
| | cross-sectional | 3,8 | 96,2 | 0,7 | | 7,6 | 92,4 | 0,7 | | |
| | ILI non hCoV | 2,7 | 97,3 | 0,15 | | 10,7 | 89,3 | 0,15 | | |
| | ILI hCoV | 7,3 | 92,7 | 0,15 | | 14,7 | 85,3 | 0,15 | | |
| | All COVID-19 | 96,6 | 3,4 | | 96,6 | 99,1 | 0,86 | | 99,1 | |
| | COVID-19 severe | 100 | 0 | 0,30 | | 100 | 0 | 0,30 | | |
| COVID-19 mild | 95,1 | 4,9 | 0,70 | | 98,8 | 1,2 | 0,70 | | | |

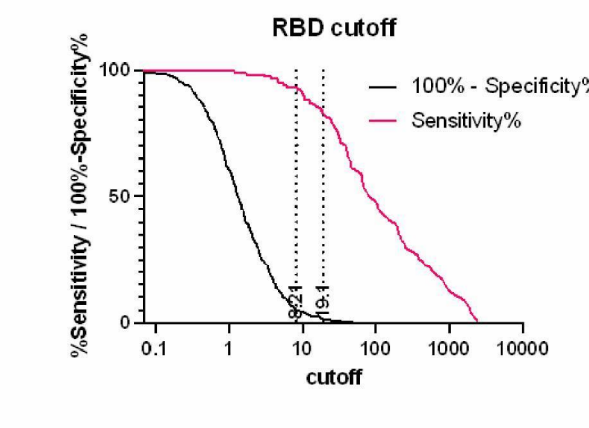
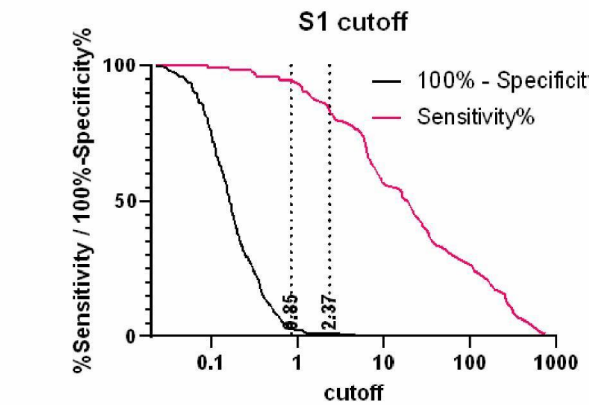
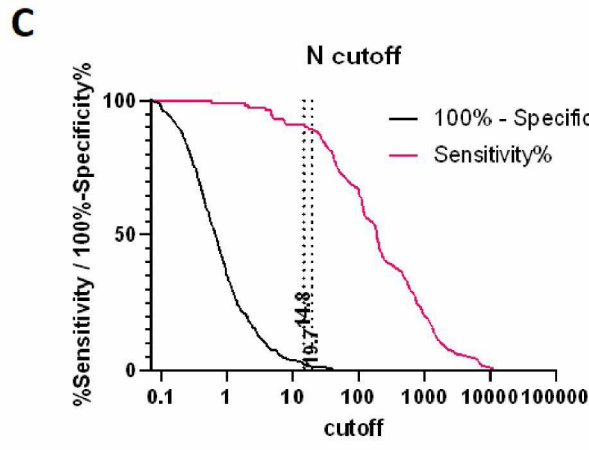
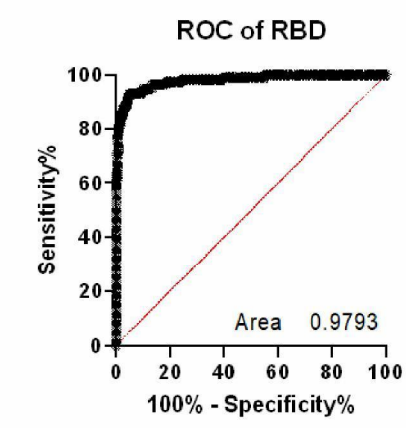
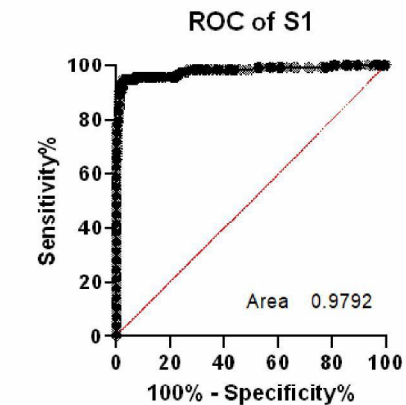
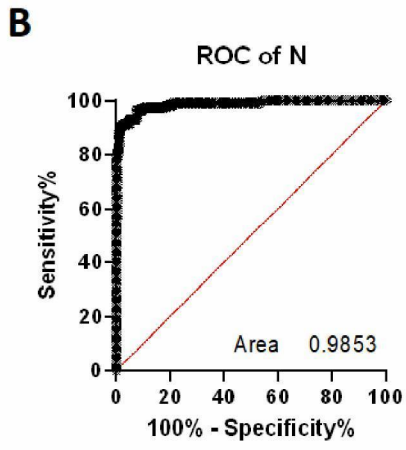
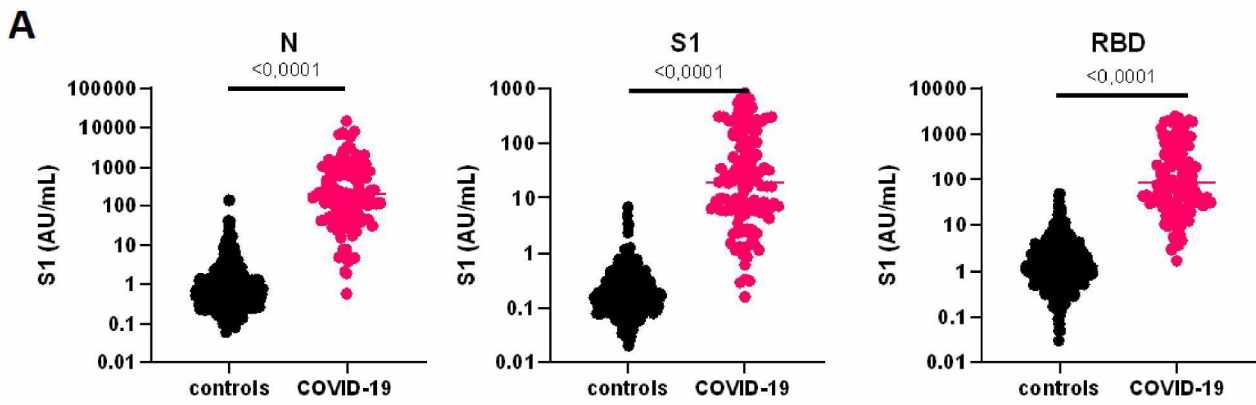
* cutoffs: N= 19,7; S1=2.37; RBD=19.1 ; # cutoffs N=14.8; S1=0.85; RBD=8.21 ; \$ weighing is applied to better reflect the frequencies of patients



Version 6mei
Probably final

Archive





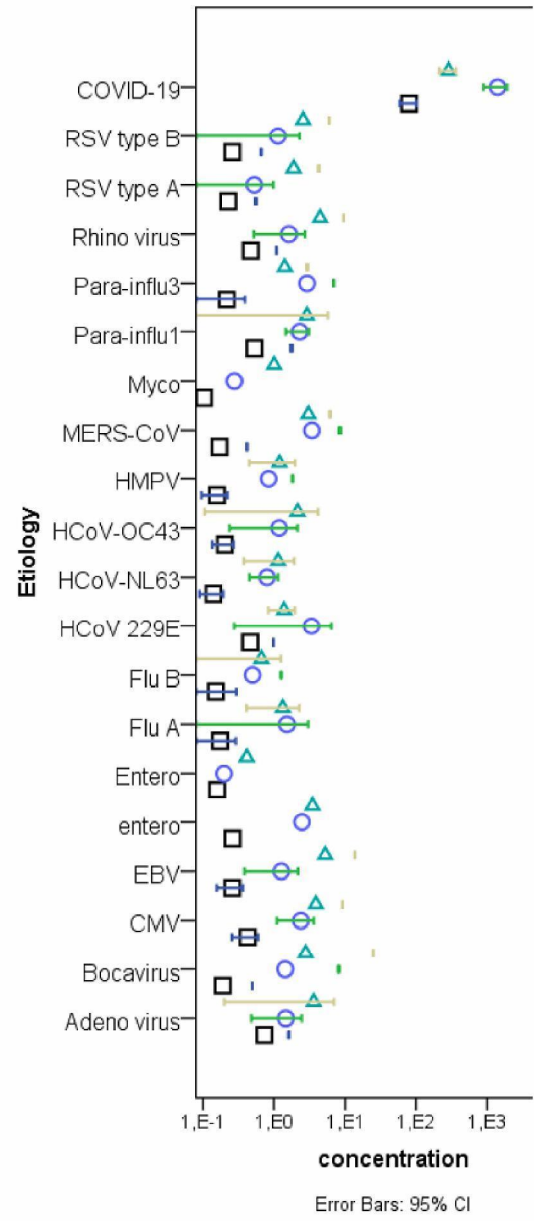
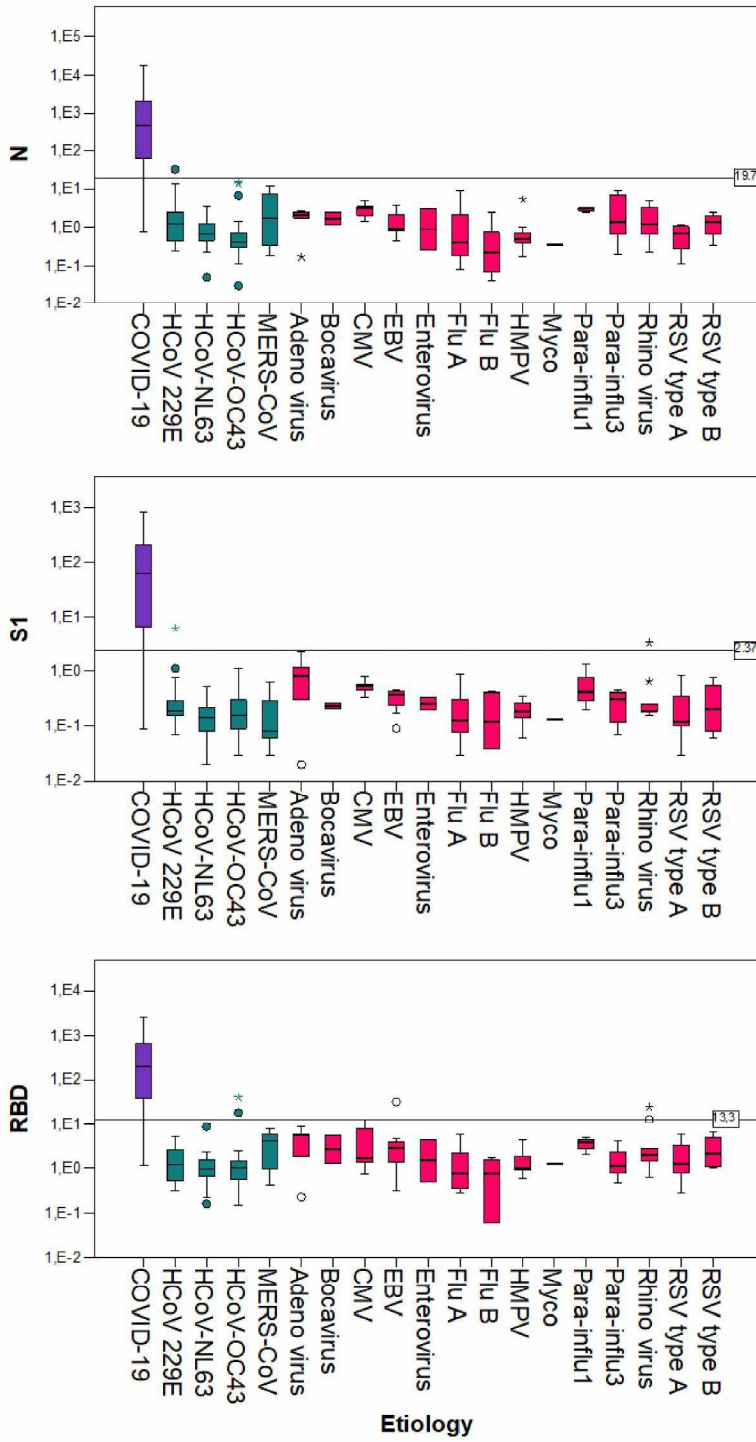


Table 1. IgG concentrations to SARS-CoV-2 antigens in controls and COVID-19 patients

| | N | S1 | RBD |
|---------------------------|---------------------|---------------------|---------------------|
| Reproducibility * | N=268 | N=268 | N=214 |
| R square | 0.982 | 0.985 | 0.988 |
| CV% | 19.1 | 25.5 | 14.6 |
| Seroconversion # | | | |
| Controls | 3.125 (1.803-4.446) | 0.515 (0.408-0.621) | 2.863 (2.350-3.377) |
| COVID-19 patients | 558.7 (199.4-918.0) | 76.22 (41.08-111.4) | 519.7 (334.2-705.1) |
| Mean difference | 555.6 (425.9-685.3) | 75.70 (62.68-88.73) | 516.8 (442.9-590.7) |
| | P<0.0001 | P<0.0001 | P<0,0001 |
| ROC analyses † | | | |
| Specificity Cutoff | 19.7 | 2.37 | 19.1 |
| Sensitivity | 89.4 | 84.4 | 83.6 |
| Specificity | 98.5 | 99.0 | 98.5 |
| Youden J statistic Cutoff | 14.8 | 0.85 | 8.21 |
| Sensitivity | 91.2 | 95.7 | 94.0 |
| Specificity | 97.8 | 97.3 | 95.1 |

* Performance data of the assay. Raw data shown in Fig 1.

Data show mean and 95% confidence intervals. Statistical test results are based on log-transformed data. Untransformed data are shown in Fig 2.

† Roc statistics of Fig 3.

N N=268
R square 0.982
CV% 19.1

Table 2. Sensitivity of the multiplex assay using different cutoffs and antibody results

| | Specificity cutoff * | | | | | Youdens'J statistic cutoff # | | | | |
|-----------------------------|----------------------|-------------|-------------|---------------------|---------------------|------------------------------|-------------|-------------|---------------------|---------------------|
| | %pos | %neg | weight \$ | weighed sensitivity | weighed specificity | %pos | %neg | weight \$ | weighed sensitivity | weighed specificity |
| N & S1 & RBD | All controls | 0 | 100 | | | 100 | 0 | 100 | | |
| | cross-sectional | 0 | 100 | 0,70 | | | 0 | 100 | 0,70 | |
| | ILI non hCoV | 0 | 100 | 0,15 | | | 0 | 100 | 0,15 | |
| | ILI hCoV | 0 | 100 | 0,15 | | | 0 | 100 | 0,15 | |
| | All COVID-19 | 68,5 | 31,5 | | 68,9 | | 85,2 | 14,8 | | 85,4 |
| | COVID-19 severe | 90,3 | 9,7 | 0,30 | | | 96,8 | 3,2 | 0,30 | |
| COVID-19 mild | 59,7 | 40,3 | 0,70 | | | 80,5 | 19,5 | 0,70 | | |
| N & S1 | All controls | 0 | 100 | | | 100 | 0 | 100 | | |
| | cross-sectional | 0 | 100 | 0,70 | | | 0 | 100 | 0,70 | |
| | ILI non hCoV | 0 | 100 | 0,15 | | | 0 | 100 | 0,15 | |
| | ILI hCoV | 0 | 100 | 0,15 | | | 0 | 100 | 0,15 | |
| | All COVID-19 | 77,1 | 22,9 | | 77,4 | | 87,2 | 12,8 | | 87,4 |
| | COVID-19 severe | 93,5 | 6,5 | 0,30 | | | 96,8 | 3,2 | 0,30 | |
| COVID-19 mild | 70,5 | 29,5 | 0,70 | | | 83,3 | 16,7 | 0,70 | | |
| N & RBD | All controls | 0 | 100 | | | 100 | 0 | 100 | | |
| | cross-sectional | 0 | 100 | 0,70 | | | 0 | 100 | 0,70 | |
| | ILI non hCoV | 0 | 100 | 0,15 | | | 0 | 100 | 0,15 | |
| | ILI hCoV | 0 | 100 | 0,15 | | | 0 | 100 | 0,15 | |
| | All COVID-19 | 76,4 | 23,6 | | 76,8 | | 86,4 | 13,6 | | 86,6 |
| | COVID-19 severe | 93,5 | 6,5 | 0,30 | | | 96,8 | 3,2 | 0,30 | |
| COVID-19 mild | 69,6 | 30,4 | 0,70 | | | 82,3 | 17,7 | 0,70 | | |
| S1 & RBD | All controls | 0 | 100 | | | 100 | 0 | 100 | | |
| | cross-sectional | 0 | 100 | 0,70 | | | 0 | 100 | 0,70 | |
| | ILI non hCoV | 0 | 100 | 0,15 | | | 0 | 100 | 0,15 | |
| | ILI hCoV | 0 | 100 | 0,15 | | | 0 | 100 | 0,15 | |
| | All COVID-19 | 72,6 | 27,4 | | 72,5 | | 90,3 | 9,7 | | 90,3 |
| | COVID-19 severe | 91,2 | 8,8 | 0,30 | | | 97,1 | 2,9 | 0,30 | |
| COVID-19 mild | 64,6 | 35,4 | 0,70 | | | 87,3 | 12,7 | 0,70 | | |
| N or S1 or RBD | All controls | 4,6 | 95,4 | | | 95,8 | 10,3 | 89,7 | | 90,9 |
| | cross-sectional | 3,8 | 96,2 | 0,7 | | | 7,6 | 92,4 | 0,7 | |
| | ILI non hCoV | 2,7 | 97,3 | 0,15 | | | 10,7 | 89,3 | 0,15 | |
| | ILI hCoV | 7,3 | 92,7 | 0,15 | | | 14,7 | 85,3 | 0,15 | |
| | All COVID-19 | 96,6 | 3,4 | | 96,1 | | 99,1 | 0,86 | | 99,0 |
| | COVID-19 severe | 100 | 0 | 0,20 | | | 100 | 0 | 0,20 | |
| COVID-19 mild | 95,1 | 4,9 | 0,80 | | | 98,8 | 1,2 | 0,80 | | |

* cutoffs: N= 19,7; S1=2.37; RBD=19.1 ; # cutoffs N=14.8; S1=0.85; RBD=8.21 ; \$ weighing is applied to better reflect the frequencies of patients

With FFX t2 data may4th added

