Supplementary Appendix of

Accuracy of COVID-19 self-tests with unsupervised nasal or nasal plus oropharyngeal self-sampling in symptomatic individuals in the Omicron period

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Supplementary material 1: short questionnaire after performing both self-tests at home (translated from Dutch). Questions or items that were added for phase-2 (oropharyngeal + nasal sampling) are indicated in red.

1. What was the result of the self-test you just performed for this study?
   - ☐ Negative
   - ☐ Positive
   - ☐ Invalid
   - ☐ I don't know: unable to perform the test
   - ☐ I don't know: unable to read the test result
   - ☐ Other, ……

2. Which sampling technique did you apply when performing the self-test, you just performed for this study?
   - ☐ first oropharyngeal, then nasal sampling
   - ☐ first nasal, then oropharyngeal sampling
   - ☐ oropharyngeal sampling only
   - ☐ nasal sampling only

3. What is your gender?
   - ☐ Male
   - ☐ Female
   - ☐ Other (non-binary / genderqueer)

4. What is your age?
   - ☐☐ years

5. What is the reason for testing?
   Multiple answers possible
   - ☐ I have (had) Covid-19 like symptoms
   - ☐ I am a close contact of a SARS-CoV-2 infected person
     - Date of last contact: ___ - ___ (day – month)

   I was notified because of:
   - The infected person is a household member □ yes / □ no
   - The infected person is a close contact (more than 15 minutes within 1.5 meters), but not a household member □ yes / □ no
   - Received notification by CoronaMelder app (English: Corona notification app) □ yes / □ no
   - Received notification by public health service (by phone or letter) □ yes / □ no
   - Received notification by SARS-CoV-2 infected person □ yes / □ no
   - Other, ……

   - ☐ GP recommended a SARS-CoV-2 test
   - ☐ Travelled to a high-pandemic country
   - ☐ I performed a self-test and tested positive
     - What was the date of the positive self-test: ___ - ___ (day – month)
   - ☐ None of the above, but…….

6. Did you receive a Covid-19 vaccination?
   - ☐ No ***GO TO QUESTION 8***
   - ☐ Yes

   If yes, how many vaccinations did you receive?
   - ☐ One
   - ☐ Two
   - ☐ Three
   - ☐ Four

   If yes, which vaccine did you receive (tip: this can be found in the CoronaCheck QR-code app under "International"/"Vaccination certificate")?
   Vaccine 1:
   - ☐ Pfizer
   - ☐ Moderna
   - ☐ AstraZeneca

☐ Janssen
☐ Unknown/Other

Vaccine 2:
☐ Pfizer
☐ Moderna
☐ AstraZeneca
☐ Janssen
☐ Unknown/Other

Vaccine 3:
☐ Pfizer
☐ Moderna
☐ AstraZeneca
☐ Janssen
☐ Unknown/Other

Vaccine 4:
☐ Pfizer
☐ Moderna
☐ AstraZeneca
☐ Janssen
☐ Unknown/Other

7. What was the date of the last vaccination (tip: this can be found in the CoronaCheck QR-code app under "International"/"Vaccination certificate")?
___ - ___ - ___ (day – month – year)

8. Did you have a positive SARS-CoV-2 test previously?
☐ No ***GO TO QUESTION 9***
☐ Yes

If yes, how often
☐☐ times

If yes, how long ago?
☐ < 2 months
☐ 2-6 months
☐ 6-12 months
☐ >12 months

If yes, how was the SARS-CoV-2 infection detected?
☐ via public health service
☐ via a commercial test sites ("testing for access" or "testing for travel")
☐ via a self-test
☐ Other (e.g., in another country, hospital, or at work): ……

If yes, via a
☐ PCR test
☐ Rapid antigen test
☐ Unknown

If yes, did this positive test occur after you had been vaccinated against COVID-19?
☐ No
☐ Yes, after:
  ☐ the first vaccination

☐ the second vaccination
☐ the third vaccination
☐ the fourth vaccination

9. At this moment, do you have any COVID-19 like symptoms?
☐ No ***GO TO QUESTION 12***
☐ Yes

10. What COVID-19 like symptoms do you currently have?
Multiple answers possible
☐ Common cold
☐ Shortness of breath
☐ Fever
☐ Coughing
☐ Loss of taste or smell
☐ Muscle ache
☐ I have other symptoms: …….

11. What was the moment you first experienced these symptoms?
☐ Today
☐ Yesterday
☐ Two days ago
☐ Three or more days ago, at …/… (day/month)

12. Have you performed a self-test before?
☐ No ***GO TO QUESTION 13***
☐ Yes

If yes, how often?
☐☐ times

If yes, why?
☐ because of COVID-19 related symptoms
☐ because of an infected household member
☐ because of a close contact of an infected individual
☐ because I had to / it was recommended by my employer
☐ because I wanted to prevent me from potentially infected someone else because of the following situation: ……
☐ Other: ……

If yes, when did you perform your most recent self-test?
☐ less than 7 days ago
☐ 1-4 weeks ago
☐ 2-3 months ago
☐ 3-6 months ago
☐ more than 6 months ago

If yes, how did you obtain the self-test?
☐ received it from the government
☐ store-bought
☐ received it from school
☐ received it from work
☐ other: ……

13. Did you experience any difficulties when performing the self-test in this study?
☐ No ***GO TO QUESTION 14***
☐ Yes

If yes,

☐ I had a problem when performing the test:
- ☐ Unsure how deep to place the swab up my throat
- ☐ Fear of placing the swab too far up my throat
- ☐ I had a gag reflex when placing the swab up my throat
- ☐ Unsure how deep to place the swab up my nose
- ☐ Fear of placing the swab too far up my nose
- ☐ Spilled fluid
- ☐ Other ……

☐ I had a problem interpreting the test:
- ☐ I saw no control line
- ☐ Liquid didn't travel through the cassette
- ☐ Unclear whether the test was positive
- ☐ Other ……

14. How difficult was it to perform oropharyngeal self-sampling, on a scale from 1 to 5?

Very easy *1* *2* *3* *4* *5* *very hard*

15. How difficult was it to perform nasal self-sampling, on a scale from 1 to 5?

Very easy *1* *2* *3* *4* *5* *very hard*

16. Do you agree with the following statements?

The instruction for use of the self-test used in this study was clear
Agree *In doubt* *Disagree*

I am confident I performed the self-test used in this study correctly
Agree *In doubt* *Disagree*

I am confident I interpreted the self-test used in this study correctly
Agree *In doubt* *Disagree*

In the future, I will perform additional oropharyngeal sampling more often
Agree *In doubt* *Disagree*

I would rather perform nasal sampling only (instead of nasal plus oropharyngeal sampling)
Agree *In doubt* *Disagree*

Supplementary material 2: short questionnaire provided 10 days after test site visit (translated from Dutch). Questions or items that were added for phase-2 (oropharyngeal + nasal sampling) are indicated in red.

1. Did you develop any COVID-19 like symptoms since your corona test at the public health service test site and the self-test performed for this study 10 days ago (see next question for list of possible symptoms)?
   - ☐ I had symptoms 10 days ago already and I experienced no new symptoms ***GO TO QUESTION 4***
   - ☐ I had symptoms 10 days ago already and I experienced new symptoms
   - ☐ I had no symptoms 10 days ago, but I experienced new symptoms
   - ☐ I had no symptoms 10 days ago and I experienced no new symptoms ***GO TO QUESTION 4***

2. What COVID-19 like symptoms did you have?
   Multiple answers possible
   - ☐ Common cold
   - ☐ Shortness of breath
   - ☐ Fever
   - ☐ Coughing
   - ☐ Loss of taste or smell
   - ☐ Muscle ache
   - ☐ I have other symptoms: …….

3. Since when do you have these symptoms?
   … / … / … (day/month/year)

4. Did you perform a corona test or was a corona test performed in the last 10 days since your corona test at the public health service test site and the self-test performed for this study?
   - ☐ No ***THIS CONCLUDES THIS QUESTIONNAIRE***
   - ☐ Yes

   If yes, what type of test was performed?
   - ☐ Test at a public health service test site
     - ☐ PCR
     - ☐ (Rapid) antigen test
     - ☐ I don't know
   - ☐ Test at another test location (e.g., a commercial test site or at work)
     - ☐ PCR
     - ☐ (Rapid) antigen test
     - ☐ I don't know
   - ☐ Test at the hospital or general practitioner
     - ☐ PCR
     - ☐ (Rapid) antigen test
     - ☐ I don't know
   - ☐ Self-test
     - ☐ with nasal sampling
     - ☐ with nasal and oropharyngeal sampling
     - ☐ with oropharyngeal sampling
   - ☐ Other, ……

5. What is the reason for testing?
   Multiple answers possible
   - ☐ I have (had) Covid-19 like symptoms
   - ☐ I am a close contact of a corona infected person
     - Date of last contact: ___ - ___ (day – month)

   I was notified because of:
   - The infected person is a household member □ yes / □ no
   - The infected person is a close contact (more than 15 minutes within 1.5 meters), but not a household member □ yes / □ no
   - Received notification by CoronaMelder app (English: Corona notification app) □ yes / □ no
   - Received notification by public health service (by phone or letter) □ yes / □ no

- Received notification by a corona infected person □ yes / □ no
- Other, ……

☐ GP recommended a corona test
☐ Travelled to a high-pandemic country
☐ I performed a self-test and tested positive
- What was the date of the positive self-test: ___ - ___ (day – month)

☐ None of the above, but…….

6. Was at least one of these new corona tests positive?
   ☐ No
   ☐ Yes
   ☐ Unclear (result of self-test not clear clear), because…….

   If yes, what was the date of this positive test: ___ - ___ (day – month)

Supplementary material 3: Specimen collection, SARS-CoV-2 diagnostic testing, and SARS-CoV-2 virus culture procedures

Specimen collection and SARS-CoV-2 diagnostic testing procedures

West Brabant region, Microvida laboratory at Bravis hospital in Roosendaal; samples from Roosendaal

One combined oropharyngeal and nasal swab (about 2.5 cm deep from the edge of the nostril) was taken per person at the Public Health Service test sites at the test sites in Roosendaal, placed in universal transport media (HiViralTM) with MagnaPure LC lysis- and binding buffer (Roche Diagnostics, The Netherlands), and transported to the Microvida laboratory in Roosendaal. RT-PCR was used as the reference standard test. RT-PCR was performed using the Cobas SARS-CoV-2 test on either the Cobas 6800 or the Cobas 8800 platform (Roche Diagnostics International, Rotkreuz, Switzerland). Cycle threshold (Ct) values for the SARS-CoV-2 E-gene were converted to viral load (genome copies/ml) using a previously established standard curve.[1,2] The assay was used in accordance with the manufacturer's instructions; amplification curves and Ct values were interpreted using the manufacturer's interpretation algorithms, which complied with the European in-vitro diagnostic devices directive.

Central- and Northeast Brabant region, Sanquin Blood Supply Foundation in Amsterdam; samples from Tilburg (Ledeboer and Ringbaan-West)

One superficial combined oropharyngeal and nasopharyngeal swab (>5 cm deep from the edge of the nostril) was taken per person at the Public Health Service test sites at the test sites in Tilburg, placed in universal transport media (HiViralTM) with MagnaPure LC lysis- and binding buffer (Roche Diagnostics, The Netherlands), and transported to the Sanquin Blood Supply Foundation in Amsterdam. RT-PCR was used as the reference standard test. RT-PCR was performed using the Cobas SARS-CoV-2 test on the Cobas 8800 platform (Roche Diagnostics International, Rotkreuz, Switzerland). Cycle threshold (Ct) values for the SARS-CoV-2 E-gene were converted to viral load (genome copies/ml) using a previously established standard curve.[1,2] The assay was used in accordance with the manufacturer's instructions; amplification curves and Ct values were interpreted using the manufacturer's interpretation algorithms, which complied with the European in-vitro diagnostic devices directive.

Rotterdam region, Reinier Haga Medical Diagnostic center in Delft: samples from Rotterdam (Rotterdam-The Hague Airport, Groothandelsgebouw) and Capelle aan den IJssel (sporthal Schenkel)

One deep combined oropharyngeal and nasopharyngeal swab (>5 cm deep from the edge of the nostril) was taken per person at the Public Health service test site in the Rotterdam region, placed directly in universal transport media (HiViralTM) with MagnaPure LC lysis- and binding buffer (Roche Diagnostics, The Netherlands), and transported to the Reinier Haga Medical Diagnostic Center. RT-PCR was used as the reference standard test. RT-PCR testing was performed in virus transport medium using the Cobas SARS-CoV-2 test on the Cobas 6800 platform. The assay was used in accordance with the manufacturer's instructions; amplification curves and cycle threshold values were interpreted using the manufacturer's interpretation algorithms, which complied with the European in-vitro diagnostic devices directive.

Viral load calculation

We used a viral load cut-off as a proxy of infectiousness of ≥5.2 log10 SARS-CoV-2 E-gene copies/mL, which was the viral load cut-off above which 95% of people with a positive RT-PCR test had a positive virus culture in a recent previous study by our group.[3] For that study, the Erasmus MC Viroscience laboratory created a standard curve by testing dilutions of a specific quantified E-gene transcript (primary standard) available from the European Virus Archive (EVAg[1]) with the RT-PCR described by Corman et al.[2] The relation between Ct value and number of E-gene copies/ml was determined by linear regression analysis. Based on this calibration curve a secondary standard derived from cell-cultured virus was prepared and quantified. Serial dilutions of this secondary standard were tested by RT-PCR to prepare a secondary standard curve by linear regression.

In that previous study, we determined whether the Cobas PCR platform at the Microvida laboratory provided comparable data to the Cobas PCR platform at the Erasmus Medical Center Viroscience laboratory by having both laboratories test the same SARS-CoV-2 viral load panel obtained from the National Public Health Institute (RIVM). The Ct values generated in the two laboratories corresponded well. Therefore, we concluded that the same formula to convert Ct values into viral loads (copies/ml) could be used for the West-Brabant and Rotterdam laboratories. This formula was $62.5 * e^{\frac{43.1-Ct}{1.607}}$. Similarly, and as an extension, the most recent (October 2021) SARS-CoV-2 viral load panel (LEQA4) obtained from the National Public Health Institute (RIVM) was tested at the Sanquin Blood Supply Foundation and the Reinier Haga Medical Diagnostic Center. The Cobas platforms at all laboratories generated Ct values that were comparable. Therefore, we concluded that the same formula could be applied to all three laboratories used in the current study.

The three laboratories used different medium/reagent volumes per swab. For each, a factor *X* was calculated based on the swab volume, dilution, and total sample volume, to allow alignment across samples with respect to the viral load in the Erasmus MC Viroscience laboratory standard. For example, if the sample volume is equal (3 ml in Tilburg compared to 3 ml in Rotterdam), but the dilution factor is higher, e.g., by the addition of lysis buffer (3 in Tilburg compared to 2 in Rotterdam), the effective viral sample would be lower than in the Rotterdam standard, thereby underestimating the actual viral load when using the above formula. To account for this, a factor $X < 1$ was used, here 6/9. If dilution was lower, a factor $X > 1$ was used. The conversion formula used therefore was $62.5 * e^{\frac{43.1-Ct}{1.607}} / X$.

Self-tests

Acon Labs Flowflex COVID-19 Antigen Home Test

The Flowflex COVID-19 Antigen Home Test is a CE-marked Ag-RDT manufactured and distributed by Acon Laboratories, Inc. San Diego, CA, USA. According to the instructions for use, a sample should be taken by placing a nasal swab less than 2.5 cm from the nostril edge in the nose and twisting it 5 times, once in each nostril. Afterwards the nose swab had to be placed in a test tube that was filled with buffer fluid and twisted for 30 seconds. Then the swab had to be twisted 5 times during which the test tube had to be squeezed lightly. While squeezing the tube, the swab could be removed from the tube, while leaving all the test fluid in the tube. After placing a cap on the tube, the tube had to be moved from left to right. Then, 4 droplets of the solvent had to be added to the testing cassette. After 15 minutes, but no later than 30 minutes, the participant had to read the test result visually from the testing cassette.

MP Biomedicals Rapid SARS-CoV-2 Antigen Test Card

The Rapid SARS-CoV-2 Antigen Test Card is a CE-marked Ag-RDT, originally manufactured by Xiamen Boson Biotech Co., Ltd., Xiamen, China and distributed by MP Biomedicals Germany GmbH, Eschwege, Germany. According to the instructions for use, first, extraction fluid had to be placed into a test tube. A sample should be taken by placing a nasal swab at least 2.5 cm deep in the nose and twisting it 3-4 times, once in each nostril. Afterwards the nose swab had to be placed in a test tube that was filled by buffer fluid and twisted 3-5 times, after which the swab had to be left in the test tube for 1 minute. While squeezing the tube, the swab could be removed from the tube, while leaving all the test fluid in the tube. Then, after placing a cap on the tube, 3 droplets of the solvent had to be added to the testing cassette. After 15 to 20 minutes, but no later than 20 minutes, the participant had to read the test result visually from the testing cassette.

Siemens-Healthineers CLINITEST Rapid COVID-19 Antigen Test

The CLINITEST Rapid COVID-19 Antigen Test is a CE-marked Ag-RDT, manufactured by Healgen Scientific Limited Liability Company, Houston, TX, USA, and distributed by Siemens Healthineers, Erlangen, Germany. According to the instructions for use, a sample should be taken by placing a nasal swab 2-4 cm from the nostril edge in the nose and twisting it 5 times, once in each nostril. Afterwards the nose swab had to be placed in a test tube that was filled with buffer fluid and twisted for 5 times, while touching the bottom and sidewalls of the test tube, after which the swab had to be left in the test tube for 1 minute. After squeezing the tube several times, the swab could be removed from the tube, while leaving all the test fluid in the tube. Then, after placing a cap on the tube, 4 droplets of the solvent had to be added to the testing cassette. After 15 minutes, the participant had to read the test result visually from the testing cassette.

References

1. Archive EV. Wuhan coronavirus 2019 E gene control. Accessed June 8 2021, 2021. https://www.european-virus-archive.com/nucleic-acid/wuhan-coronavirus-2019-e-gene-control
2. Corman VM, Landt O, Kaiser M, et al. Detection of 2019 novel coronavirus (2019-nCoV) by real-time RT-PCR. *Euro Surveill*. Jan 2020;25
3. Schuit E, Veldhuijzen IK, Venekamp RP, et al. Diagnostic accuracy of rapid antigen tests in asymptomatic and presymptomatic close contacts of individuals with confirmed SARS-CoV-2 infection: cross sectional study. *BMJ*. 2021;374:n1676.

Supplementary material 4: Discussion on subgroup effects based on gender and age.

We found differences in sensitivity between men and women (Flowflex and Clinitest with nasal sampling) and those aged 16-40 and >40 (MPBio with nasal sampling). To further assess these differences cross-tabulation was performed of gender and age groups with other subgroup characteristics for which we found distinct differences in sensitivity: confirmatory testing as reason for testing (yes [higher sensitivity] vs. no), vaccination status (no [higher sensitivity] vs. one or more vaccinations) and having had a previous SARS-CoV-2 infection (yes vs. no [higher sensitivity]). Cross-tabulation showed that women in the Flowflex study group less often visited the test site for confirmatory testing (42% vs. 49%) and more often had had a previous SARS-CoV-2 infection (28% vs. 18%) than men, both of which may explain the lower overall sensitivity in women vs. men with Flowflex. In the Clinitest study group, differences were smaller between women and men for these subgroup characteristics but were consistently favouring men, which may contribute to the lower sensitivity seen in women. In the MPBio study group, participants aged 40 years and older were less often confirmatory testers (25.1% vs. 32.5%), and less often not vaccinated (3.9% vs. 11.7%) than participants aged 16 to 40 years old, both of which may have contributed to the lower sensitivity in the older participants. Apparently, underrepresentation of confirmatory testers and non-vaccinators in older participants was not counterbalanced by the fact that older participants less often had had a SARS-CoV-2 infection (19.3% vs. 25.1%) than younger participants. To conclude, we do not expect the diagnostic accuracy of the three Ag-RDTs truly differs according to gender or age.

Figure S1 Flow of study participants in the Delta-Omicron transition period

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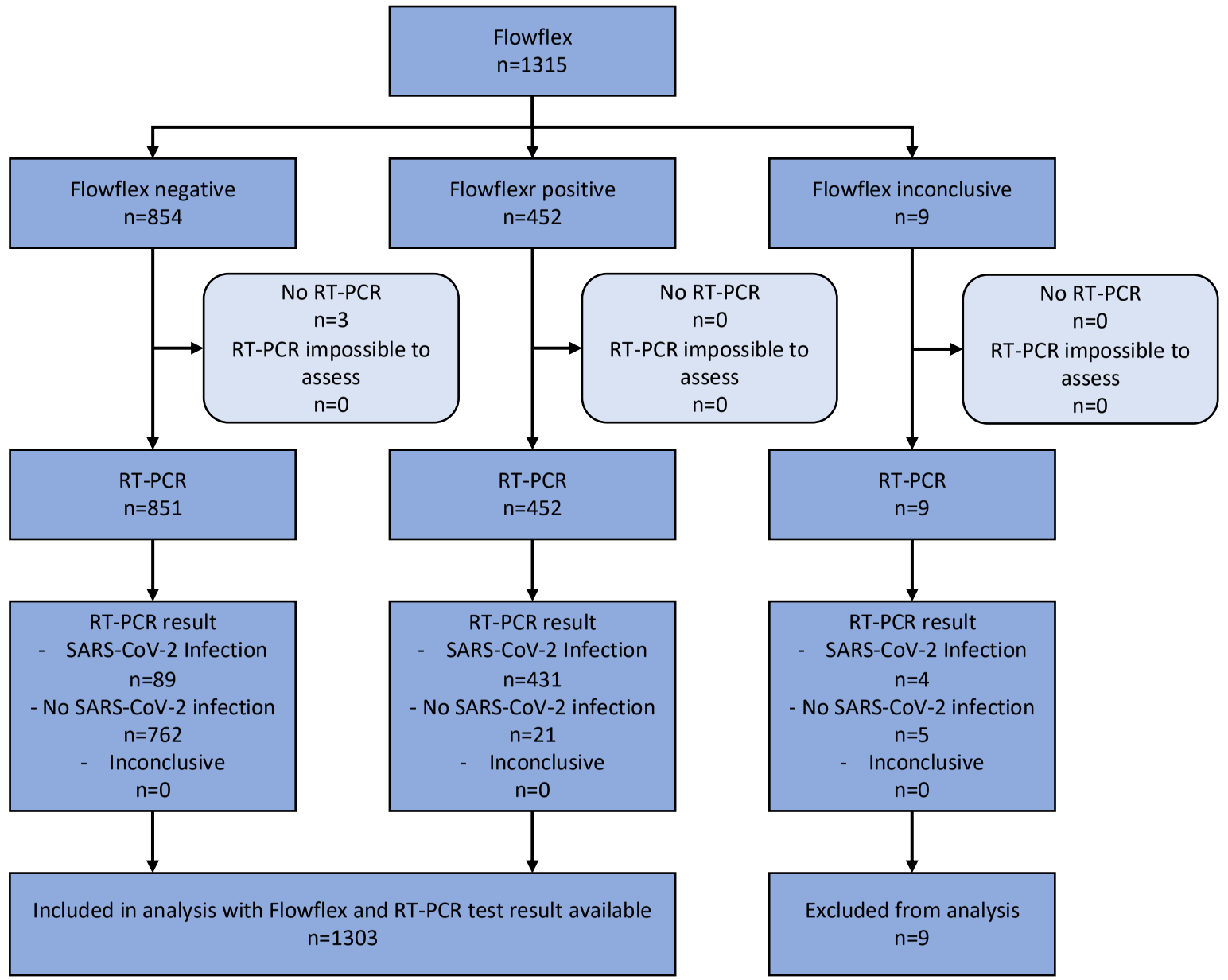
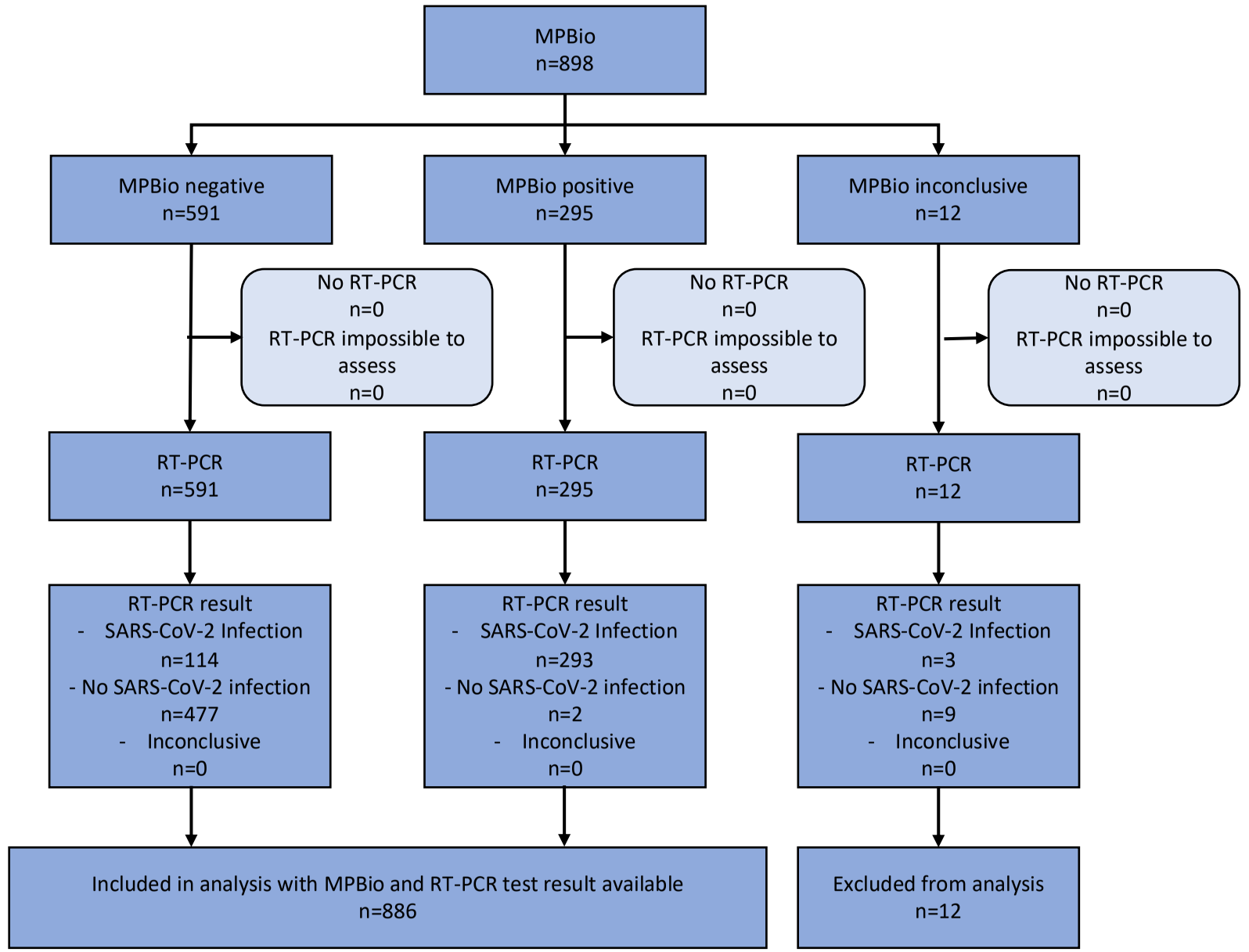
Flowflex (*Rotterdam*)



MPBio (*Tilburg*)



Clinitest (*Roosendaal*)

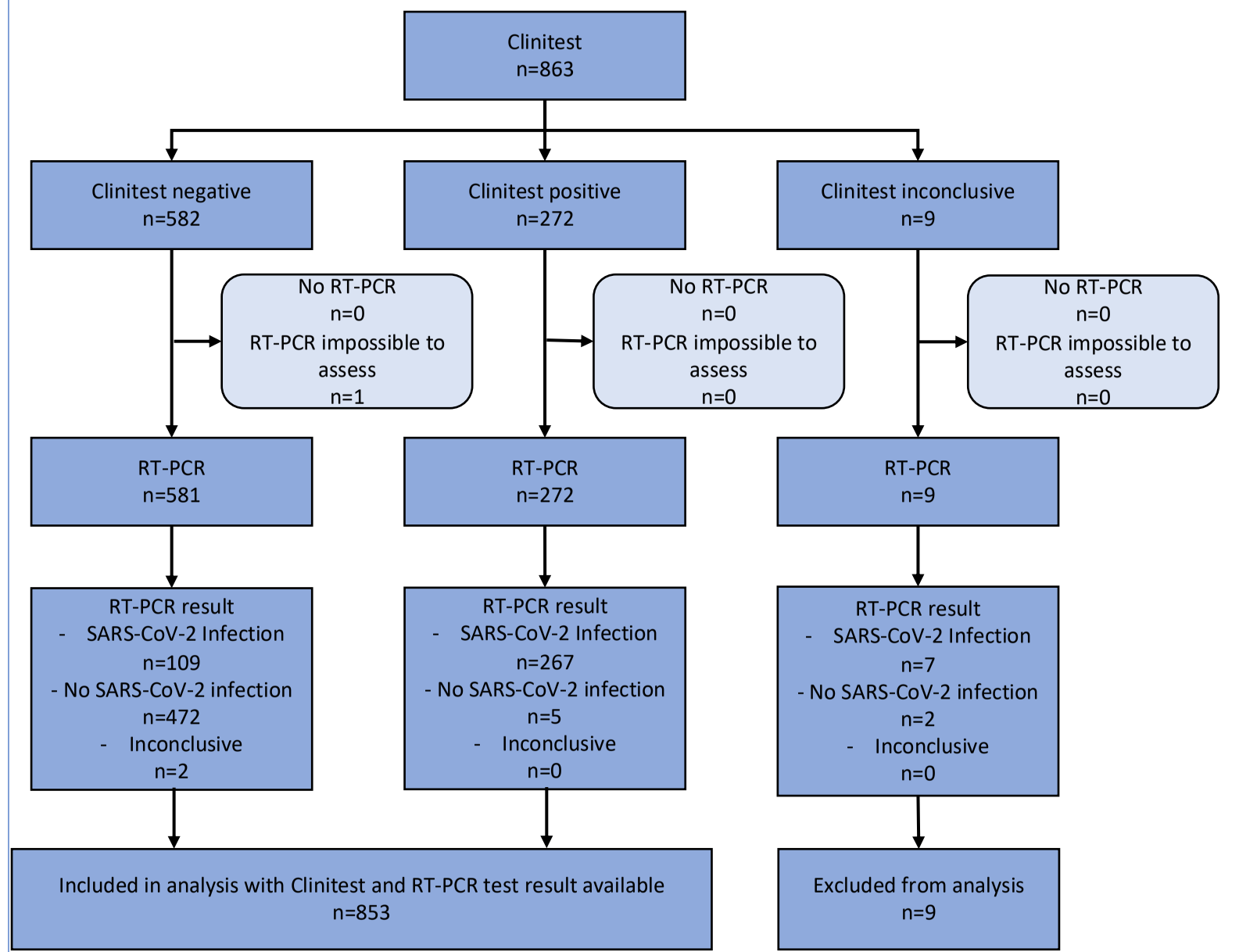


Figure S2 Sensitivities with 95% confidence intervals of the antigen rapid test-RT-PCR reference standard test comparisons after applying a viral load cut-off as a proxy for infectiousness, and stratified according to COVID-19 vaccination status, previous infection status, sex, and age, with nasal sampling for all three tests (top) and oropharyngeal plus nasal sampling for MPBio and Clinitest (bottom). The vertical line indicates the sensitivity of the Ag-RDT in the respective overall study population before application of a viral load cut-off, and the number of positive RT-PCR tests out of the total or subgroup between parentheses.

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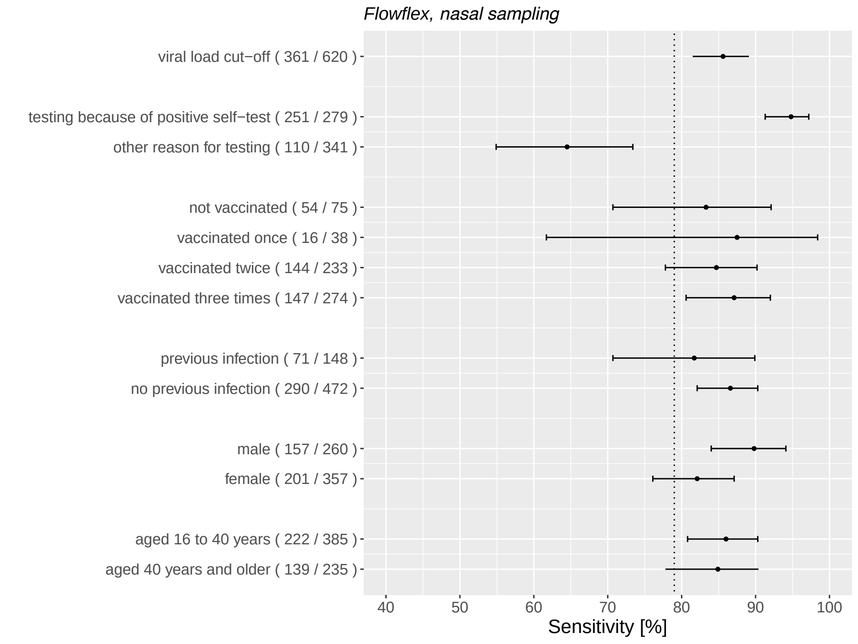
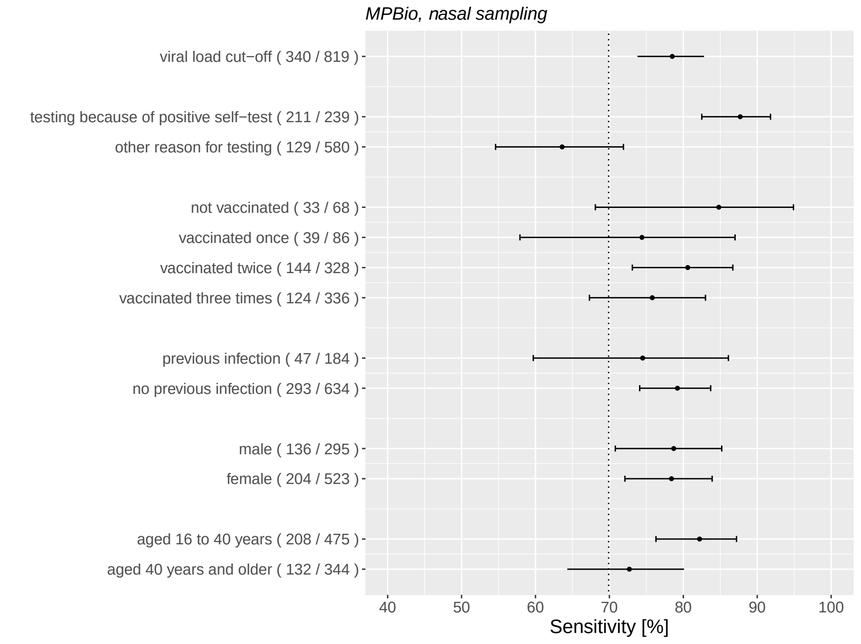
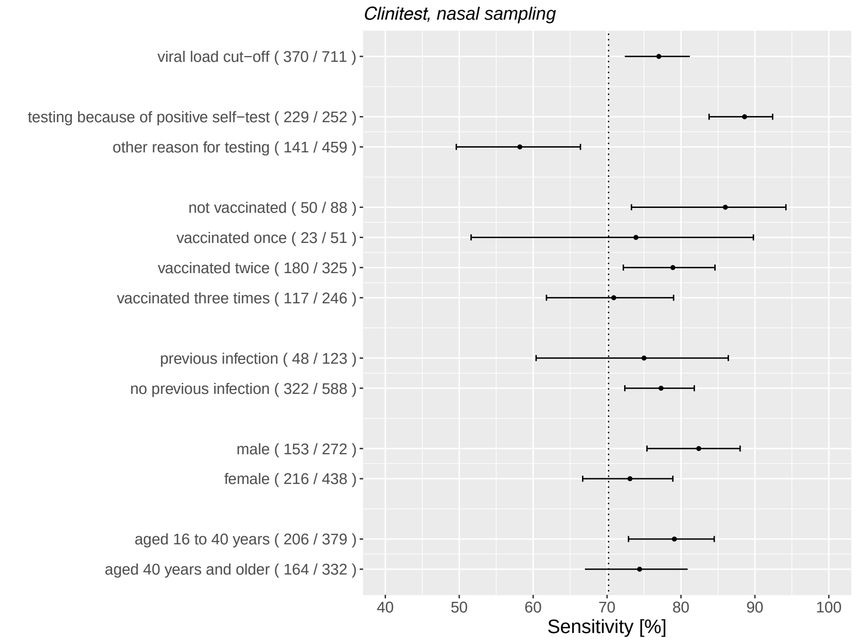
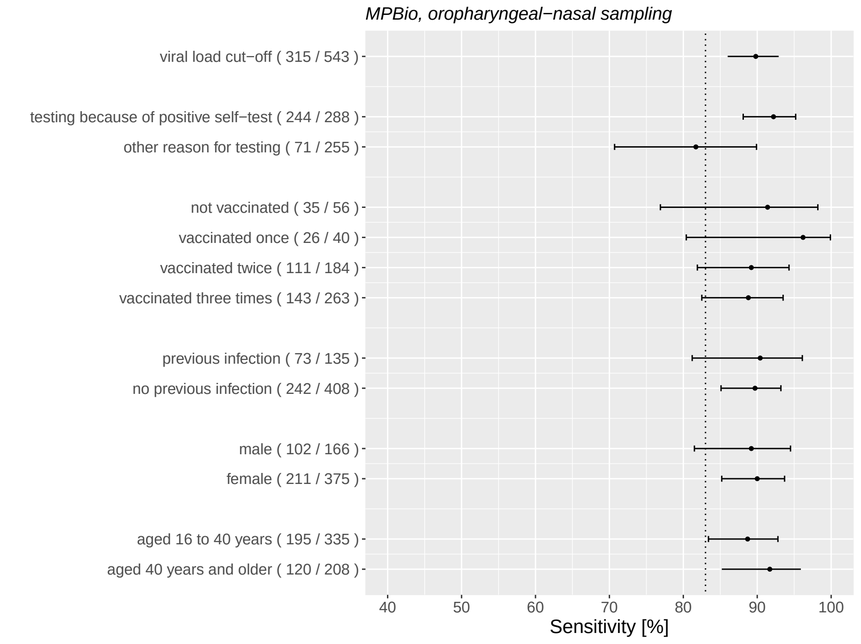

Flowflex, nasal sampling
viral load cut−off (361 / 620)
testing because of positive self−test (251 / 279)
other reason for testing (110 / 341)
not vaccinated (54 / 75)
vaccinated once (16 / 38)
vaccinated twice (144 / 233)
vaccinated three times (147 / 274)
previous infection (71 / 148)
no previous infection (290 / 472)
male (157 / 260)
female (201 / 357)
aged 16 to 40 years (222 / 385)
aged 40 years and older (139 / 235)
40 50 60 70 80 90 100
Sensitivity [%]


MPBio, nasal sampling
viral load cut−off (340 / 819)
testing because of positive self−test (211 / 239)
other reason for testing (129 / 580)
not vaccinated (33 / 68)
vaccinated once (39 / 86)
vaccinated twice (144 / 328)
vaccinated three times (124 / 336)
previous infection (47 / 184)
no previous infection (293 / 634)
male (136 / 295)
female (204 / 523)
aged 16 to 40 years (208 / 475)
aged 40 years and older (132 / 344)
40 50 60 70 80 90 100
Sensitivity [%]


Clinitest, nasal sampling
viral load cut−off (370 / 711)
testing because of positive self−test (229 / 252)
other reason for testing (141 / 459)
not vaccinated (50 / 88)
vaccinated once (23 / 51)
vaccinated twice (180 / 325)
vaccinated three times (117 / 246)
previous infection (48 / 123)
no previous infection (322 / 588)
male (153 / 272)
female (216 / 438)
aged 16 to 40 years (206 / 379)
aged 40 years and older (164 / 332)
40 50 60 70 80 90 100
Sensitivity [%]


MPBio, oropharyngeal−nasal sampling
viral load cut−off (315 / 543)
testing because of positive self−test (244 / 288)
other reason for testing (71 / 255)
not vaccinated (35 / 56)
vaccinated once (26 / 40)
vaccinated twice (111 / 184)
vaccinated three times (143 / 263)
previous infection (73 / 135)
no previous infection (242 / 408)
male (102 / 166)
female (211 / 375)
aged 16 to 40 years (195 / 335)
aged 40 years and older (120 / 208)
40 50 60 70 80 90 100
Sensitivity [%]

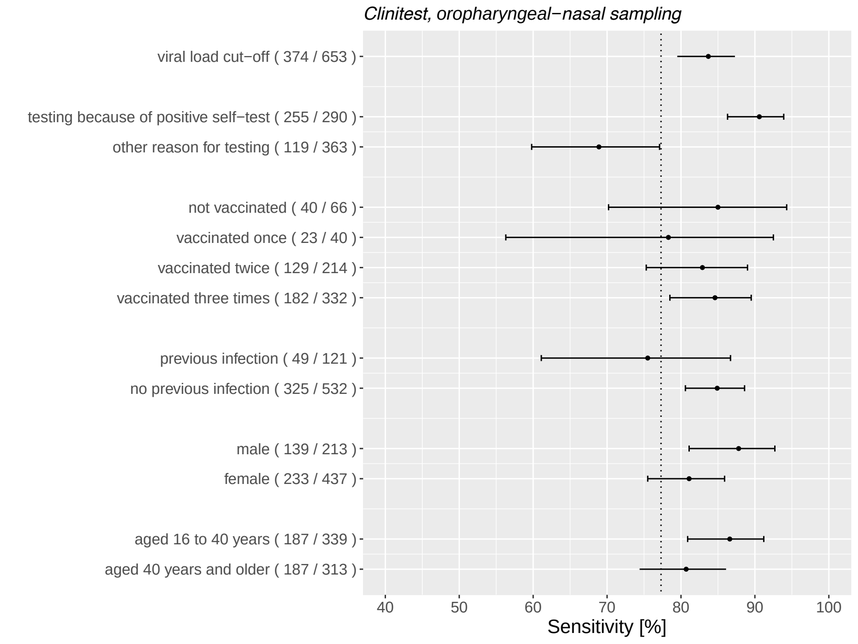
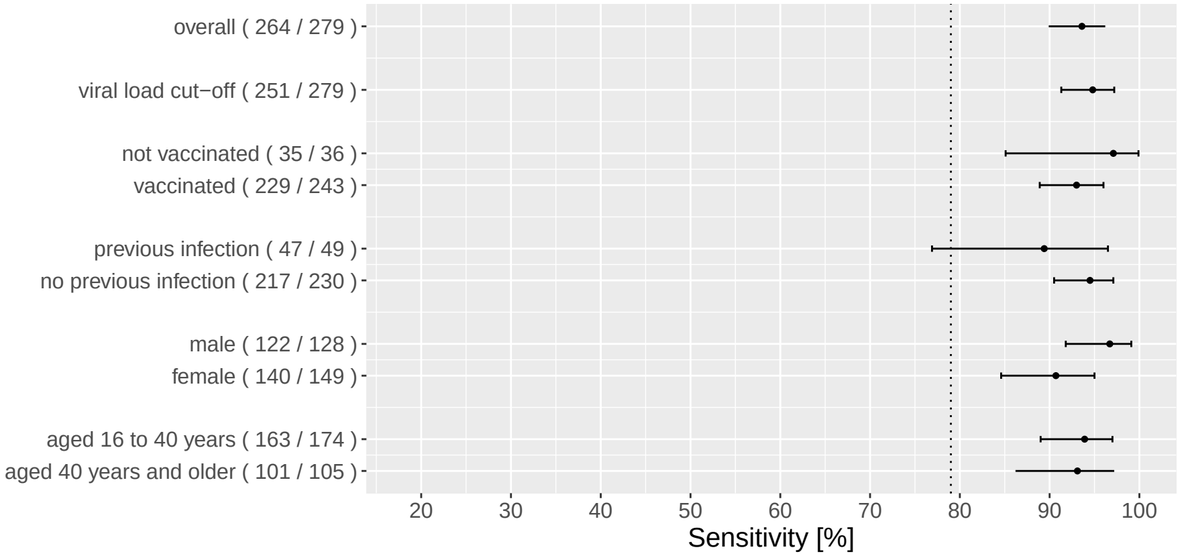

Clinitest, oropharyngeal−nasal sampling
viral load cut−off (374 / 653)
testing because of positive self−test (255 / 290)
other reason for testing (119 / 363)
not vaccinated (40 / 66)
vaccinated once (23 / 40)
vaccinated twice (129 / 214)
vaccinated three times (182 / 332)
previous infection (49 / 121)
no previous infection (325 / 532)
male (139 / 213)
female (233 / 437)
aged 16 to 40 years (187 / 339)
aged 40 years and older (187 / 313)
40 50 60 70 80 90 100
Sensitivity [%]

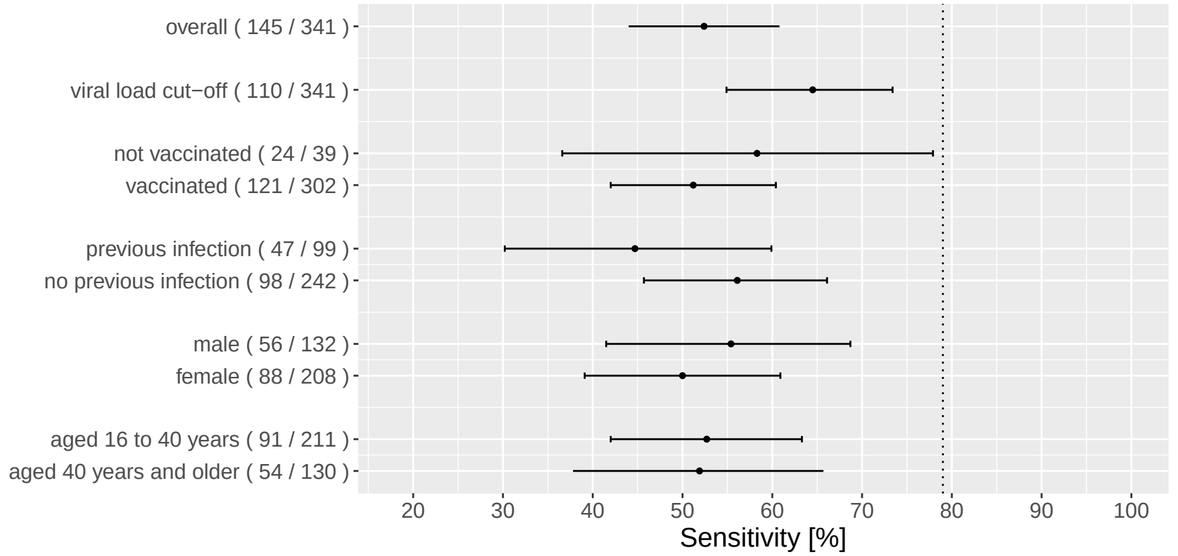
Figure S3 Sensitivities with 95% confidence intervals of the antigen rapid test-RT-PCR reference standard test comparisons stratified according to COVID-19 vaccination status, previous infection status, sex, and age, with nasal sampling, stratified by confirmatory testing (left) vs. other reason for testing (right). The vertical line indicates the sensitivity of the Ag-RDT in the respective overall study population, and the number of positive RT-PCR tests out of the total or subgroup between parentheses.

See next page

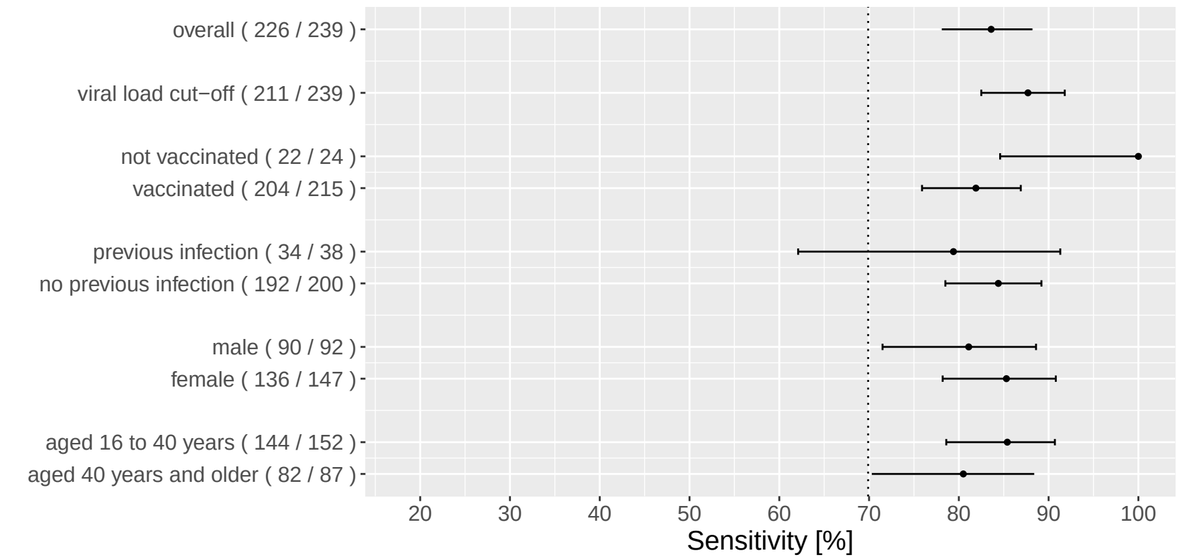
Flowflex, testing because of positive self-test



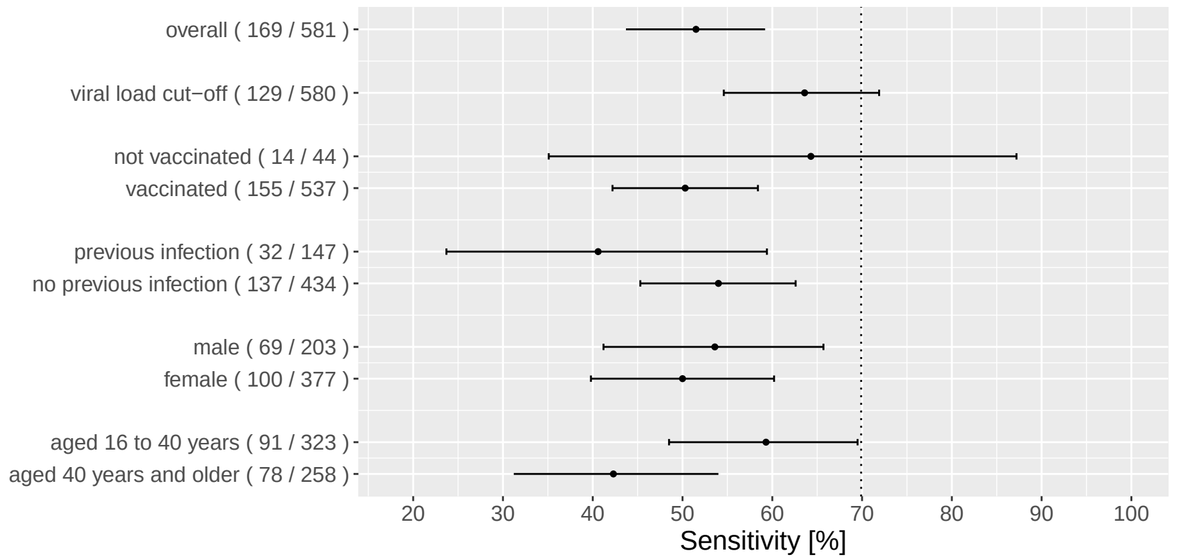
Flowflex, testing for other reasons



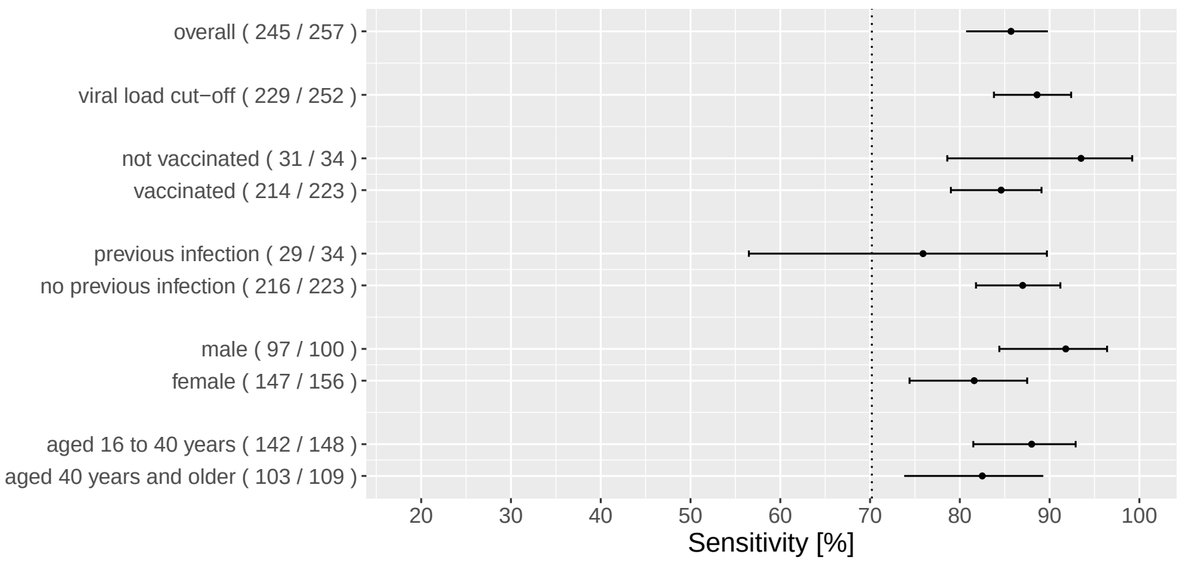
MPBio, testing because of positive self-test



MPBio, testing for other reasons



Clinitest, testing because of positive self-test



Clinitest, testing for other reasons

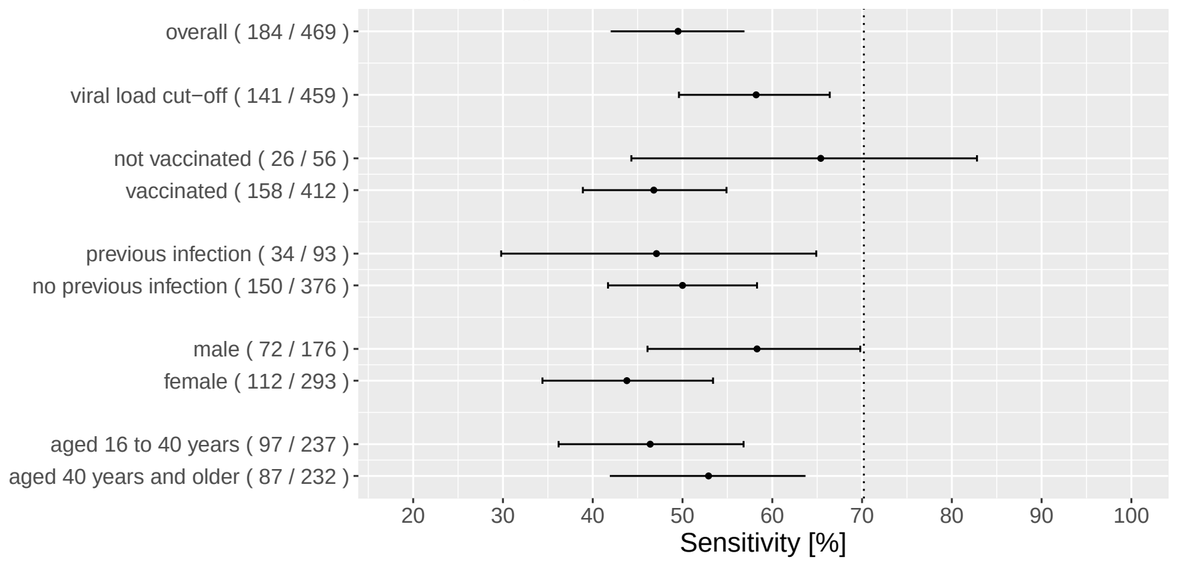
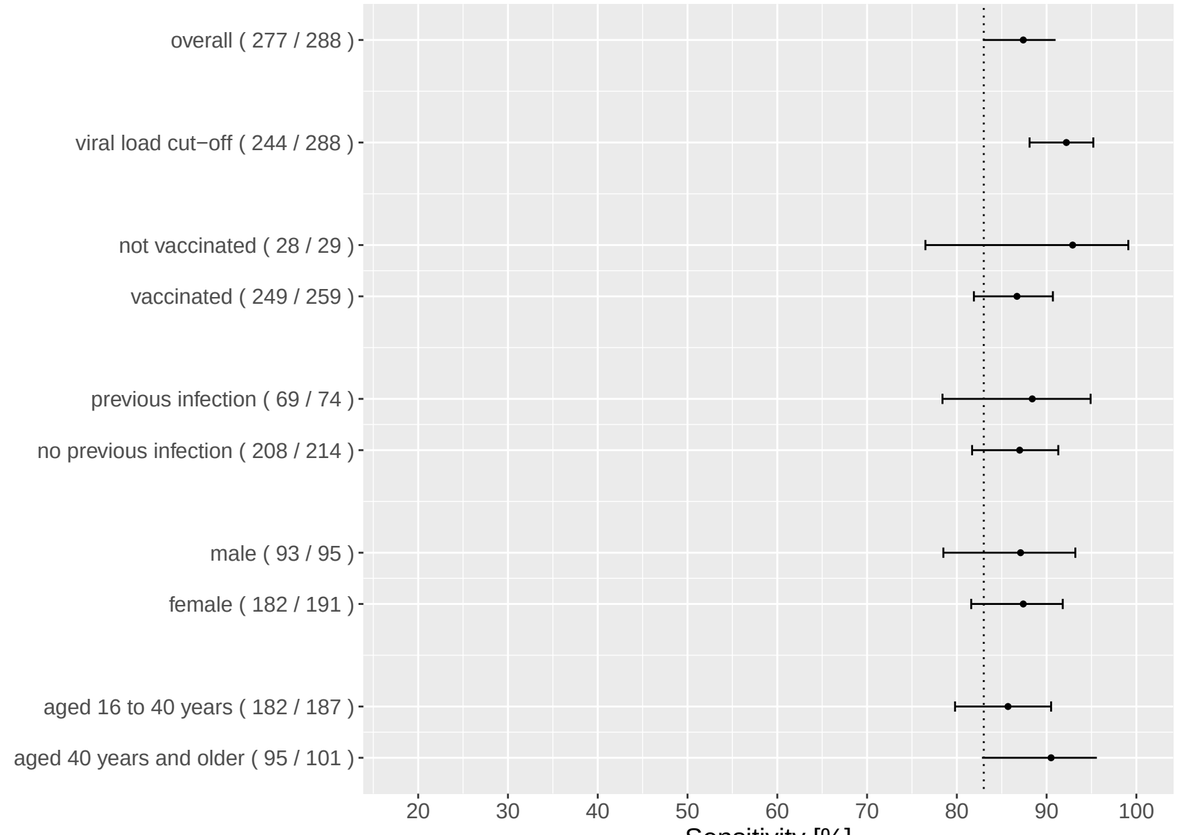


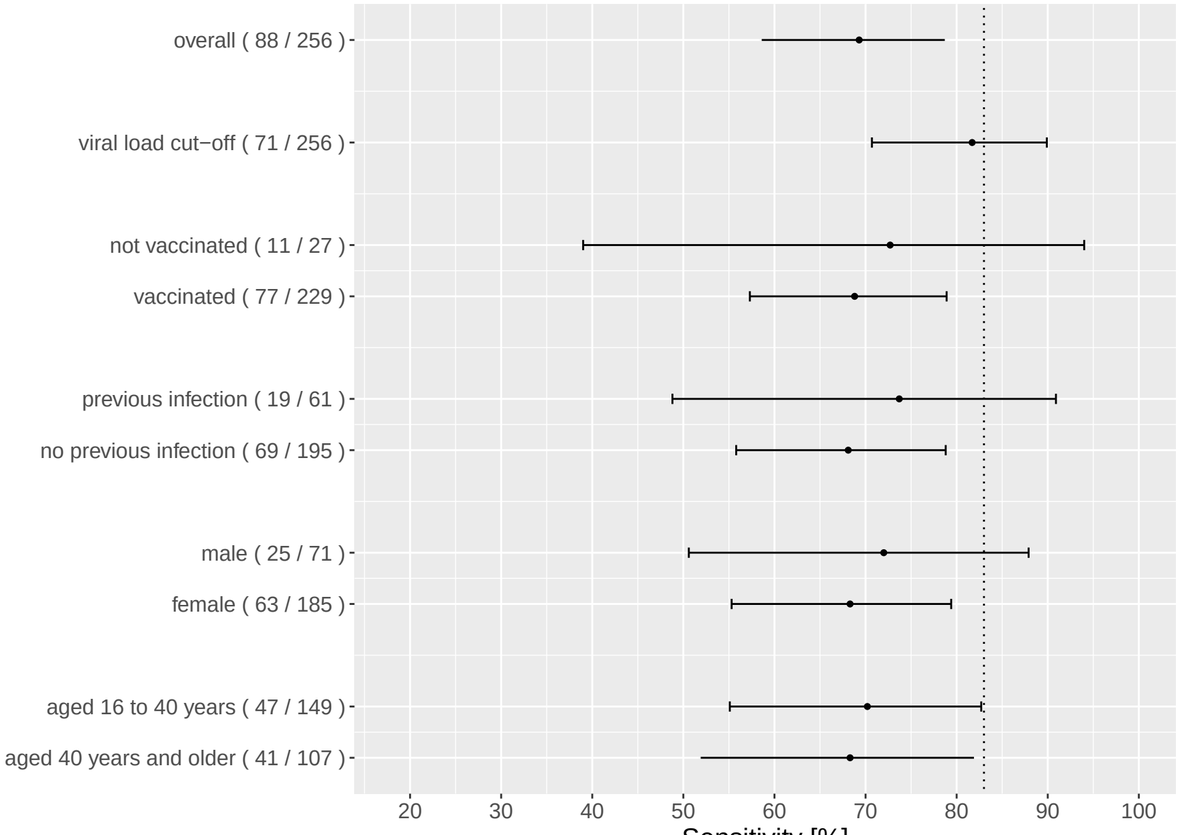
Figure S4 Sensitivities with 95% confidence intervals of the antigen rapid test-RT-PCR reference standard test comparisons stratified according to COVID-19 vaccination status, previous infection status, sex, and age, with OP-N sampling, stratified by confirmatory testing (left) vs. other reason for testing (right). The vertical line indicates the sensitivity of the Ag-RDT in the respective overall study population, and the number of positive RT-PCR tests out of the total or subgroup between parentheses.

See next page

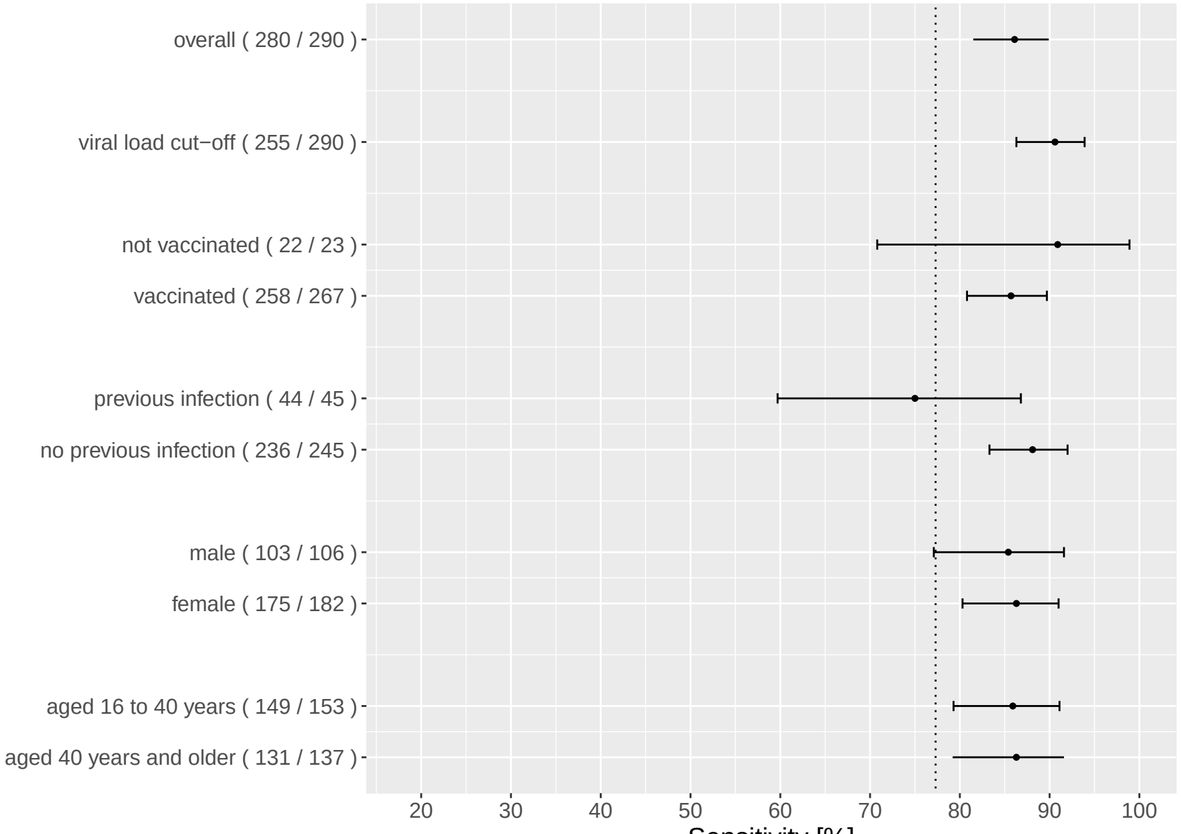
MPBio, testing because of positive self-test



MPBio, testing for other reasons



Clinitest, testing because of positive self-test



Clinitest, testing for other reasons

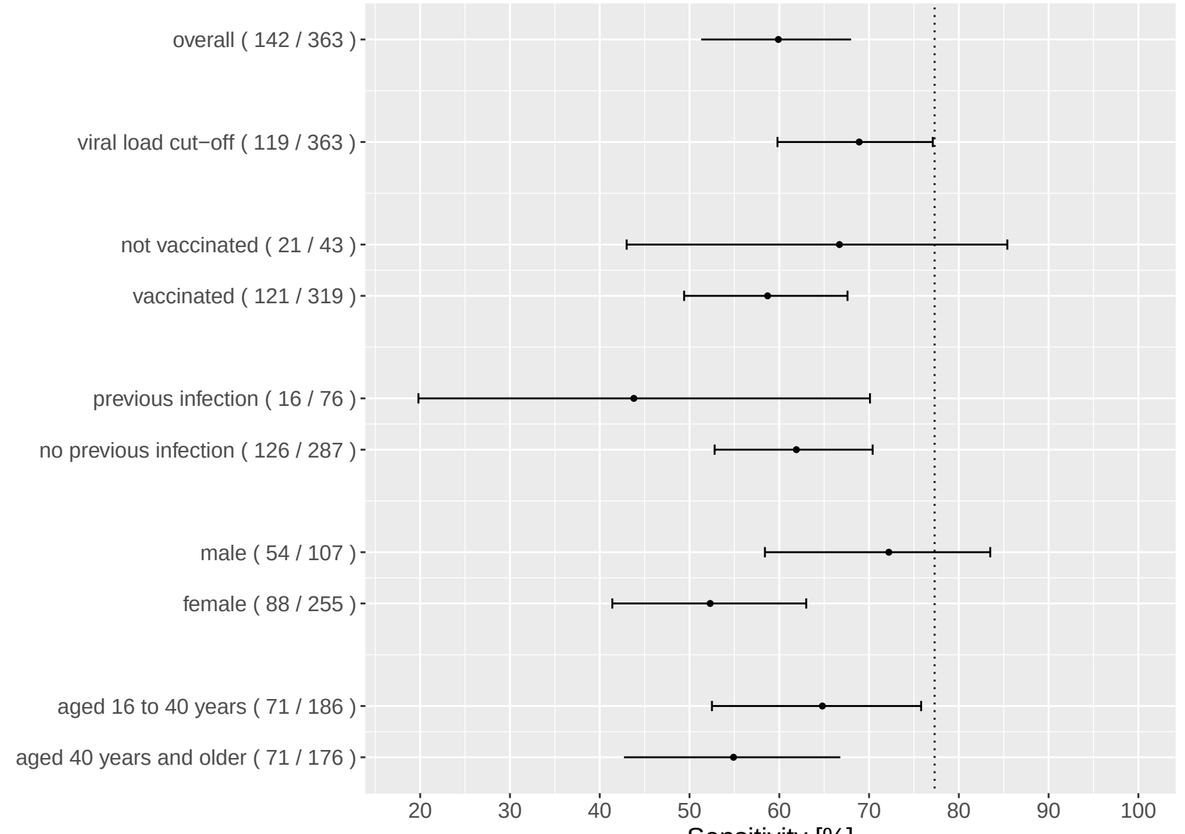
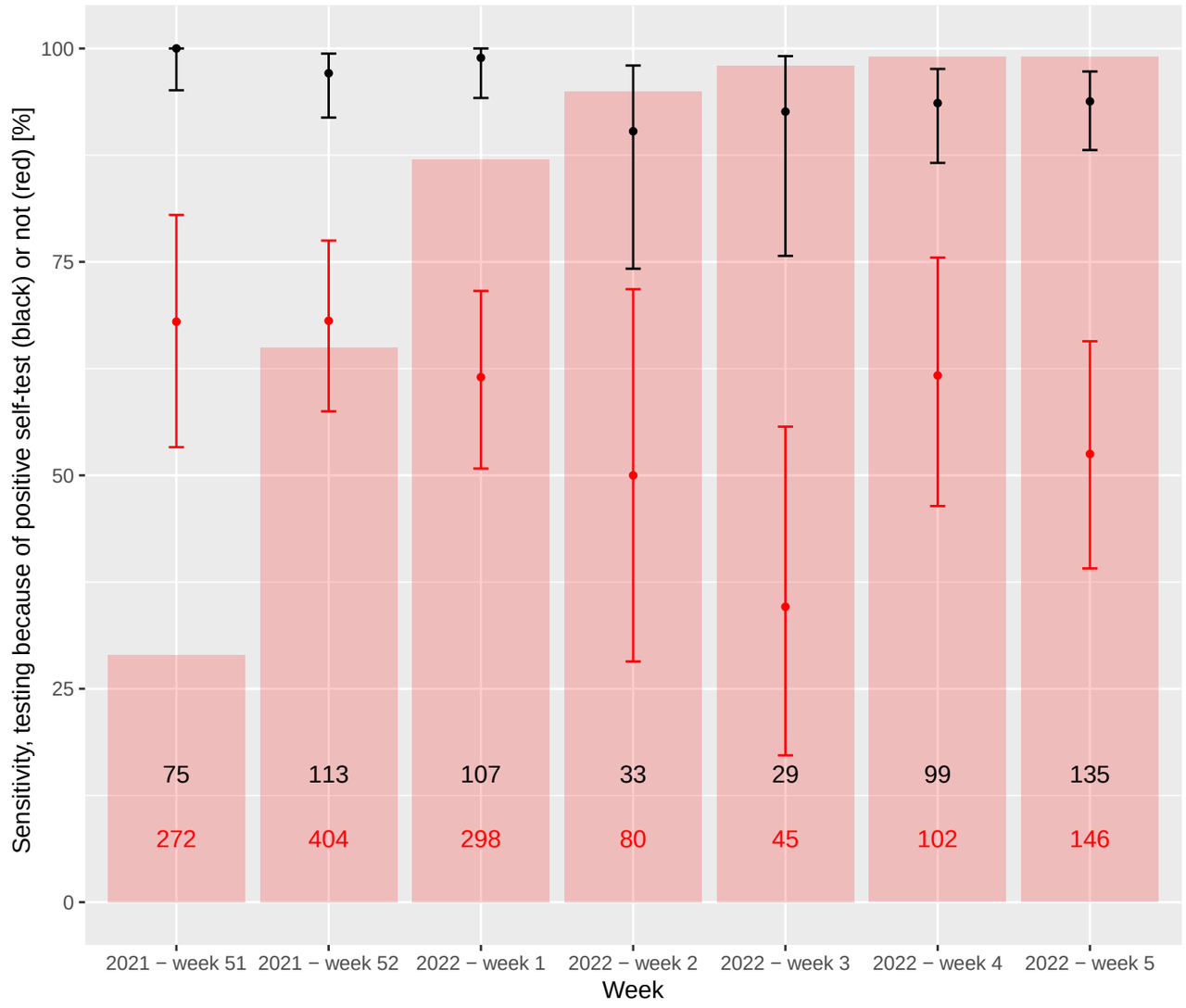


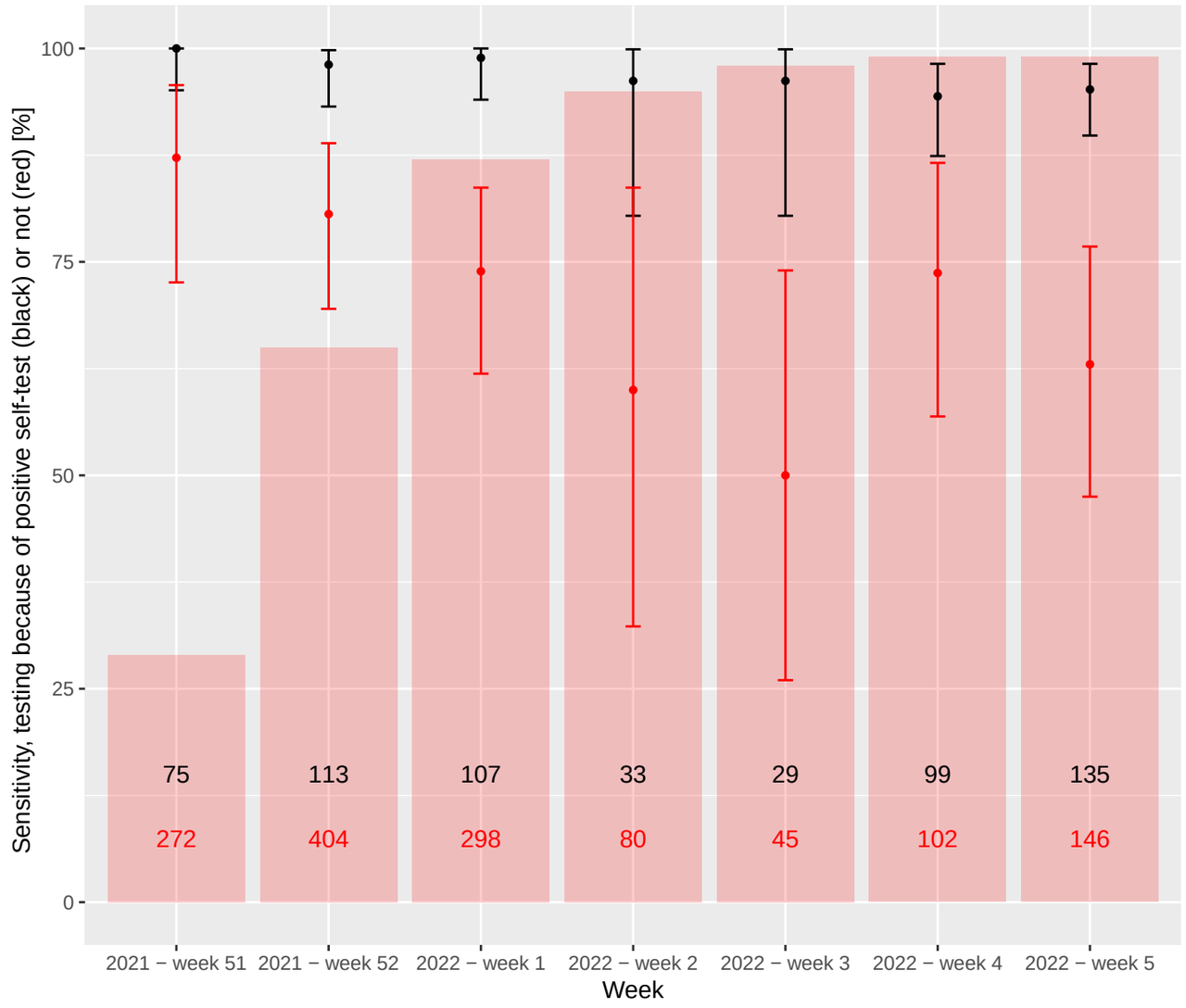
Figure S5 Sensitivities with 95% confidence intervals of the antigen rapid test with nasal sampling-RT-PCR reference standard test comparison by week of inclusion stratified by confirmatory testing (black) or other reason for testing (red), before (left) and after application of a viral load cut-off (right). The histogram indicates the percentage of the Omicron variant according to the national pathogen surveillance, while the numbers indicate the number of participants included in each week, stratified by reason for testing.

See next page

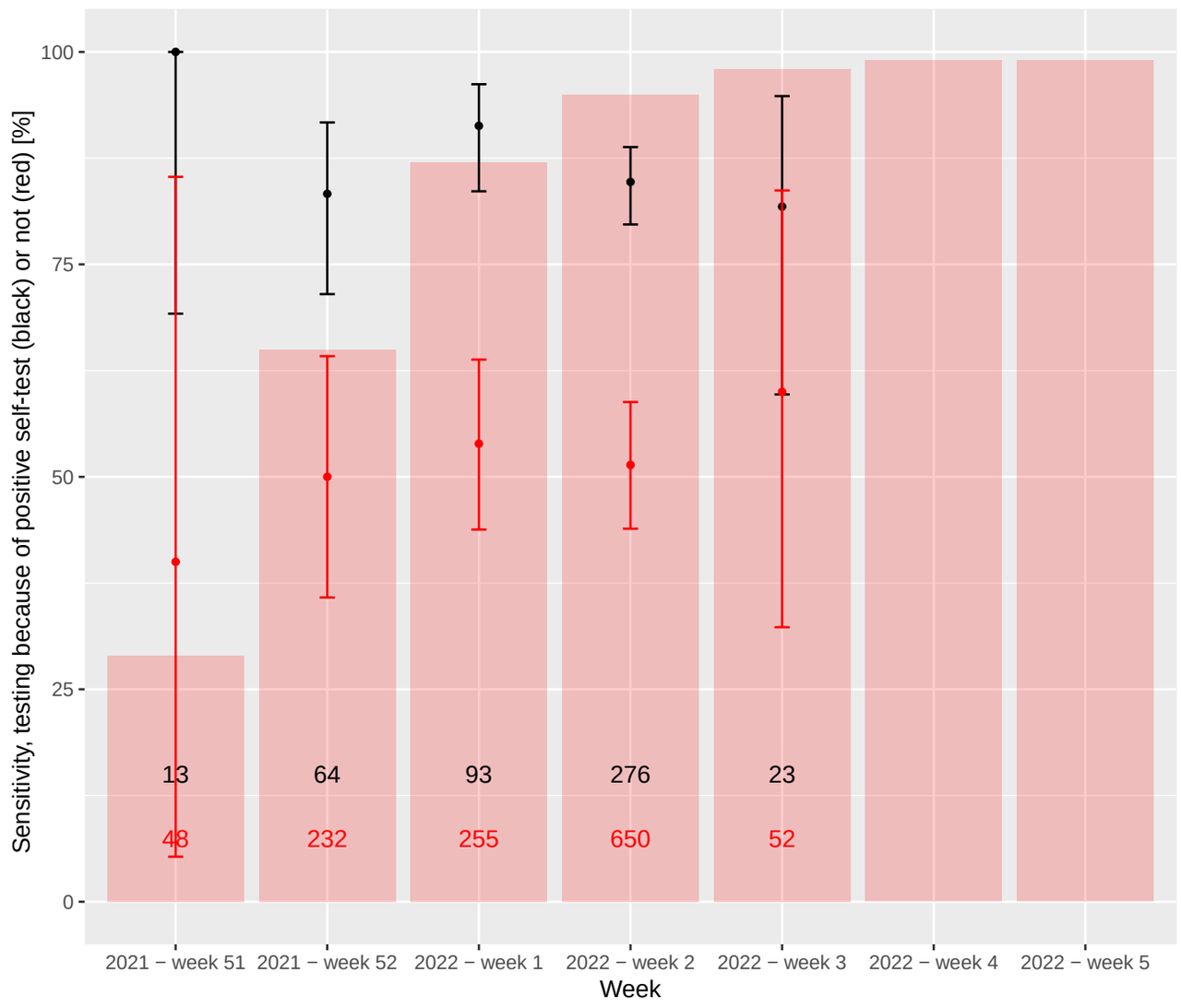
Flowflex



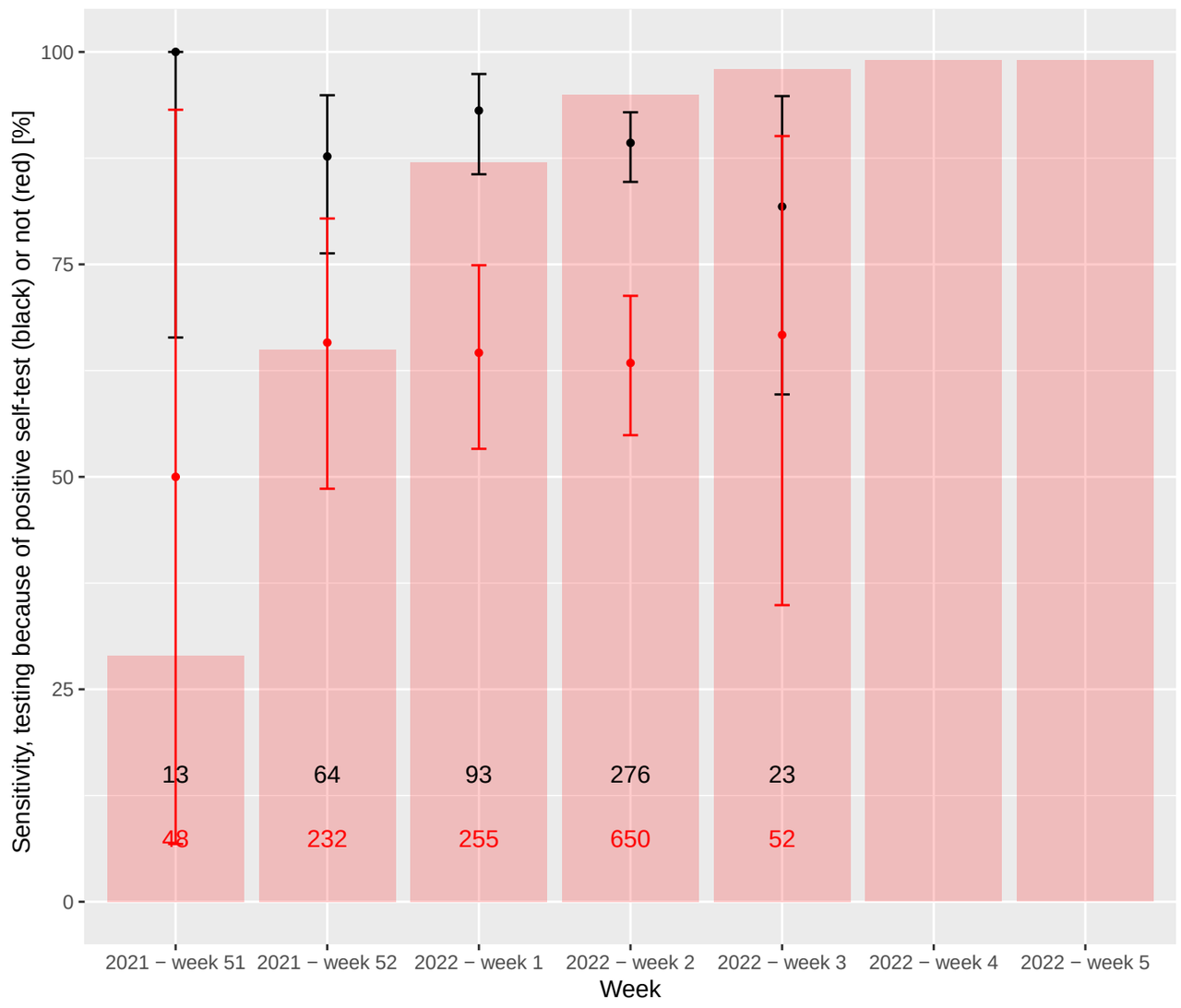
Flowflex at viral load cut−off



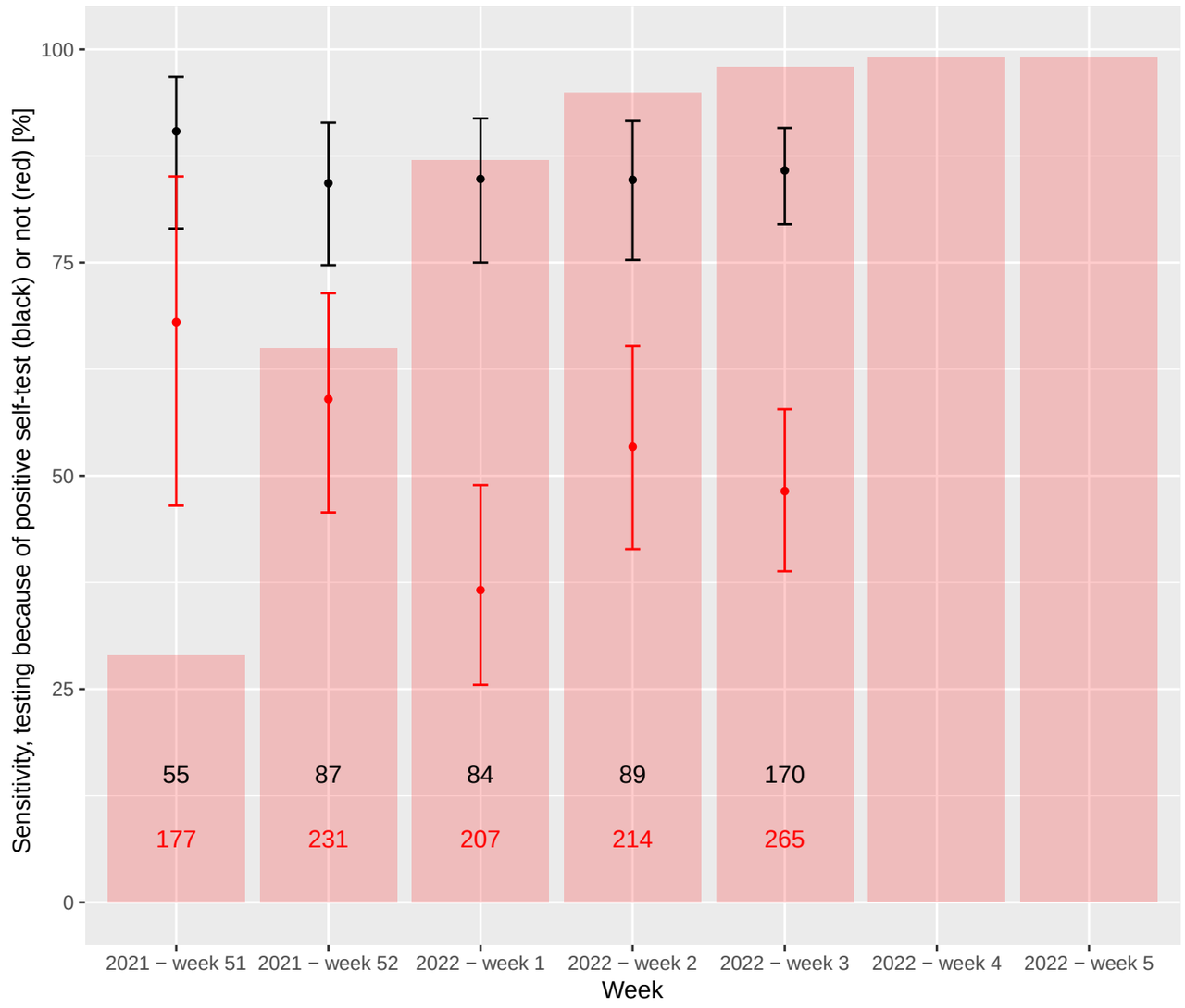
MPBio



MPBio at viral load cut−off



Clinitest



Clinitest at viral load cut−off

Table S1 Baseline characteristics of the study population during the Delta-Omicron transition period, stratified by rapid antigen test

Test result available from the reference test and…	Flowflex	MPBio	Clinitest
Location	Rotterdam	Tilburg	Roosendaal
Inclusion period	Dec 22 – Jan 11	Dec 24 – Jan 11	Dec 21 – Jan 11
Sample size	N = 1303	N = 866	N = 853
Age [years], mean (SD)	37 (13)	38 (15)	41 (15)
Range (min-max)	16-80	16-79	16-79
Sex, female, No. (%)	718 (55.1)	567 (64.0)	524 (61.4)
Reason for testing, No. (%)[4]			
Positive self-test	304 (23.3)	230 (26.0)	228 (26.7)
Symptoms	930 (71.4)	592 (66.8)	590 (69.2)
Close contact[1]	183 (14.0)	164 (18.5)	130 (15.2)
Other reason	106 (8.1)	73 (8.2)	68 (8.0)
Vaccination status			
Not vaccinated	109 (8.4)	61 (6.9)	81 (9.5)
Vaccinated with at least one dose, No. (%)	1194 (91.6)	825 (93.1)	772 (90.5)
Number of vaccinations received, No. (%)[2]			
1	158 (13.2)	106 (12.8)	109 (14.1)
2	858 (71.9)	533 (64.6)	546 (70.7)
3	177 (14.8)	186 (22.5)	117 (15.2)
4	1 (0.1)	0 (0)	0 (0)
Unknown	0 (0)	0 (0)	0 (0)
Type of initial vaccine, No. (%)[2]			
Pfizer	924 (77.4)	539 (65.3)	483 (62.6)
Moderna	97 (8.1)	101 (12.2)	121 (15.7)
Astra Zeneca	76 (6.4)	104 (12.6)	101 (13.1)
Janssen	91 (7.6)	79 (9.6)	63 (8.2)
Unknown/Other	6 (0.5)	2 (0.2)	4 (0.5)
Type of booster vaccine, No. (%)[2]			
Pfizer	131 (11.0)	116 (14.1)	82 (10.6)
Moderna	64 (5.4)	75 (9.1)	42 (5.4)
No booster received	993 (83.2)	621 (75.3)	641 (83.0)
Unknown	6 (0.5)	13 (1.6)	7 (0.9)
At least one prior SARS-CoV-2 infection, No. (%)	317 (24.4)	161 (18.2)	156 (18.3)
Less than 2 months ago	34 (10.7)	16 (9.9)	15 (9.6)

2 to 6 months ago	56 (17.7)	20 (12.4)	15 (9.6)
6 to 12 months ago	113 (35.6)	69 (42.9)	52 (33.3)
More than 12 months ago	114 (36.0)	56 (34.8)	73 (46.8)
Unknown	0 (0)	0 (0)	1 (0.6)
Symptom onset, No. (%)[3]			
On day of sampling	148 (11.4)	69 (7.8)	58 (6.8)
A day before sampling	492 (37.8)	285 (32.2)	313 (36.7)
Two days before sampling	341 (26.2)	289 (32.6)	249 (29.2)
Three or more days before sampling	321 (24.6)	243 (27.4)	232 (27.2)
Unknown	1 (0.1)	0 (0)	1 (0.1)
Type of symptoms (self-reported), No. (%)[3,4]			
Common cold	1113 (85.4)	771 (87.0)	730 (85.6)
Shortness of breath	172 (13.2)	148 (16.7)	145 (17.0)
Fever	257 (19.7)	194 (21.9)	177 (20.8)
Coughing	592 (45.4)	430 (48.5)	440 (51.6)
Loss of taste or smell	60 (4.6)	40 (4.5)	44 (5.2)
Muscle ache	210 (16.1)	181 (20.4)	168 (19.7)
Other symptoms	130 (10.0)	117 (13.2)	98 (11.5)
Previous experience with using self-tests, No. (%)	1236 (94.9)	851 (96.0)	795 (93.5)
Performed last self-test, No. (%)[2]			
Less than 7 days ago	1011 (81.8)	723 (85.0)	644 (81.0)
1 to 4 weeks ago	144 (11.7)	77 (9.0)	96 (12.1)
More than 1 months ago	77 (6.2)	50 (5.9)	55 (6.9)
Unknown	4 (0.3)	1 (0.1)	0 (0)
Number of performed self-tests, No. (%)			
1-3	299 (24.2)	211 (24.8)	251 (31.6)
4-6	362 (29.3)	254 (29.9)	248 (31.2)
7-10	294 (23.8)	189 (22.2)	168 (21.1)
>10	279 (22.6)	196 (23.1)	128 (12.6)

SD=standard deviation.

[1] In the Netherlands, individuals are notified of a close contact by the Dutch public health service test-and-trace program, and/or the Dutch contact tracing mobile phone application (the CoronaMelder app) and/or an individual with a confirmed SARS-CoV-2 infection (index case).

[2] percentage calculated as proportion of those vaccinated, or those that had previous experience in performing a self-test

[3] Percentage calculated as proportion of those with symptoms at time of sampling

[4] Totals add up to a number higher than the number of participants, and individuals with symptoms at the time of sampling because individuals could report more than one reason for testing, or symptom, respectively.

Table S2 2x2 tables for primary and secondary analyses of the RT-PCR reference test – Acon Labs Flowflex COVID-19 Antigen Home Test comparison – nasal sampling

Analysis	Subgroup				
Primary			Ref +	Ref -	Total
		Test +	323	6	329
		Test -	86	205	291
		Total	409	211	620
Secondary (stratified):					
Infectiousness viral load cut-off¶			Ref +	Ref -	Total
		Test +	309	20	329
		Test -	52	239	291
		Total	361	259	620
Vaccinated (at least at least once):	Yes		Ref +	Ref -	Total
		Test +	275	6	281
		Test -	75	189	264
		Total	350	195	545
	No		Ref +	Ref -	Total
		Test +	48	0	48
		Test -	11	16	27
		Total	59	16	75
Previous SARS-CoV-2 infection:	Yes		Ref +	Ref -	Total
		Test +	63	1	64
		Test -	31	53	84
		Total	94	54	148
	No		Ref +	Ref -	Total
		Test +	260	5	265
		Test -	55	152	207
		Total	315	157	472
Sex:	Female		Ref +	Ref -	Total
		Test +	171	2	173
		Test -	57	127	184
		Total	228	129	357
	Male		Ref +	Ref -	Total
		Test +	149	4	153
		Test -	29	78	107
		Total	178	82	260
Age [years]	≥16 to ≤40		Ref +	Ref -	Total
		Test +	201	5	206
		Test -	53	126	179
		Total	254	131	385
	>40		Ref +	Ref -	Total
		Test +	122	1	123
		Test -	33	79	112
		Total	155	80	235
Reasons for testing was positive self-test:	Yes		Ref +	Ref -	Total
		Test +	247	3	250
		Test -	17	12	29
		Total	264	15	279
	No		Ref +	Ref -	Total
		Test +	76	3	79
		Test -	69	193	262
		Total	145	196	341

Ref = RT-PCR reference standard test; Test = Flowflex

¶Viral load cut-off for infectiousness, defined as viral load above which 95% of people with a positive RT-PCR test result had a positive viral culture[1] was 5.2 log10 SARS-CoV-2 E-gene copies/mL

Table S3 2x2 tables for primary and secondary analyses of the RT-PCR reference test – MP Biomedicals Rapid SARS-CoV-2 Antigen Test Card comparison – nasal sampling

Analysis	Subgroup				
Primary			Ref +	Ref -	Total
		Test +	276	5	281
		Test -	119	420	539
		Total	395	425	820
Secondary (stratified):					
Infectiousness viral load cut-off¶			Ref +	Ref -	Total
		Test +	267	14	281
		Test -	73	465	538
		Total	340	479	819
Vaccinated (at least at least once):	Yes		Ref +	Ref -	Total
		Test +	245	3	248
		Test -	114	390	504
		Total	359	393	752
	No		Ref +	Ref -	Total
		Test +	31	2	33
		Test -	5	30	35
		Total	36	32	68
Previous SARS-CoV-2 infection:	Yes		Ref +	Ref -	Total
		Test +	40	0	40
		Test -	26	119	145
		Total	66	119	185
	No		Ref +	Ref -	Total
		Test +	236	5	241
		Test -	93	300	393
		Total	329	305	634
Sex:	Female		Ref +	Ref -	Total
		Test +	166	3	169
		Test -	70	285	355
		Total	236	288	524
	Male		Ref +	Ref -	Total
		Test +	110	2	112
		Test -	49	134	183
		Total	159	136	295
Age [years]	≥16 to ≤40		Ref +	Ref -	Total
		Test +	177	2	179
		Test -	58	238	296
		Total	235	240	475
	>40		Ref +	Ref -	Total
		Test +	99	3	102
		Test -	61	182	243
		Total	160	185	345
Reasons for testing was positive self-test:	Yes		Ref +	Ref -	Total
		Test +	189	2	191
		Test -	37	11	48
		Total	226	13	239
	No		Ref +	Ref -	Total
		Test +	87	3	90
		Test -	82	409	491
		Total	169	412	581

Ref = RT-PCR reference standard test; Test = MPBio

¶Viral load cut-off for infectiousness, defined as viral load above which 95% of people with a positive RT-PCR test result had a positive viral culture[1] was 5.2 log10 SARS-CoV-2 E-gene copies/mL.

Table S4 2x2 tables for primary and secondary analyses of the RT-PCR reference test – Siemens-Healthineers CLINITEST Rapid COVID-19 Antigen Test – nasal sampling

Analysis	Subgroup				
Primary			Ref +	Ref -	Total
		Test +	301	2	303
		Test -	128	295	423
		Total	429	297	726
Secondary (stratified):					
Infectiousness viral load cut-off¶			Ref +	Ref -	Total
		Test +	285	9	294
		Test -	85	332	417
		Total	370	341	711
Vaccinated (at least at least once):	Yes		Ref +	Ref -	Total
		Test +	255	0	255
		Test -	117	263	380
		Total	372	263	635
	No		Ref +	Ref -	Total
		Test +	46	2	48
		Test -	11	31	42
		Total	57	33	90
Previous SARS-CoV-2 infection:	Yes		Ref +	Ref -	Total
		Test +	38	2	40
		Test -	25	62	87
		Total	63	64	127
	No		Ref +	Ref -	Total
		Test +	263	0	263
		Test -	103	233	336
		Total	366	233	599
Sex:	Female		Ref +	Ref -	Total
		Test +	169	2	171
		Test -	90	188	278
		Total	259	190	449
	Male		Ref +	Ref -	Total
		Test +	131	0	131
		Test -	38	107	145
		Total	169	107	276
Age [years]	≥16 to ≤40		Ref +	Ref -	Total
		Test +	170	1	171
		Test -	69	145	214
		Total	239	146	385
	>40		Ref +	Ref -	Total
		Test +	131	1	132
		Test -	59	150	209
		Total	190	151	341
Reasons for testing was positive self-test:	Yes		Ref +	Ref -	Total
		Test +	210	1	211
		Test -	35	11	46
		Total	245	12	257
	No		Ref +	Ref -	Total
		Test +	91	1	92
		Test -	93	284	377
		Total	184	285	469

Ref = RT-PCR reference standard test; Test = Clinitest

¶Viral load cut-off for infectiousness, defined as viral load above which 95% of people with a positive RT-PCR test result had a positive viral culture[1] was 5.2 log10 SARS-CoV-2 E-gene copies/mL.

Table S5 2x2 tables for primary and secondary analyses of the RT-PCR reference test – MP Biomedicals Rapid SARS-CoV-2 Antigen Test Card comparison – oropharyngeal-nasal sampling

Analysis	Subgroup				
Primary			Ref +	Ref -	Total
		Test +	303	4	307
		Test -	62	175	237
		Total	365	179	544
Secondary (stratified):					
Infectiousness viral load cut-off¶			Ref +	Ref -	Total
		Test +	283	24	307
		Test -	32	205	237
		Total	315	229	544
Vaccinated (at least at least once):	Yes		Ref +	Ref -	Total
		Test +	269	4	273
		Test -	57	158	215
		Total	326	162	488
	No		Ref +	Ref -	Total
		Test +	34	0	34
		Test -	5	17	22
		Total	39	17	56
Previous SARS-CoV-2 infection:	Yes		Ref +	Ref -	Total
		Test +	75	1	76
		Test -	13	46	59
		Total	88	47	135
	No		Ref +	Ref -	Total
		Test +	228	3	231
		Test -	49	129	178
		Total	277	132	409
Sex:	Female		Ref +	Ref -	Total
		Test +	202	3	205
		Test -	43	128	171
		Total	245	131	376
	Male		Ref +	Ref -	Total
		Test +	99	1	100
		Test -	19	47	66
		Total	118	48	166
Age [years]	≥16 to ≤40		Ref +	Ref -	Total
		Test +	189	3	192
		Test -	40	104	144
		Total	229	107	336
	>40		Ref +	Ref -	Total
		Test +	114	1	115
		Test -	22	71	93
		Total	136	72	208
Reasons for testing was positive self-test:	Yes		Ref +	Ref -	Total
		Test +	242	1	243
		Test -	35	10	45
		Total	277	11	288
	No		Ref +	Ref -	Total
		Test +	61	3	64
		Test -	27	165	192
		Total	88	168	256

Ref = RT-PCR reference standard test; Test = MPBio

¶Viral load cut-off for infectiousness, defined as viral load above which 95% of people with a positive RT-PCR test result had a positive viral culture[1] was 5.2 log10 SARS-CoV-2 E-gene copies/mL.

Table S6 2x2 tables for primary and secondary analyses of the RT-PCR reference test – Siemens-Healthineers CLINITEST Rapid COVID-19 Antigen Test – oropharyngeal-nasal sampling

Analysis	Subgroup				
Primary			Ref +	Ref -	Total
		Test +	326	7	333
		Test -	96	224	320
		Total	422	231	653
Secondary (stratified):					
Infectiousness viral load cut-off¶			Ref +	Ref -	Total
		Test +	313	20	333
		Test -	61	259	320
		Total	374	279	653
Vaccinated (at least at least once):	Yes		Ref +	Ref -	Total
		Test +	292	4	296
		Test -	87	203	290
		Total	379	207	586
	No		Ref +	Ref -	Total
		Test +	34	2	36
		Test -	9	21	30
		Total	43	23	66
Previous SARS-CoV-2 infection:	Yes		Ref +	Ref -	Total
		Test +	40	3	43
		Test -	20	58	78
		Total	60	61	121
	No		Ref +	Ref -	Total
		Test +	286	4	290
		Test -	76	166	242
		Total	362	170	532
Sex:	Female		Ref +	Ref -	Total
		Test +	197	6	203
		Test -	66	168	234
		Total	263	174	437
	Male		Ref +	Ref -	Total
		Test +	127	1	128
		Test -	30	55	85
		Total	157	56	213
Age [years]	≥16 to ≤40		Ref +	Ref -	Total
		Test +	174	4	178
		Test -	46	115	161
		Total	220	119	339
	>40		Ref +	Ref -	Total
		Test +	152	3	155
		Test -	50	108	158
		Total	202	111	313
Reasons for testing was positive self-test:	Yes		Ref +	Ref -	Total
		Test +	241	2	243
		Test -	39	8	47
		Total	208	10	290
	No		Ref +	Ref -	Total
		Test +	85	5	90
		Test -	57	216	273
		Total	142	221	363

Ref = RT-PCR reference standard test; Test = Clinitest

¶Viral load cut-off for infectiousness, defined as viral load above which 95% of people with a positive RT-PCR test result had a positive viral culture[1] was 5.2 log10 SARS-CoV-2 E-gene copies/mL.

Table S7 Diagnostic accuracy parameters for the three Ag-RDTs in symptomatic individuals in the Omicron era, stratified by reasons for testing. Values are percentages (95% confidence interval) unless stated otherwise. Categories for reasons of testing are not mutually exclusive; participants could report multiple reasons for testing. If a participant indicated multiple reasons, he or she was included in the analysis of both categories.

Analysis	Sampling	No.	Prevalence* [%]	Sensitivity [%] (95%CI)	Specificity [%] (95%CI)	PPV [%] (95%CI)	NPV [%] (95%CI)
Flowflex							
Reason for testing:							
Positive self-test	N	279	94.6	93.6 (89.9-96.2)	80.0 (51.9-95.7)	98.8 (96.5-99.8)	41.4 (23.5-61.1)
Symptoms	N	405	59.8	77.7 (71.9-82.8)	96.9 (93.0-99.0)	97.4 (94.1-99.2)	74.5 (68.1-80.2)
Close contact	N	72	51.4	51.4 (34.4-68.1)	100 (90.0-100)	100 (82.4-100)	66.0 (51.7-78.5)
Other reason	N	49	69.4	61.8 (43.6-77.8)	100 (78.2-100)	100 (83.9-100)	53.6 (33.9-72.5)
MPBio							
Reason for testing:							
Positive self-test	N	239	94.6	83.6 (78.1-88.2)	84.6 (54.6-98.1)	99.0 (96.3-99.9)	22.9 (12.0-37.3)
	OP-N	288	96.2	87.4 (82.9-91.0)	90.9 (58.7-99.8)	99.6 (97.7-100)	22.2 (11.2-37.1)
Symptoms	N	510	41.2	66.7 (59.9-73.0)	98.7 (96.6-99.6)	97.2 (93.0-99.2)	80.9 (76.5-84.8)
	OP-N	355	60.8	79.6 (73.6-84.8)	87.1 (92.8-99.2)	97.7 (94.3-99.4)	75.4 (68.4-81.5)
Close contact	N	170	37.1	60.3 (47.2-72.4)	99.1 (94.9-100)	97.4 (86.5-99.9)	80.9 (73.1-87.3)
	OP-N	58	37.9	77.3 (54.6-92.2)	100 (90.3-100)	100 (80.5-100)	87.8 (73.8-95.9)
Other reason	N	54	46.3	64.0 (42.5-82.0)	100 (88.1-100)	100 (79.4-100)	76.3 (59.8-88.6)
	OP-N	15	60.0	55.6 (21.2-86.3)	100 (54.1-100)	100 (47.8-100)	60.0 (26.2-87.8)
Clinitest							
Reason for testing:							
Positive self-test	N	257	95.3	85.7 (80.7-89.8)	91.7 (61.5-99.8)	99.5 (97.4-100)	23.9 (12.6-38.8)
	OP-N	290	96.6	86.1 (81.5-89.9)	80.0 (44.4-97.5)	99.2 (97.1-99.9)	17.0 (7.6-30.8)
Symptoms	N	459	54.5	69.6 (63.5-75.2)	99.0 (96.6-99.9)	98.9 (96.0-99.9)	73.1 (67.6-78.2)
	OP-N	390	57.9	76.1 (70.0-81.5)	98.2 (94.7-99.6)	98.3 (95.1-99.6)	74.9 (68.5-80.5)
Close contact	N	144	49.3	49.3 (37.2-61.4)	100 (95.1-100)	100 (90.0-100)	67.0 (57.3-75.7)
	OP-N	99	44.4	65.9 (50.1-79.5)	92.7 (82.4-98.0)	87.9 (71.8-96.6)	77.3 (65.3-86.7)
Other reason	N	29	34.5	70.0 (34.8-93.3)	100 (82.4-100)	100 (59.0-100)	86.4 (65.1-97.1)
	OP-N	24	50.0	58.3 (29.1-70.9)	91.7 (61.5-99.8)	87.5 (47.3-99.7)	68.8 (41.3-89.0)

Flowflex=Acon Labs Flowflex COVID-19 Antigen Home Test; MPBio=MP Biomedicals Rapid SARS-CoV-2 Antigen Test Card; Clinitest=Siemens-Healthineers CLINITEST Rapid COVID-19 Antigen Test; NPV=negative predictive value; PPV=positive predictive value; N = nasal sampling; OP-N = combined oropharyngeal and nasal sampling.

Table S8 User experience of the three Ag-RDTs. Up to 3% of the participants in the three nasal sampling groups and up to 5.5% in the OP-N sampling groups reported problems with conducting and/or interpreting the self-test. The higher percentage in the OP-N sampling group was primarily explained by fear or uncertainty of placing the swab (far) up the throat. Participants that performed nasal sampling more often indicated they performed and interpreted the test correctly than those that performed OP-N sampling.

	Flowflex	MPBio		Clinitest	
	Nasal sampling	Nasal sampling	Oropharyngeal-nasal sampling	Nasal sampling	Oropharyngeal-nasal sampling
	N = 1938	N = 1706	N = 543	N = 1579	N = 653
Did you experience problems when conducting the test? No. (%)					
Yes	30 (1.5)	49 (2.9)	25 (2.6)	36 (2.3)	36 (5.5)
I had a problem when performing the test, No. (%)	23 (76.7)	36 (73.5)	21 (84.0)	30 (83.3)	29 (80.6)
Unsure how deep to place the swab up my nose	8 (34.8)	3 (8.3)	1 (4.8)	1 (3.3)	1 (3.4)
Unsure how deep to place the swab up my throat	N/A	N/A	5 (23.8)	N/A	5 (17.2)
Fear of placing the swab too far up my nose	7 (30.4)	3 (8.3)	2 (9.5)	3 (10.0)	2 (6.9)
Fear of placing the swab too far up my throat	N/A	N/A	4 (19.0)	N/A	9 (31.0)
Spilled fluid	1 (4.3)	1 (2.8)	0 (0)	1 (3.3)	0 (0)
Other, unspecified	13 (56.6)	33 (91.7)	8 (38.1)	27 (90)	7 (24.1)
I had a problem interpreting the test, No. (%)	3 (10.0)	6 (12.2)	2 (8.0)	5 (13.9)	4 (11.1)
I saw no control line	1 (33.3)	6 (100)	2 (100)	5 (100)	4 (100)
Liquid didn’t travel through the cassette	1 (33.3)	2 (33.3)	1 (50.0)	5 (100)	4 (100)
Unclear whether the test was positive	3 (100)	2 (33.3)	1 (50.0)	1 (20.0)	2 (50.0)
Other, unspecified	1 (33.3)	2 (33.3)	2 (100)	4 (80)	2 (50.0)
The user manual was clear, No. (%)					
Agree	1879 (97.0)	1659 (97.2)	516 (95.0)	1323 (83.8)	575 (88.1)
Neutral	42 (2.2)	34 (2.0)	20 (3.7)	110 (7.0)	38 (5.8)
Disagree	11 (0.6)	8 (0.5)	5 (0.9)	144 (9.1)	37 (5.7)
Unknown	2 (0.1)	5 (0.3)	2 (0.4)	2 (0.1)	3 (0.5)
I am sure I performed the test correctly, No. (%)					
Agree	1838 (94.8)	1598 (93.7)	490 (90.2)	1525 (96.6)	584 (89.4)
In doubt	90 (4.6)	97 (5.7)	48 (8.8)	50 (3.2)	60 (9.2)
Disagree	4 (0.2)	5 (0.3)	3 (0.6)	2 (0.1)	5 (0.8)
Unknown	6 (0.3)	6 (0.3)	2 (0.4)	2 (0.1)	4 (0.6)
I am sure I interpreted the test correctly, No. (%)					
Agree	1915 (98.8)	1671 (98.0)	525 (96.7)	1555 (98.5)	634 (97.1)

In doubt	12 (0.6)	23 (1.3)	13 (2.4)	19 (1.2)	14 (2.1)
Disagree	3 (0.2)	5 (0.3)	3 (0.6)	3 (0.2)	1 (0.2)
Unknown	8 (0.4)	7 (0.4)	2 (0.4)	2 (0.1)	4 (0.6)

N/A = not applicable

Table S9 Follow-up information of participants that were tested again within 10 days after the initial test. Follow-up information was not available for 56/211 (26.5%), and 110/425 (25.9%), and 98/297 (33.0%) of nasal sampling participants with Flowflex, MPBio, and Clinitest and 45/179 (25.1%) and 79/231 (34.2%) of OP-N sampling participants with MPBio and Clinitest, that had an Ag-RDT result available and were tested negative with RT-PCR at their initial test site visit, respectively. Of the remaining participants 75/155 (48.4%), 161/315 (51.1%), and 100/199 (50.3%), and 61/134 (45.5%) and 68/152 (44.7%) were not tested within 10 days after the initial test, respectively. Overall, follow-up information was available for 71.1% (955/1343) of participants with a negative RT-PCR test result at baseline. Of this group, 51.3% (490/955) reported to have been re-tested within 10 days, with 16.3% of them (80/490) testing positive.

Test result available from the reference test and…	Flowflex	MPBio		Clinitest	
Type of sampling	Nasal	Nasal	OP-N	Nasal	OP-N
Sample size (Test during 10-day follow-up)	N = 80/155 (51.6%)	N = 154/315 (48.9%)	N = 73/134 (54.5%)	N = 99/199 (49.7%)	N = 84/152 (55.3%)
Type of test, No. (%)					
public health service COVID-19 test sites	25 (31.3)	50 (32.5)	26 (35.6)	36 (29.5)	29 (34.5)
RT-PCR test	23 (92.0)	45 (90.0)	24 (92.3)	33 (91.7)	25 (86.2)
Ag-RDT	0 (0)	2 (4.0)	1 (3.8)	0 (0)	1 (3.4)
Unknown	2 (8.0)	3 (6.0)	1 (3.8)	3 (8.3)	3 (10.3)
Other test site (incl. at work)	5 (6.3)	6 (3.9)	3 (4.1)	3 (2.5)	4 (4.8)
RT-PCR test	2 (40.0)	4 (66.7)	1 (33.3)	1 (33.3)	2 (50.0)
Ag-RDT	3 (60.0)	2 (33.3)	2 (66.7)	2 (66.7)	2 (50.0)
Unknown	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)
Hospital or general practitioner test site	2 (2.5)	1 (0.6)	0 (0)	1 (0.8)	0 (0)
RT-PCR test	1 (50.0)	0 (0)	N/A	0 (0)	N/A
Ag-RDT	1 (50.0)	0 (0)	N/A	1 (100)	N/A
Unknown	0 (0)	1 (100)	N/A	0 (0)	N/A
Self-test	68 (85.0)	119 (77.3)	61 (83.6)	80 (80.8)	74 (88.1)
Positive test result, No. (%)	11 (13.8)	19 (12.3)	16 (22.2)	18 (18.4)	16 (19.0)

OP-N = oropharyngeal and nasal; N/A = not applicable