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# **Performance evaluation of two SARS-CoV-2 IgG/IgM rapid tests (Covid-Presto and NG-Test) and one IgG automated immunoassay (Abbott)**

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# INTRODUCTION

1  
2  
3 Severe Acute Respiratory Syndrome Coronavirus 2 (SARS-CoV-2) is the etiological  
4 agent of the Coronavirus Disease 19 (COVID-19) pandemic. SARS-CoV-2 RNA RT-  
5 PCR from a naso-pharyngeal swab is the gold standard test to diagnose COVID-19.  
6 Serological tests are also available allowing serological surveys in different  
7 populations, in particular patients presenting strong COVID-19 suspicions with  
8 negative PCR. Serological tests also make it possible to catch up later with  
9 undiagnosed people at time of active infection, since antibodies have been found in  
10 almost all people who have been in contact with SARS-CoV-2 within a variable  
11 period depending on the severity of the infection [1,2]. Furthermore, studies showed  
12 that the kinetics of appearance of IgM and IgG were relatively close [3].  
13 Two types of tests are available to detect anti-SARS-CoV-2 antibodies: rapid lateral  
14 flow tests and automated immunoassays. Several studies have assessed analytical  
15 performances of the automated immunoassays [4-7]. On the other hand, although a  
16 very large number of rapid tests have been developed, few of them have been  
17 reliably evaluated with a suitable serum panel. However, this is very important to  
18 have data about the efficacy of these rapid tests to reliably detect anti-SARS-CoV-2  
19 antibodies, since their increasing use in the world.

20 The aim of this study was to assess the analytical performances (sensitivity and  
21 specificity) and agreement of two rapid tests and one automated immunoassay for  
22 detecting antibodies against SARS-CoV-2.

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## MATERIALS AND METHODS

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### **Patients and serum samples**

28 This evaluation was performed on 262 serum samples collected in the Virology  
29 Laboratory of Bichat-Claude Bernard and Saint-Louis University-Hospitals both in  
30 Paris, France.

31 Eighty-eight serum samples were collected from 54 patients with a confirmed  
32 COVID-19 diagnosis by a positive nasopharyngeal sample RT-PCR. Median age was  
33 52 years (range: 27-80), 36 were males. Among them, 29 were hospitalized in  
34 intensive care, 11 in infectious diseases.

35 We constituted a negative panel of 120 sera, all collected before November 2019, to  
36 assess the specificity, including samples for testing as part of routine clinical care  
37 (n=56) and serum samples corresponding to a cross-reactivity panel (n=64). These  
38 latter consisted of coronaviruses (HKU1, NL63, 229E and OC43; n=20), malarial  
39 (n=26), respiratory viruses (Influenza A [n=2], Influenza B [n=1] Respiratory Syncytial  
40 Virus [n=2], Metapneumovirus [n=1], Rhinovirus [n=1]), sera with acute CMV

41 infection (n=2), acute EBV infection (n=1), HIV-HBV co-infection (n=1), and acute  
42 Parvovirus B19 infection (n=1), Toxoplasma (n=1). In addition, we assessed five  
43 samples containing autoantibodies (four rheumatoid factor and one systemic lupus  
44 erythematosus).

45 We also assessed the serum of 54 health-care workers who presented clinical  
46 symptoms during the epidemic period for whom SARS-CoV-2 RT-PCR was negative  
47 or not carried out.

48

#### 49 **Rapid lateral flow tests**

50 We evaluated two lateral flow tests: Covid-Presto® test rapid Covid-19 IgG/IgM  
51 (AAZ, Boulogne-Billancourt, France) and NG-Test® IgM-IgG COVID-19 (NG Biotech,  
52 Guipry, France) according to the manufacturer's instructions. Five and ten  
53 microliters of serum for Covid-Presto® test and NG-Test®, respectively, were added  
54 and results were read and interpreted 10 minutes after depositing serum.

55

#### 56 **Automated immunoassay**

57 Abbott SARS-CoV-2 IgG kit (chemiluminescent microparticle immunoassay) (Abbott,  
58 IL, USA) was performed according to the manufacturer's instructions. The assay cut-  
59 off is an index of 1.40 and the assigned grey zone is comprised between 1.12 and 1.68.

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63 **Statistical analysis**

64 All statistical analyses were performed using Excel. To assess sensitivity, RT-PCR  
65 results were chosen as gold standard. Cohen kappa statistics and absolute agreement  
66 were calculated to evaluate the agreement between the different tests.

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68 **Ethics**

69 All participants were not opposed to the collection of their data.

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## RESULTS

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74 **Sensitivity assessment**

75 Sensitivity of Covid-Presto® test was assessed on 88 samples collected between day 4  
76 and day 42 after onset of symptoms and sensitivity of the NG-Test® was assessed on a  
77 subgroup of 59 samples among the 88 samples tested with Covid-Presto® test,  
78 collected between days 7 and 28 after onset of symptoms (Table 1).

79 Sensitivity of Covid-Presto® test for IgM was 67% (n=12/18), 88% (n=29/33) and 76%  
80 (n=28/37) for samples collected between days 4 and 9, between days 10 and 14, and  
81 after 14 days after onset of symptoms, respectively. Sensitivity of Covid-Presto® test  
82 for IgG was 72% (n=13/18), 94% (n=31/33) and 100% (n=37/37) for samples collected  
83 between days 4 and 9, between days 10 and 14, and after 14 days after onset of

84 symptoms, respectively. When combining IgM and IgG, sensitivity of Covid-Presto®  
85 test was 83% (n=15/18), 97% (n=32/33) and 100% (n=37/37) for samples collected  
86 between days 4 and 9, between days 10 and 14, and after 14 days after onset of  
87 symptoms, respectively.

88 Sensitivity of NG-Test® for IgM was 83% (n=5/6), 100% (n=22/22) and 97% (n=30/31)  
89 for samples collected between days 7 and 9 after, between days 10 and 14, and after  
90 14 days after onset of symptoms, respectively. Sensitivity of NG-Test® test for IgG  
91 was 83% (n=5/6), 96% (n=21/22) and 97% (n=30/31) for samples collected between  
92 days 7 and 9, between days 10 and 14, and after 14 days after onset of symptoms,  
93 respectively. When combining IgM and IgG, sensitivity of NG-Test® test was 83%  
94 (n=5/6), 100% (n=22/22) and 97% (n=30/31) for samples collected between days 7 and  
95 9, between days 10 and 14, and after 14 days after onset of symptoms, respectively.

96 Among the 59 serum samples of this PCR positive panel tested by the two rapid  
97 tests, 57 were compared with Abbott SARS-CoV-2 IgG automated immunoassay.  
98 Sensitivity of Abbott IgG test was 67% (n=4/6), 100% (n=22/22) and 100% (n=29/29) for  
99 samples collected between days 7 and 9, between days 10 and 14, and after 14 days  
100 after onset of symptoms, respectively.

101 Agreement between Abbott assay and rapid tests (IgM/IgG combined) was of 96.5%  
102 (n=55/57). In one case, the two rapid tests detected IgG that were not detected by  
103 Abbott (index=0.94), this sample was collected between days 7 and 9 after symptoms  
104 onset. For the second case, IgG were detected in the greyzone of Abbott (index=1.45)

105 but not by NG-Test®. This latter sample was collected between days 10 and 14 after  
106 symptoms onset and IgM were positive with the two rapid tests.

107

### 108 **Specificity assessment**

109 Specificity of Covid-Presto® test was assessed on 120 samples described in the  
110 methods section. Specificity of NG-Test® and Abbott assay was assessed on a  
111 subgroup of 52 samples among the 120 samples tested with Covid-Presto® test (Table  
112 1).

113 Specificity of Covid-Presto® test assessed on 120 samples was 100% for IgM and  
114 98.3% for IgG. For IgG one false positive result was observed with one sample  
115 containing malarial antibodies, and one false positive result was observed with one  
116 pre-epidemic sample.

117 Specificity of NG-Test® assessed on 52 samples was 86.5% for IgM and 96.2% for IgG.  
118 Regarding the seven samples false positive for IgM, two were from pre-epidemic  
119 panel and five were from samples containing malarial antibodies. Regarding IgG, the  
120 two false-positive samples belonged to the pre-epidemic panel.

121 Specificity of Abbott SARS-CoV-2 IgG kit was 96.2%. The two false-positive samples  
122 had low titers (1.12 and 2.28), both samples contained malarial antibodies.

123

### 124 **Agreement between the two lateral flow tests and the automated immunoassay**

125 Agreement between the assays was performed on 163 samples: (i) 57 samples from  
126 the positive panel, (ii) 52 samples from the negative panel for which results were

127 obtained for the two rapid tests and the Abbott SARS-CoV-2 IgG assay, and (iii) 54  
128 samples collected from health workers (Table 2).

129 Absolute agreement between Covid-Presto<sup>®</sup> and NG-Test<sup>®</sup> was 82.8% (n=135/163,  
130  $\kappa=0.643$ ) and 96.9% (n=158/163,  $\kappa=0.937$ ) for IgM and IgG, respectively. Regarding  
131 IgM results, 26 samples were negative with Covid-Presto<sup>®</sup> and positive with NG-  
132 Test<sup>®</sup>, including 8 NG-Test<sup>®</sup> false-positive results (6 malarial and 2 pre-epidemic  
133 sera). Seventeen of the 18 remaining samples were sera of COVID-positive patients  
134 collected after day 14 after symptoms onset. In addition, two sera were negative with  
135 NG-Test<sup>®</sup> and positive with Covid-Presto<sup>®</sup> test, both corresponding to COVID-  
136 positive patients (collected at day 8 and day 14 after symptoms onset). Regarding  
137 IgG results, three samples were negative with Covid-Presto<sup>®</sup> and positive with NG-  
138 Test<sup>®</sup> including 2 false-positive pre-epidemic sera and one serum from a COVID-  
139 positive patient collected at day 13 after symptoms onset. Furthermore, two sera  
140 were negative with NG-Test<sup>®</sup> and positive with Covid-Presto<sup>®</sup> test: one serum from a  
141 COVID-positive patient collected after day 14 after symptoms onset and one serum  
142 from a healthcare worker.

143 Evaluation between Covid-Presto<sup>®</sup> IgG test and Abbott SARS-CoV-2 IgG assay was  
144 performed on 159 available samples with sufficient volume of serum, showing an  
145 absolute agreement of 96.9% (n=154/159,  $\kappa=0.936$ ). Three samples were found  
146 negative with Covid-Presto<sup>®</sup> IgG test and positive with Abbott SARS-CoV-2 IgG, all  
147 with low Abbott index (1.87, 2.16, 2.57). Two of these three samples were issued from  
148 healthcare workers and the remaining one was from a COVID-positive patient (day

149 13 after symptoms onset) showing positive IgM. Two were positive with Covid-  
150 Presto® IgG test and negative with Abbott SARS-CoV-2 IgG (index:0.58 and 0.78),  
151 both samples belonged to the group of healthcare workers.

152 Assessment between NG-Test® IgG and Abbott SARS-CoV-2 IgG was performed on  
153 153 available samples with sufficient volume of serum showing an agreement of  
154 97.4% (n=149/153,  $\kappa=0.947$ ). One sample was positive in the greyzone with Abbott  
155 SARS-CoV-2 IgG assay (index: 1.45) and negative with NG-Test® IgG corresponding  
156 to the serum of a COVID-positive patient (day 11 after symptoms onset). Three  
157 samples were negative with Abbott SARS-CoV-2 IgG assay (index: 0.07, 0.12 and  
158 0.94) and positive with NG-Test® corresponding to 2 false-positive pre-epidemic sera  
159 and one COVID-positive patient (day 8 after symptoms onset).

160 Predictive positive and negative values were calculated for 210, 113 and 111 serum  
161 samples for Covid-Presto®, NG-Test® and Abbott tests, respectively. For the rapid  
162 tests, IgM and IgG results were combined for this analysis. All patients with positive  
163 PCR were considered COVID-19 positive and all pre-pandemic samples were  
164 considered COVID-19 negative. PPV was 97.7%, 89.1% and 96.6% for Covid-Presto®,  
165 NG-Test® and Abbott tests, respectively. NPV was 97.5%, 95.9% and 94.3% for Covid-  
166 Presto®, NG-Test® and Abbott, respectively.

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## DISCUSSION

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171

172 In the present study, we evaluated two different lateral flow tests (Covid-Presto® and  
173 NG-Test®) and compared their performances to that of the automated Abbott  
174 immunoassay using the same samples panel.

175

176 Sensitivity has been assessed using a panel of 88 serum samples of COVID-19-  
177 infected patients (confirmed with a positive PCR), serum was collected between day  
178 4 and day 42 after symptoms onset. Sensitivity for IgM, among the samples collected  
179 before day 9 after symptoms onset, was 67% and 83% for Covid-Presto® test and NG-  
180 Test®, respectively. In the recent study of Nicol et al., they found sensitivity of NG-  
181 Test for IgM of 43.8% for the samples collected before day 7 after symptoms onset  
182 and of 81.8% among all samples [5]. The excellent sensitivity of Covid-Presto® test  
183 observed in our study confirmed the findings of the Prazuck et al. study showing  
184 100% of sensitivity in samples collected more than 15 days after symptoms [8].

185 Among some samples collected before day 10 after symptoms onset, a simultaneous  
186 detection of IgM and IgG antibodies has been detected. These findings are in line  
187 with the antibodies kinetics described for IgM and IgG also using lateral flow rapids,  
188 as previously described with other techniques [3]. In the present study for Covid-  
189 Presto® test, it allowed to increase the sensitivity from 67% when only IgM are taken  
190 into account to 83% when both IgM and IgG are taken into account, highlighting the

191 important added value to interpret the rapid tests by combining IgM and IgG  
192 antibodies.

193 Sensitivity for IgG in samples collected later than 10 days after symptoms onset was  
194 excellent with the different tests being equal to 97.1%, 96.2% and 100% for Covid-  
195 Presto®, NG-Test®, and Abbott, respectively. Thus, both rapid tests showed an  
196 excellent sensitivity for IgG with a very good agreement with Abbott. A previous  
197 study assessing Abbott test performance showed sensitivity of 100% for IgG for  
198 samples collected after 15 days after symptoms onset and of 69% for samples  
199 collected between 9 and 14 days after symptoms onset [6]. In this latter study, results  
200 sensitivity for IgG were similar using NG-Test® [6]. In another study, IgG sensitivity  
201 of Abbott test was 91.8% for patients hospitalized 15 days after symptoms onset and  
202 95.7% for patients non-hospitalized 20 days after symptoms onset.

203 A limitation of our study could be that most of the patients of the positive panel  
204 presented severe infections, since 74% of them were hospitalized in infectious disease  
205 unit or in intensive care. Interestingly, among the 14 out-patients, samples were  
206 collected for 9 of them 10 days after symptoms onset, showing positive IgM and/or  
207 IgG in seven cases with Covid-Presto® test. Insufficient quantity of serum for these  
208 patients was available to also test with NG-Test® and Abbott. Previous studies have  
209 reported that the kinetics and intensity of immune response could differ depending  
210 on the disease severity [1,2], thus it will be needed to also evaluate rapid tests in mild  
211 and pauci-symptomatic patients. Another limitation is the difference in the number  
212 of tested samples for the early panel (serum samples collected before 9 days after

213 symptoms onset) between the two rapid tests that which can bias the comparison  
214 between these tests for this group. A limitation is that we make this evaluation from  
215 serum samples and not from capillary blood specimens.

216 Regarding specificity evaluation, a crucial point for rapid tests, we used a large panel  
217 with 120 pre-endemic samples including 64 representatives of different profiles that  
218 can generate possible cross-reactivity. In our study, we showed an excellent  
219 specificity, above 96% in all cases and equal to 100% for IgM with Covid-Presto® test.

220 The excellent specificity of Covid-Presto® test was also observed in the study of  
221 Prazuck et al. [8]. In our study, the only issue regarding specificity is for IgM with  
222 NG-Test®, since specificity is only of 86.5%. However, this low specificity is mainly  
223 due to cross-reactivity with sera containing reactivity malarial antibodies. In the  
224 study of Nicol et al. IgM specificity with NG-Test® was 95.3% [5], higher than in our  
225 study, however their negative panel contained no serum with malaria antibodies.

226 Regarding automated immunoassay, we showed a very good specificity of 96.2% for  
227 IgG with Abbott, confirming previous results of 99.3%, 99.6% and 100% [9]. Serum  
228 samples containing malarial antibodies are absent or underrepresented in the  
229 negative panel of the other studies, although they are known to generate possible  
230 cross reactivity. This is very important to include it in the negative panel, since this is  
231 a differential diagnosis in patients returning from malaria endemic region with Flu-  
232 like symptoms. Overall, in our study, we observed a very good PPV and NPV for  
233 both rapid tests.

234

235 In conclusion, analytical performances for detection of anti-SARS-CoV-2 IgG  
236 antibodies by two lateral flow rapid tests are very good and quite comparable to  
237 those obtained with automated immunoassay. However, serological tests should be  
238 used after day 10 following symptoms onset. Before this, RT-PCR is the gold  
239 standard test for COVID-19 diagnosis. The interpretation by combining IgM and IgG  
240 increased sensitivity of rapid tests. The presence of isolated IgM should be cautiously  
241 interpreted due to the possible false-positive reactions. Finally, the rapid tests must  
242 be reliably evaluated with adequate and large panels including early seroconversion  
243 and possible cross-reactive samples, before their large use and particular interest in  
244 low-resource settings.

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**Table 1.** Performance characteristics of two rapid tests and one automated immunoassay.

Test	Sensitivity				Specificity		
	Days after symptoms onset			Overall	Pre-epidemic samples	Cross-reactivity panel	Overall
	≤9	10-14	>14				
<b>IgM</b>							
Covid-Presto®	67 % (12/18)	88 % (29/33)	76 % (28/37)	78.4 % (69/88)	100 % (56/56)	100 % (64/64)	100 % (120/120)
NG-Test®	83 % (5/6)	100 % (22/22)	97 % (30/31)	96.6 % (57/59)	91.7 % (22/24)	82.1 % (23/28)	86.5 % (45/52)
<b>IgG</b>							
Covid-Presto®	72 % (13/18)	94 % (31/33)	100 % (37/37)	92.0 % (81/88)	98.2 % (55/56)	98.4 % (63/64)	98.3 % (118/120)
NG-Test®	83 % (5/6)	96 % (21/22)	97 % (30/31)	94.9 % (56/59)	91.7 % (22/24)	100 % (28/28)	96.2 % (50/52)
Abbott	67 % (4/6)	100 % (22/22)	100 % (29/29)	96.5 % (55/57)	100 % (24/24)	92.9 % (26/28)	96.2 % (50/52)
<b>IgM + IgG</b>							
Covid-Presto®	83 % (15/18)	97 % (32/33)	100 % (37/37)	95.5 % (84/88)	NA	NA	NA
NG-Test®	83 % (5/6)	100 % (22/22)	97 % (30/31)	96.6 % (57/59)	NA	NA	NA

NA: not applicable

**Table 2.** Agreement between two rapid tests and one automated immunoassay.

Test		Covid-Presto®	NG-Test®	Abbott
Covid-Presto®	IgM		82.8 % (n = 135/163)	NA
	IgG		96.9 % (n = 158/163)	96.9 % (n = 154/159)
NG-Test®	IgM			NA
	IgG			97.4 % (n = 149/153)
Abbott				

NA: not applicable