

Clinical Study Report

Viral antigen detection reagent (colloidal gold)

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Ltd**

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Study Summary

The Viral antigen detection reagent (colloidal gold) produced by Tai zhou Sun Trine Biotechnology Co.,Ltd was clinically validated according to the requirements of the "Management Measures for Registration of In Vitro Diagnostic Reagents (Trial Implementation)". Here, we evaluated the analytical specificity and diagnostic Sensitivity and Specificity of the kit. See the following for specific assessment methods, results, analysis and evaluation.

1. Cross reactivity and Interference Study

1.1 Cross-Reactivity Study

1.1.1 Main product

Product name: Viral antigen detection reagent (colloidal gold)

LOT 1: 20201012 LOT 2: 20201019 LOT 3: 20201026

1.1.2 Purpose

The following study was done to assess the cross-reactivity and exclusivity of the Viral antigen detection reagent (colloidal gold) in high concentration.

1.1.3 Method

Some related pathogens, high prevalence disease agents and pathogenic flora that are reasonably likely to be encountered in the clinical specimen with certain concentration were tested by each batch of the COVID-19 Antigen Rapid Test. Every specimen was tested for three times.

1.1.4 Interpretation of results

- Negative: There is only a red line appears in the quality control area (C), and no line appears in the test area (T).
- Positive: There are two red lines appear. One is in the test area (T) and the other is in the quality control area (C).
- Invalid: There is no red line displays in the quality control area (C). This indicates that the incorrect operation or the test cassette has deteriorated or damaged. Repeat the test with a new kit. If the problem persists, stop using this lot number immediately and contact your local supplier.

1.1.5 Acceptance criteria

The Virus or Bacteria culture with certain concentration in negative clinical matrix should not influence the negative results.

1.1.6 Results

Microorganism	Concentration	LOT1	LOT2	LOT3
human coronavirus OC 43	1.05×10^5 pfu/mL	No	No	No
human coronavirus 229E	1.26×10^5 pfu/mL	No	No	No
human coronavirus NL63	1.47×10^5 pfu/mL	No	No	No
human coronavirus HKU1	1.36×10^5 pfu/mL	No	No	No
MERS coronavirus	1.61×10^5 pfu/mL	No	No	No

No: Only the C line appears, indicates negative.

Yes: Two red lines appear, indicates Positive.

1.1.7 Conclusion

Related pathogens, high prevalence disease agents and pathogenic flora that are reasonably likely to be encountered in the clinical specimen were tested. It showed that no cross-reactivity was found in these tests with the Viral antigen detection reagent (colloidal gold).

1.2 Interference Study

1.2.1 Main product

Product name: Viral antigen detection reagent (colloidal gold)

LOT 1: 20201012 LOT 2: 20201019 LOT 3: 20201026

1.2.2 Purpose

The following study was done to confirm that the substances do not interfere with the performance of the Viral antigen detection reagent (colloidal gold) in certain concentration.

1.2.3 Method

A group of 8 kinds of potentially interference substances were tested by each batch of Viral antigen detection reagent (colloidal gold) to assess the potential for interference that are reasonably likely to be encountered in the clinical specimen. Each sample was tested in three times.

1.2.4 Interpretation of results

- Negative: There is only a red line appears in the quality control area (C), and no line appears in the test area (T).
- Positive: There are two red lines appear. One is in the test area (T) and the other is in the quality control area (C).
- Invalid: There is no red line displays in the quality control area (C). This indicates that the incorrect operation or the test cassette has deteriorated or damaged. Repeat the test with a new kit. If the problem persists, stop using this lot number immediately and contact your local supplier.

1.2.5 Acceptance criteria

All of potentially interference substances spiked into the negative samples should be tested negative.

All of potentially interference substances spiked into the positive samples should be tested positive.

1.2.6 Results

Interfering samples	Concentration	Interference (Yes/No)					
		LOT 1		LOT 2		LOT 3	
		Negative sample	Positive sample	Negative sample	Positive sample	Negative sample	Positive sample
bilirubin	20 mg/dL	No	No	No	No	No	No
hemoglobin	500 mg/dL	No	No	No	No	No	No
triglyceride	2000 mg/dL	No	No	No	No	No	No
rheumatoid factor	596 IU/ml	No	No	No	No	No	No
mucin	30 mg/mL	No	No	No	No	No	No
The new influenza A (H1N1) virus	$1.05 \times 10^{5.67}$ pfu/mL	No	No	No	No	No	No
Influenza B virus	$0.7 \times 10^{6.67}$ pfu/mL	No	No	No	No	No	No
Respiratory syncytial virus	1.47×10^5 pfu/mL	No	No	No	No	No	No

1.2.7 Conclusion

No interference was observed between any of the potentially interference substances tested with the Viral antigen detection reagent (colloidal gold).

2.Diagnostic Sensitivity And Specificity

2.1 Purpose

To validate the diagnostic sensitivity and specificity of Viral antigen detection reagent (colloidal gold)

2.2 Materials

- Product name: Viral antigen detection reagent (colloidal gold)
- Lot No.: P20200106
- The registration certificate number is 20203400057
- Detection Kit For Coronavirus Disease 2019 (COVID-19) (PCR Fluorescence Probe) produced by Shanghai Zhijiang Biotechnology Co., Ltd

2.3 Experimental design and research method selection

2.3.1 Sample size and Sample size determination basis

- (1) Should meet statistical requirements .This study uses a single target value method to calculate the sample size. In order to meet statistical requirements, and take into account the possible fall-off and other factors.

2.3.2 Sample selection basis, selection criteria, exclusion criteria and elimination criteria .

- (1) Crowd selection criteria :

- Suspected cases and clinically diagnosed cases of COVID-19 infection;
- The remaining samples after routine clinical testing;
- The collection and processing of samples meet the requirements of standard laboratory operations and product instructions;
- The relevant information of the sample is complete, including subject number, age, gender, sample type, etc.

- (2) Exclusion criteria:

- The sample collection time or information is not clear;
- Insufficient sample size due to errors in test operation;
- It was found that the specimen preservation process was contaminated before the test operation;

- (3) Elimination criteria:

- Samples with human error in the test;
- Other samples that failed to complete the test due to various reasons;
- Any other reason considered by the principal investigator of each clinical trial institution.

Among them, after excluding or excluding some samples according to the above standards, it must be ensured that the sample size meets the requirements of this test plan.

2.4 Study Design

2.4.1 Sample collection

Subjects were patients admitted to the hospital due to a respiratory virus infection; Total about 211 samples were collected; Total 107 anterior nasal swab samples from COVID-19 infected patients and 104 non-COVID-19 infected anterior nasal swab samples were tested.

2.4.2 Sample requirement

All the samples were confirmed by Detection Kit For Coronavirus Disease 2019 (COVID-19) (PCR Fluorescence Probe) produced by Shanghai Zhijiang Biotechnology Co., Ltd. Samples were to be randomly chosen and double-black labeled.

2.4.3 Test conduction

- All tests were performed by the clinical technicians in each clinical laboratory according to the manufacturer's instructions using the confirmed samples.
- Visual interpretations of the results of COVID-19 Antigen Test were made independently by the clinical technician.

2.5 Evaluation Criteria

- Negative: Only a red line appears in the quality control area (C), and no line appears in the test area (T).
- Positive: Two red lines appear. One is in the test area (T) and the other is in the quality control area (C).
- Invalid : No red line displays in the quality control area (C). This indicates that the incorrect operation or the test cassette has deteriorated or damaged. Repeat the test with a new kit. If the problem persists, stop using this lot number immediately and contact your local supplier.

2.6 Results

211 samples were collected from selected subjects, total 107 anterior nasal swab samples from COVID-19 infected patients and 104 non-COVID-19 infected anterior nasal swab samples were tested. All samples were confirmed by nucleic acid test (RT-PCR). Calculated the specificity and sensitivity, the results are as follows:

Table: 211 samples test results

Assessment reagent	Nucleic acid test (PCR)		Total
	Positive(+)	Negative(-)	
Positive(+)	95	2	97
Negative(-)	12	102	114
Total	107	104	211

- ① Clinical sensitivity (positive coincidence rate): $95/107 = 88.79\%$
 95% confidence interval: $p \pm 1.96 \times [p(1-p)/n]^{1/2} = 81.41\% - 93.47\%$
 Clinical specificity (negative coincidence rate): $102/104 = 98.08\%$
 95% Confidence interval: $p \pm 1.96 \times [p(1-p)/n]^{1/2} = 93.26\% - 99.47\%$
 Overall coincidence rate: $197/211 = 93.36\%$
 95% Confidence interval: $p \pm 1.96 \times [p(1-p)/n]^{1/2} = 89.17\% - 96.01\%$

② Consistency coefficient Kappa value (K)

$$Kappa=(P_A - P_e) / (1 - P_e)$$

$$P_A=(A + D) / (A + B + C + D)=0.9336$$

$$P_e=[(A + B) (A + C) + (C + D) (B + D)] / (A + B + C + D)^2$$
$$=0.4994$$

$$Kappa=(P_A - P_e) / (1 - P_e)=0.8675$$

Kappa= 0.8675 (K>0.75), it can be considered that the strength of agreement between the assessment reagent result and the nucleic acid test result is extremely high. (K=0.8675>0.75)

Clinical Record Sample No.	Gender	Age	Sample type	Nucleic acid test Results	Assessment reagent test results	Diagnosis
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1	M	35	anterior nasal swab	+	+	COVID-19
2	M	56	anterior nasal swab	+	-	COVID-19
3	F	85	anterior nasal swab	+	+	COVID-19
4	F	59	anterior nasal swab	-	-	Normal medical
5	M	25	anterior nasal swab	+	+	COVID-19
6	M	67	anterior nasal swab	+	+	COVID-19
7	M	37	anterior nasal swab	+	+	COVID-19
8	F	68	anterior nasal swab	+	+	COVID-19
9	F	60	anterior nasal swab	+	+	COVID-19
10	F	80	anterior nasal swab	-	-	Normal medical
11	M	73	anterior nasal swab	+	+	COVID-19
12	M	42	anterior nasal swab	-	-	Normal medical
13	M	59	anterior nasal swab	-	-	Normal medical
14	F	34	anterior nasal swab	-	-	Normal medical
15	M	63	anterior nasal swab	-	-	Normal medical
16	M	55	anterior nasal swab	+	+	COVID-19
17	F	68	anterior nasal swab	+	+	COVID-19
18	M	81	anterior nasal swab	-	-	Normal medical
19	M	68	anterior nasal swab	+	+	COVID-19
20	M	57	anterior nasal swab	-	-	Normal medical
21	F	66	anterior nasal swab	+	+	COVID-19
22	M	57	anterior nasal swab	+	+	COVID-19

23	F	46	anterior nasal swab	+	+	COVID-19
24	F	51	anterior nasal swab	-	-	Normal medical
25	F	47	anterior nasal swab	-	-	Normal medical
26	M	33	anterior nasal swab	+	+	COVID-19
27	M	43	anterior nasal swab	+	+	COVID-19
28	F	19	anterior nasal swab	-	-	Normal medical
29	F	43	anterior nasal swab	-	-	Normal medical
30	M	10	anterior nasal swab	+	-	COVID-19
31	F	60	anterior nasal swab	+	+	COVID-19
32	F	41	anterior nasal swab	-	-	Normal medical
33	M	71	anterior nasal swab	-	-	Normal medical
34	M	61	anterior nasal swab	-	-	Normal medical
35	F	67	anterior nasal swab	+	+	COVID-19
36	M	80	anterior nasal swab	-	-	Normal medical
37	F	71	anterior nasal swab	+	+	COVID-19
38	M	63	anterior nasal swab	+	+	COVID-19
39	M	35	anterior nasal swab	+	+	COVID-19
40	F	73	anterior nasal swab	-	-	Normal medical
41	F	58	anterior nasal swab	-	-	Normal medical
42	M	32	anterior nasal swab	-	+	Normal medical
43	M	57	anterior nasal swab	-	-	Normal medical
44	F	56	anterior nasal swab	+	-	COVID-19

45	M	61	anterior nasal swab	-	-	Normal medical
46	M	46	anterior nasal swab	-	-	Normal medical
47	M	51	anterior nasal swab	-	-	Normal medical
48	M	74	anterior nasal swab	+	+	COVID-19
49	M	49	anterior nasal swab	-	-	Normal medical
50	M	49	anterior nasal swab	+	+	COVID-19
51	M	28	anterior nasal swab	+	+	COVID-19
52	M	48	anterior nasal swab	-	-	Normal medical
53	M	67	anterior nasal swab	+	+	COVID-19
54	F	65	anterior nasal swab	+	+	COVID-19
55	F	56	anterior nasal swab	+	+	COVID-19
56	F	59	anterior nasal swab	+	+	COVID-19
57	F	67	anterior nasal swab	-	-	Normal medical
58	M	62	anterior nasal swab	+	+	COVID-19
59	F	25	anterior nasal swab	+	+	COVID-19
60	F	48	anterior nasal swab	+	+	COVID-19
61	M	48	anterior nasal swab	-	-	Normal medical
62	F	37	anterior nasal swab	-	-	Normal medical
63	M	68	anterior nasal swab	-	-	Normal medical
64	M	73	anterior nasal swab	-	-	Normal medical
65	F	66	anterior nasal swab	-	-	Normal medical
66	M	35	anterior nasal swab	-	-	Normal medical

67	M	29	anterior nasal swab	-	-	Normal medical
68	F	66	anterior nasal swab	+	+	COVID-19
69	M	54	anterior nasal swab	+	-	COVID-19
70	F	56	anterior nasal swab	-	-	Normal medical
71	M	82	anterior nasal swab	+	+	COVID-19
72	F	67	anterior nasal swab	+	+	COVID-19
73	F	60	anterior nasal swab	-	-	Normal medical
74	M	59	anterior nasal swab	+	+	COVID-19
75	F	56	anterior nasal swab	+	+	COVID-19
76	F	43	anterior nasal swab	+	+	COVID-19
77	F	62	anterior nasal swab	+	+	COVID-19
78	M	47	anterior nasal swab	-	-	Normal medical
79	M	54	anterior nasal swab	-	-	Normal medical
80	F	57	anterior nasal swab	-	-	Normal medical
81	M	57	anterior nasal swab	+	+	COVID-19
82	F	50	anterior nasal swab	+	+	COVID-19
83	F	38	anterior nasal swab	+	-	COVID-19
84	M	57	anterior nasal swab	+	+	COVID-19
85	M	33	anterior nasal swab	-	-	Normal medical
86	F	88	anterior nasal swab	+	+	COVID-19
87	M	79	anterior nasal swab	-	-	Normal medical
88	F	64	anterior nasal swab	+	+	COVID-19

89	M	71	anterior nasal swab	+	-	COVID-19
90	F	51	anterior nasal swab	-	-	Normal medical
91	M	67	anterior nasal swab	-	-	Normal medical
92	M	44	anterior nasal swab	-	-	Normal medical
93	F	64	anterior nasal swab	+	+	COVID-19
94	M	68	anterior nasal swab	-	-	Normal medical
95	F	49	anterior nasal swab	+	+	COVID-19
96	F	38	anterior nasal swab	-	-	Normal medical
97	M	31	anterior nasal swab	+	+	COVID-19
98	F	35	anterior nasal swab	+	+	COVID-19
99	M	58	anterior nasal swab	+	-	COVID-19
100	F	74	anterior nasal swab	+	+	COVID-19
101	M	51	anterior nasal swab	+	+	COVID-19
102	M	56	anterior nasal swab	-	-	Normal medical
103	M	61	anterior nasal swab	+	+	COVID-19
104	M	89	anterior nasal swab	-	-	Normal medical
105	M	77	anterior nasal swab	+	+	COVID-19
106	F	60	anterior nasal swab	+	+	COVID-19
107	M	49	anterior nasal swab	+	+	COVID-19
108	M	39	anterior nasal swab	-	-	Normal medical
109	F	58	anterior nasal swab	+	+	COVID-19
110	F	50	anterior nasal swab	+	+	COVID-19

111	F	85	anterior nasal swab	+	+	COVID-19
112	F	65	anterior nasal swab	-	-	Normal medical
113	F	62	anterior nasal swab	+	+	COVID-19
114	M	69	anterior nasal swab	-	-	Normal medical
115	F	30	anterior nasal swab	-	-	Normal medical
116	F	52	anterior nasal swab	-	-	Normal medical
117	M	69	anterior nasal swab	+	+	COVID-19
118	F	82	anterior nasal swab	-	-	Normal medical
119	F	75	anterior nasal swab	-	-	Normal medical
120	M	80	anterior nasal swab	-	-	Normal medical
121	M	64	anterior nasal swab	+	+	COVID-19
122	F	40	anterior nasal swab	-	-	Normal medical
123	M	63	anterior nasal swab	-	-	Normal medical
124	M	56	anterior nasal swab	+	+	COVID-19
125	M	66	anterior nasal swab	-	-	Normal medical
126	M	63	anterior nasal swab	+	+	COVID-19
127	M	23	anterior nasal swab	-	-	Normal medical
128	M	68	anterior nasal swab	+	+	COVID-19
129	F	35	anterior nasal swab	-	-	Normal medical
130	M	50	anterior nasal swab	-	-	Normal medical
131	F	33	anterior nasal swab	+	+	COVID-19
132	F	50	anterior nasal swab	-	-	Normal medical

133	M	66	anterior nasal swab	+	+	COVID-19
134	F	68	anterior nasal swab	-	-	Normal medical
135	M	70	anterior nasal swab	+	+	COVID-19
136	F	52	anterior nasal swab	-	-	Normal medical
137	F	61	anterior nasal swab	+	+	COVID-19
138	F	65	anterior nasal swab	-	-	Normal medical
139	F	30	anterior nasal swab	+	+	COVID-19
140	M	79	anterior nasal swab	-	-	Normal medical
141	M	69	anterior nasal swab	-	-	Normal medical
142	F	70	anterior nasal swab	-	-	Normal medical
143	F	53	anterior nasal swab	+	-	COVID-19
144	F	69	anterior nasal swab	-	-	Normal medical
145	M	61	anterior nasal swab	+	+	COVID-19
146	F	50	anterior nasal swab	+	+	COVID-19
147	M	59	anterior nasal swab	+	+	COVID-19
148	M	43	anterior nasal swab	-	-	Normal medical
149	M	49	anterior nasal swab	-	-	Normal medical
150	F	59	anterior nasal swab	-	-	Normal medical
151	F	71	anterior nasal swab	+	+	COVID-19
152	F	67	anterior nasal swab	+	+	COVID-19
153	F	80	anterior nasal swab	-	-	Normal medical
154	F	67	anterior nasal swab	+	+	COVID-19

155	M	32	anterior nasal swab	-	-	Normal medical
156	M	48	anterior nasal swab	-	-	Normal medical
157	F	50	anterior nasal swab	+	-	COVID-19
158	M	36	anterior nasal swab	-	+	Normal medical
159	M	42	anterior nasal swab	-	-	Normal medical
160	F	56	anterior nasal swab	+	+	COVID-19
161	M	46	anterior nasal swab	-	-	Normal medical
162	F	49	anterior nasal swab	-	-	Normal medical
163	M	61	anterior nasal swab	+	+	COVID-19
164	M	75	anterior nasal swab	+	+	COVID-19
165	F	34	anterior nasal swab	+	+	COVID-19
166	M	62	anterior nasal swab	+	+	COVID-19
167	F	50	anterior nasal swab	-	-	Normal medical
168	F	48	anterior nasal swab	+	+	COVID-19
169	M	65	anterior nasal swab	+	-	COVID-19
170	F	56	anterior nasal swab	+	+	COVID-19
171	F	25	anterior nasal swab	+	+	COVID-19
172	F	37	anterior nasal swab	+	+	COVID-19
173	F	29	anterior nasal swab	-	-	Normal medical
174	F	38	anterior nasal swab	-	-	Normal medical
175	F	66	anterior nasal swab	-	-	Normal medical
176	M	79	anterior nasal swab	+	+	COVID-19

177	M	39	anterior nasal swab	-	-	Normal medical
178	M	28	anterior nasal swab	-	-	Normal medical
179	M	75	anterior nasal swab	-	-	Normal medical
180	F	67	anterior nasal swab	-	-	Normal medical
181	M	39	anterior nasal swab	+	+	COVID-19
182	F	22	anterior nasal swab	+	+	COVID-19
183	F	27	anterior nasal swab	-	-	Normal medical
184	F	26	anterior nasal swab	+	+	COVID-19
185	F	36	anterior nasal swab	-	-	Normal medical
186	M	36	anterior nasal swab	+	+	COVID-19
187	F	51	anterior nasal swab	-	-	Normal medical
188	M	41	anterior nasal swab	+	+	COVID-19
189	F	29	anterior nasal swab	+	+	COVID-19
190	F	22	anterior nasal swab	-	-	Normal medical
191	M	36	anterior nasal swab	-	-	Normal medical
192	M	63	anterior nasal swab	-	-	Normal medical
193	F	38	anterior nasal swab	+	+	COVID-19
194	F	62	anterior nasal swab	+	+	COVID-19
195	F	33	anterior nasal swab	+	-	COVID-19
196	F	55	anterior nasal swab	-	-	Normal medical
197	F	49	anterior nasal swab	-	-	Normal medical
198	F	23	anterior nasal swab	+	+	COVID-19

199	F	12	anterior nasal swab	-	-	Normal medical
200	F	37	anterior nasal swab	-	-	Normal medical
201	F	61	anterior nasal swab	-	-	Normal medical
202	M	51	anterior nasal swab	-	-	Normal medical
203	F	67	anterior nasal swab	-	-	Normal medical
204	F	57	anterior nasal swab	+	-	COVID-19
205	F	61	anterior nasal swab	-	-	Normal medical
206	F	36	anterior nasal swab	-	-	Normal medical
207	M	51	anterior nasal swab	+	+	COVID-19
208	F	40	anterior nasal swab	-	-	Normal medical
209	F	32	anterior nasal swab	-	-	Normal medical
210	F	48	anterior nasal swab	+	+	COVID-19
211	F	76	anterior nasal swab	+	+	COVID-19