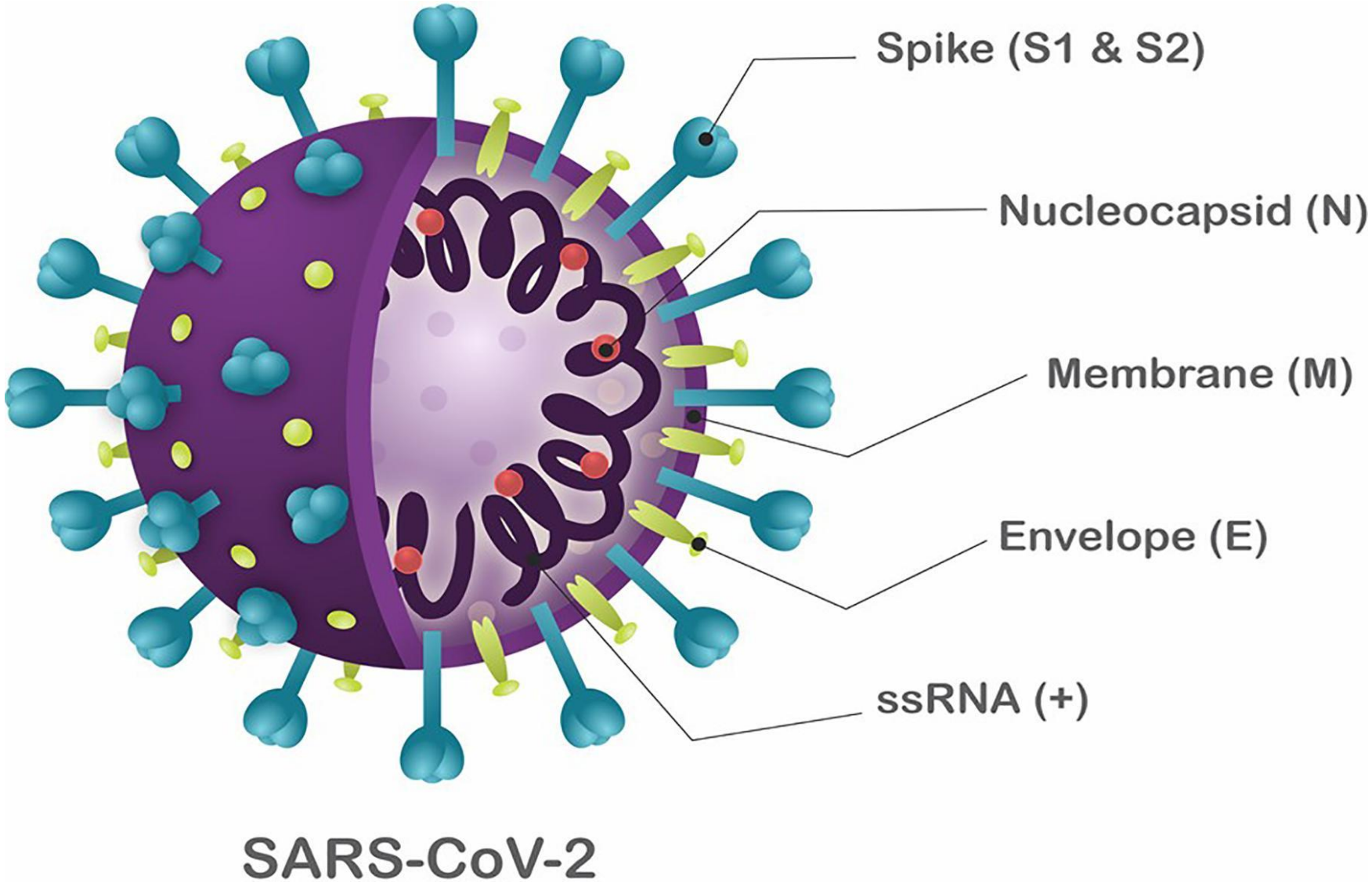
Development of an indexed score to identify the most suitable biological material to assess SARS-CoV-2

Marina Almeida-Silva, Renata Cervantes, Edna Ribeiro and Ana Marques-Ramos

H&TRC - Health and Technology Research Center

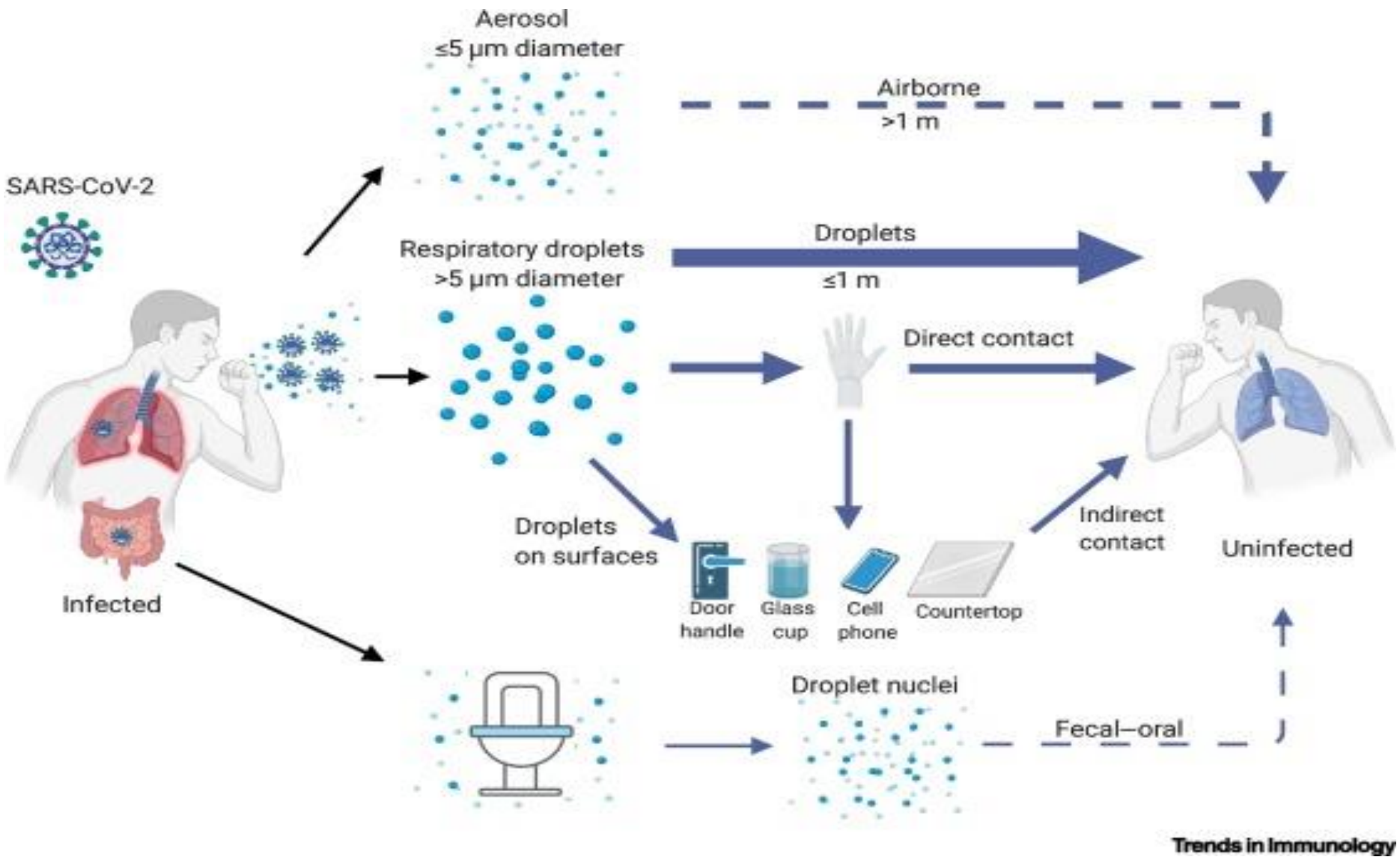
ISES – 19 to 21 March, Berlin, Germany

Schematic structure of SARS-CoV-2



Santos, I. A., et al (2020). Antivirals Against Coronaviruses: Candidate Drugs for SARS-CoV-2 Treatment?. *Frontiers in microbiology*, *11*, 1818.

SARS-CoV-2 - Transmission Routes



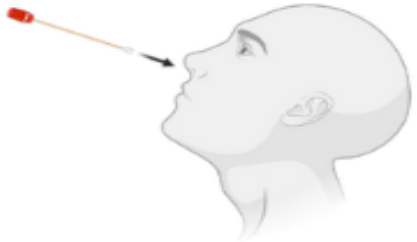
Harrison, A.G. et al, Mechanisms of SARS-CoV-2 Transmission and Pathogenesis, (2020)

SARS-CoV-2 - Testing

COVID-19 Diagnostic Test through PCR

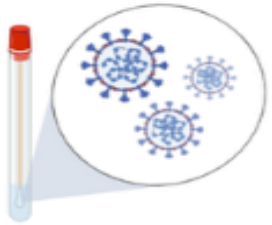
1 Nasopharyngeal swab <15 min

Cotton swab is inserted into nostril to absorb secretions.



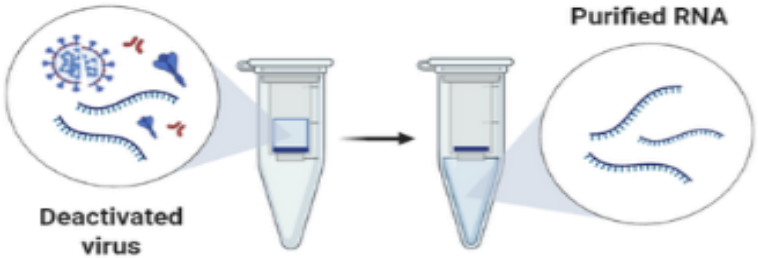
2 Collected specimen 0-72 h

Specimen is stored at 2-8°C for up to 72 hours or proceed to RNA extraction.



3 RNA extraction ~45 min

Purified RNA is extracted from deactivated virus.



4 RT-PCR ~1 h per primer set

Purified RNA is reverse transcribed to DNA and amplified by PCR.

Primers and probes for screening

E_Forward: ACAGGTACGTTAATAGTTAATAGCGT
E_Probe1: FAM-ACACTAGCCATCCTTACTGCGCTTCG-BBQ
E_Reverse: ATATTGCAGCAGTACGCACACA

E gene
First-line screening tool

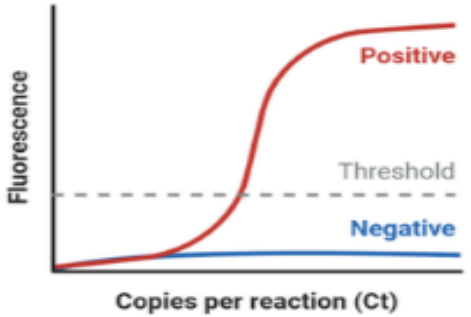
RdRp_Forward: GTGARATGGTCATGTGTGGCGG
RdRp_Probe1: FAM-CCAGGTGGWACRTCATCMGGTGATGC-BBQ
RdRp_Probe2: FAM-CAGGTGGAACCTCATCAGGAGATGC-BBQ
RdRp_Reverse: CARATGTTAAASACACTATTAGCATA

RdRp gene
Confirmatory testing

* N gene testing is not further used because it is slightly less sensitive.

5 Test results real-time

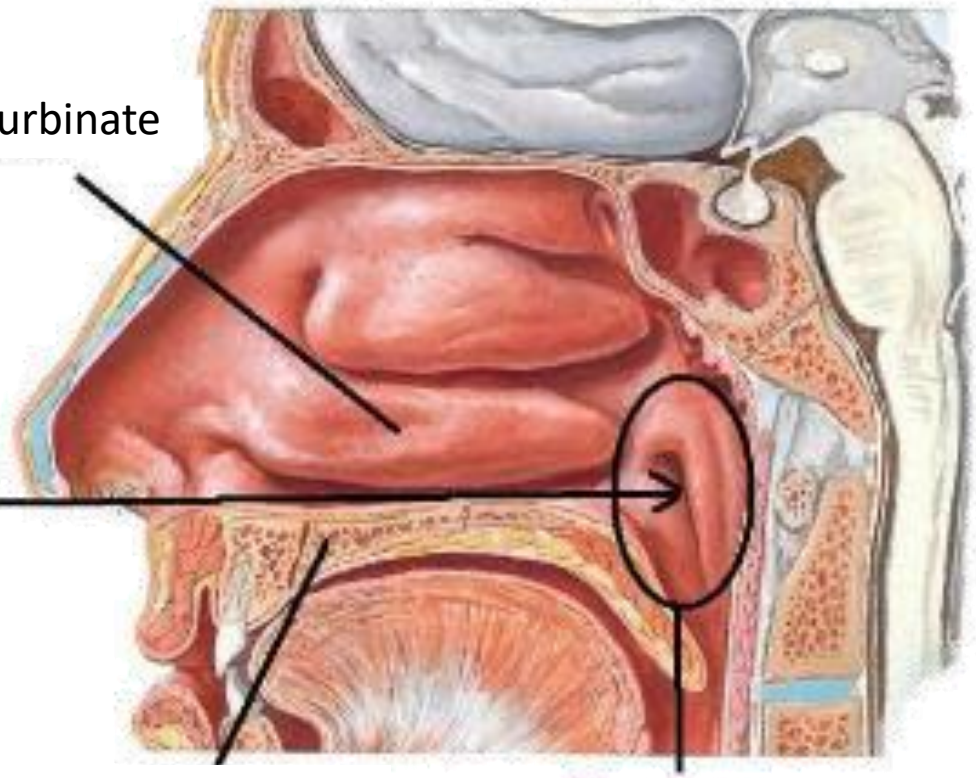
Positive SARS-CoV2 patients cross the threshold line within 40.00 cycles (< 40.00 Ct).



Brandeis Innovation, SARS-CoV-2 Testing: Demystifying the Terminology, (2021)

SARS-CoV-2 - Specimens

Nasopharyngeal swab
(NPS)



Oropharyngeal swab
(OPS)

Swab here in posterior oral pharynx

SARS-CoV-2 – GOALS

Development of an indexed score to identify the most suitable biological material to assess SARS-CoV-2

Assess the alternative methods through a review of existing literature

Specimen

Criteria

Score

SARS-CoV-2 – Alternative Specimens

A comprehensive scientific analysis was carried out to determine the types of samples used to diagnose SARS-CoV-2.

Urine

Sputum

Nasopharyngeal

Oropharyngeal

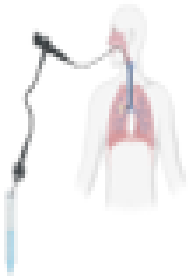
Bronchoalveolar

Salive

Faeces

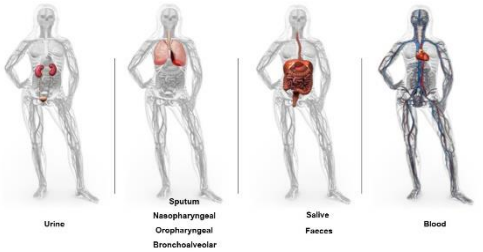
Blood

SARS-CoV-2 – Alternative Specimens

Sampling Methods								
Description	Urine	Sputum	Nasopharyngeal swab	Oropharyngeal swab	Bronchoalveolar Lavage	Saliva	Feces	Blood

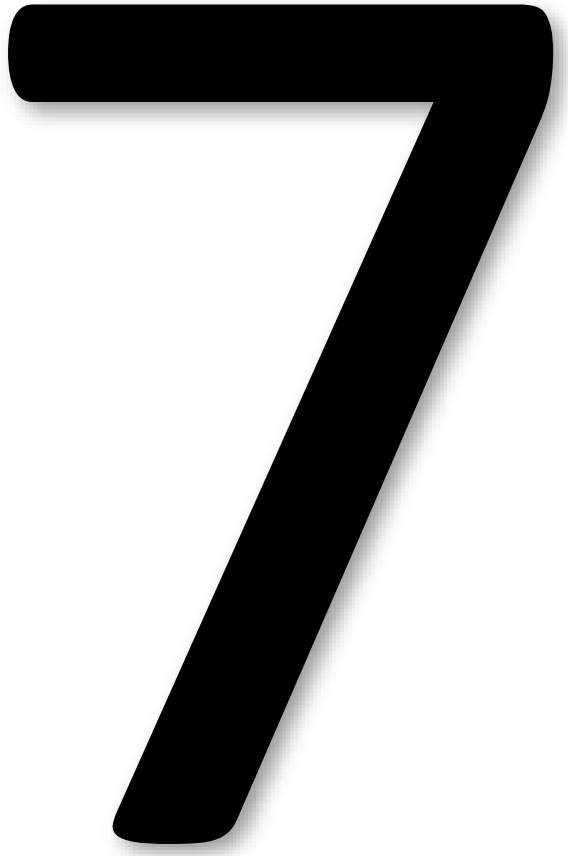
Based on scientific research, it was assumed that the sample for diagnostic examination for SARS-CoV-2 can be extracted from:

- Urinary System - urine
- Respiratory System - sputum, nasopharyngeal (NPSs), oropharyngeal (OPS), bronchoalveolar (BAL)
- Digestive System - saliva and faeces
- Circulatory System - blood

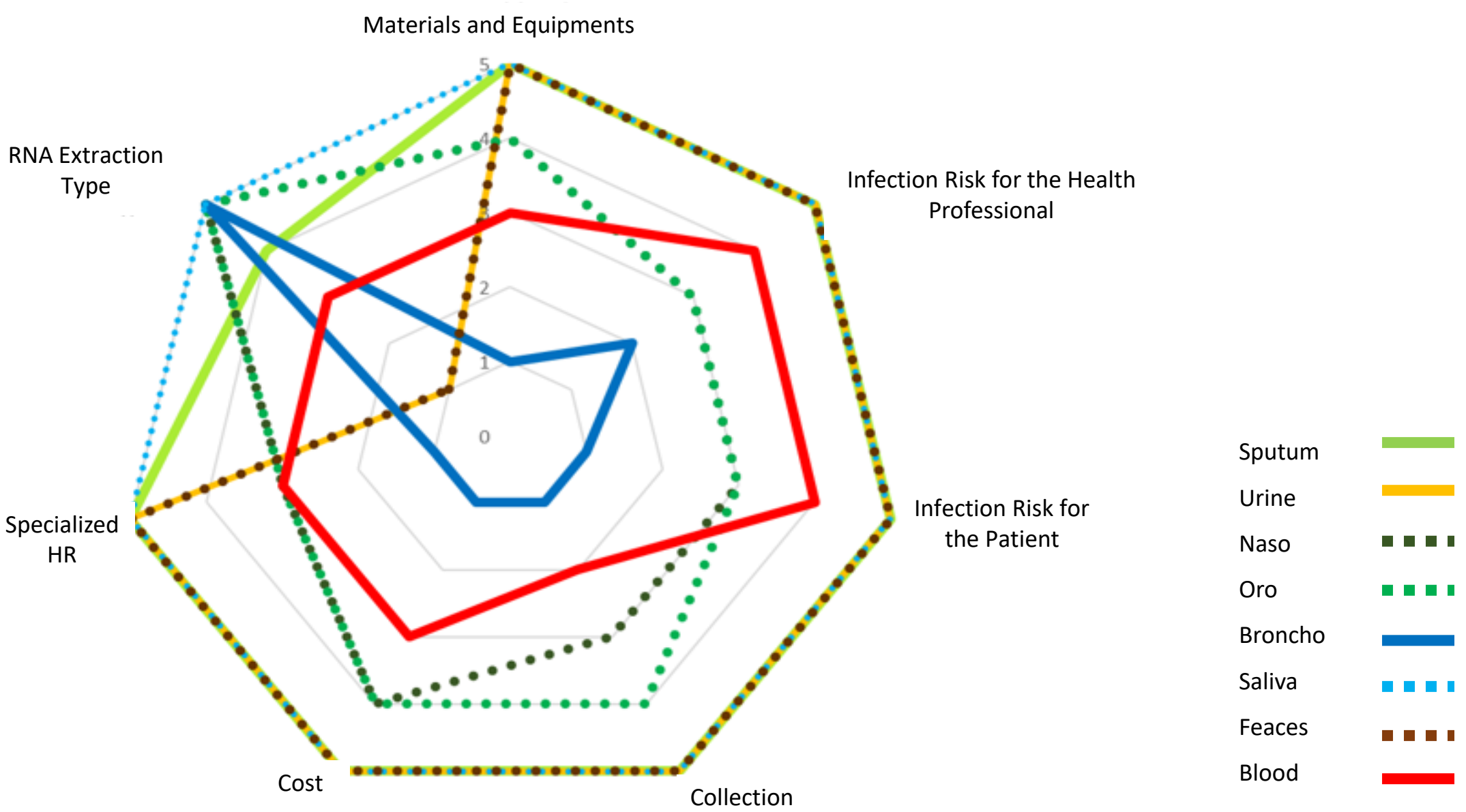


SARS-CoV-2 – Criteria

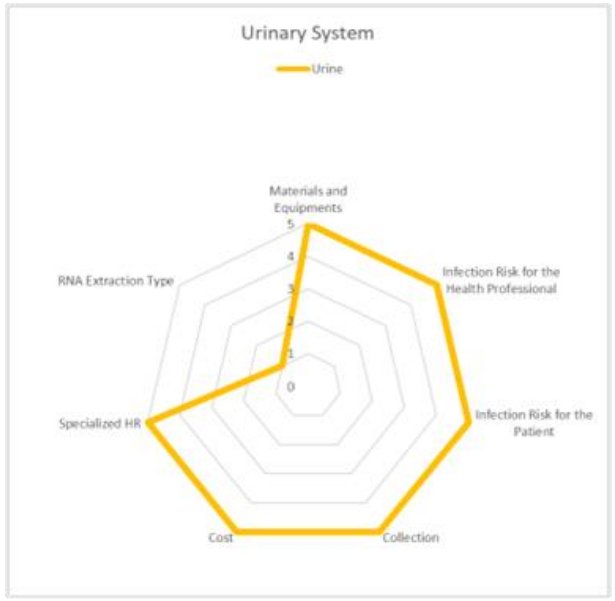
1. Materials and Equipment
2. Infection Risk for the Health Professional
3. Infection Risk for the Patient
4. Collection
5. Cost
6. Specialized HR
7. RNA Extraction Type $\bar{x}$

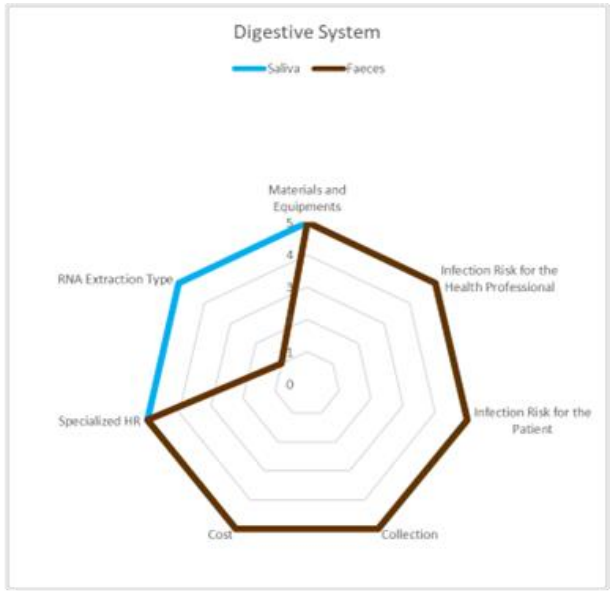


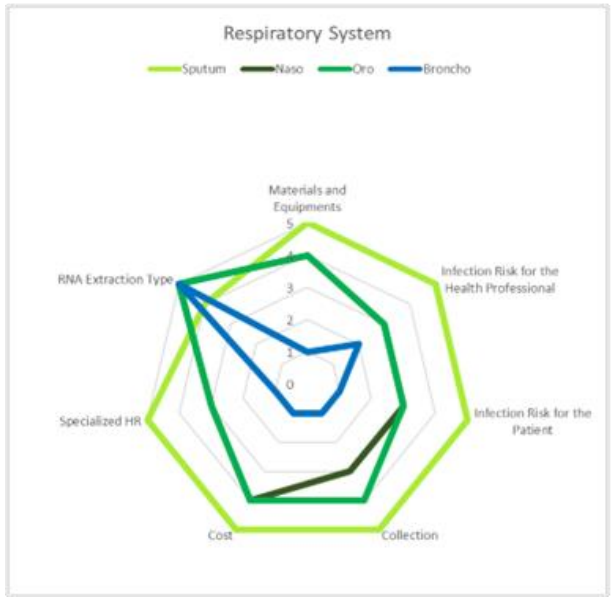
SARS-CoV-2 – Results / Score of the selected criteria according to each studied specimen

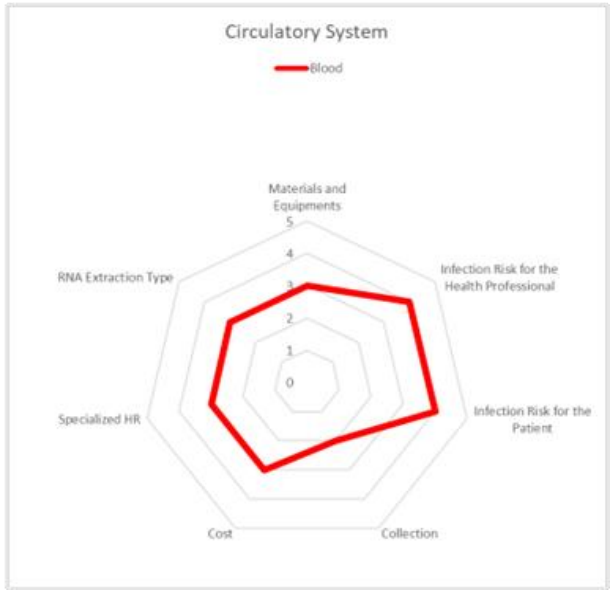


SARS-CoV-2 – Results / Score of the selected criteria according to each studied specimen









Sputum

Urine

Naso

Oro

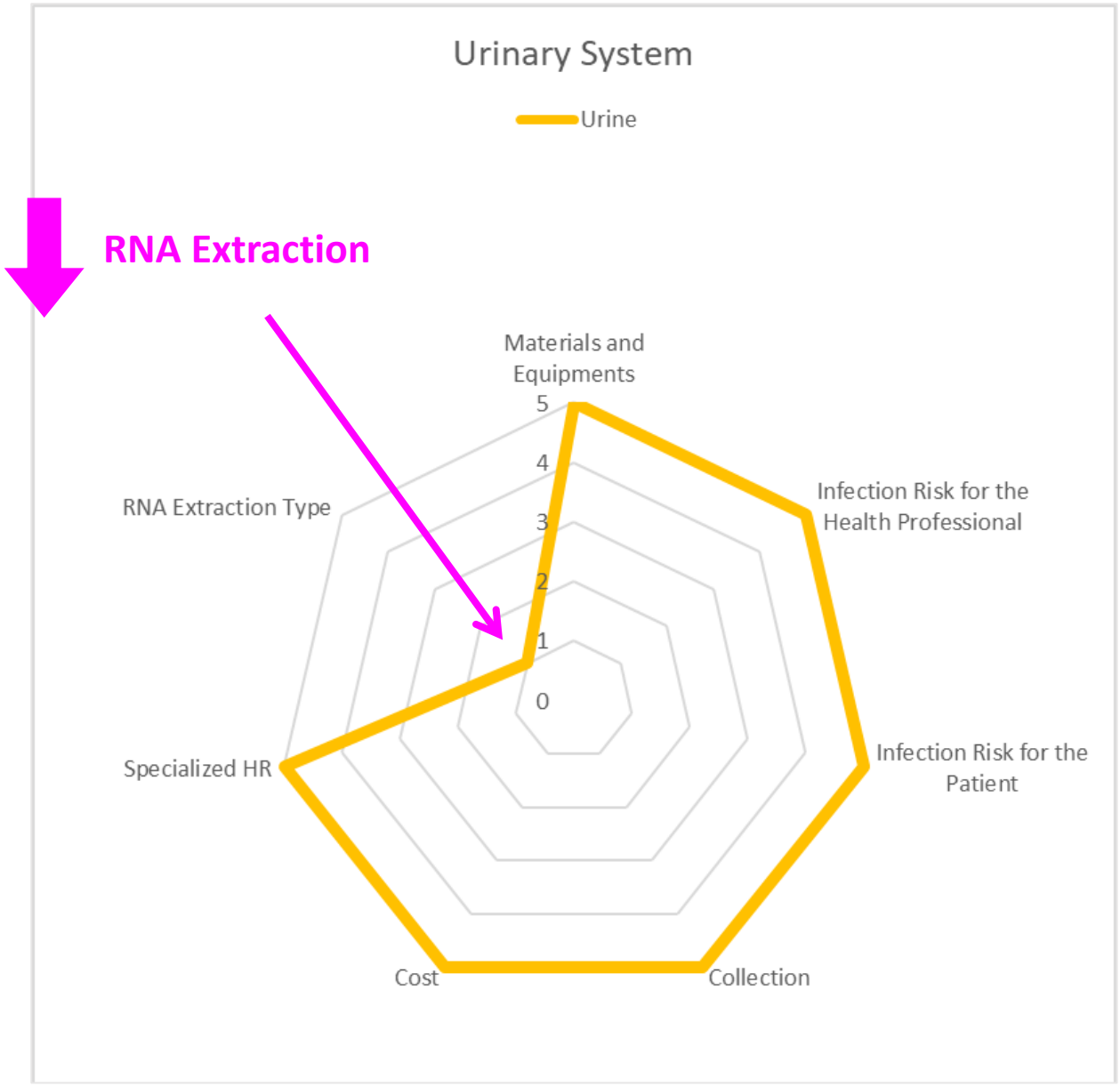
Broncho

Saliva

Feaces

Blood

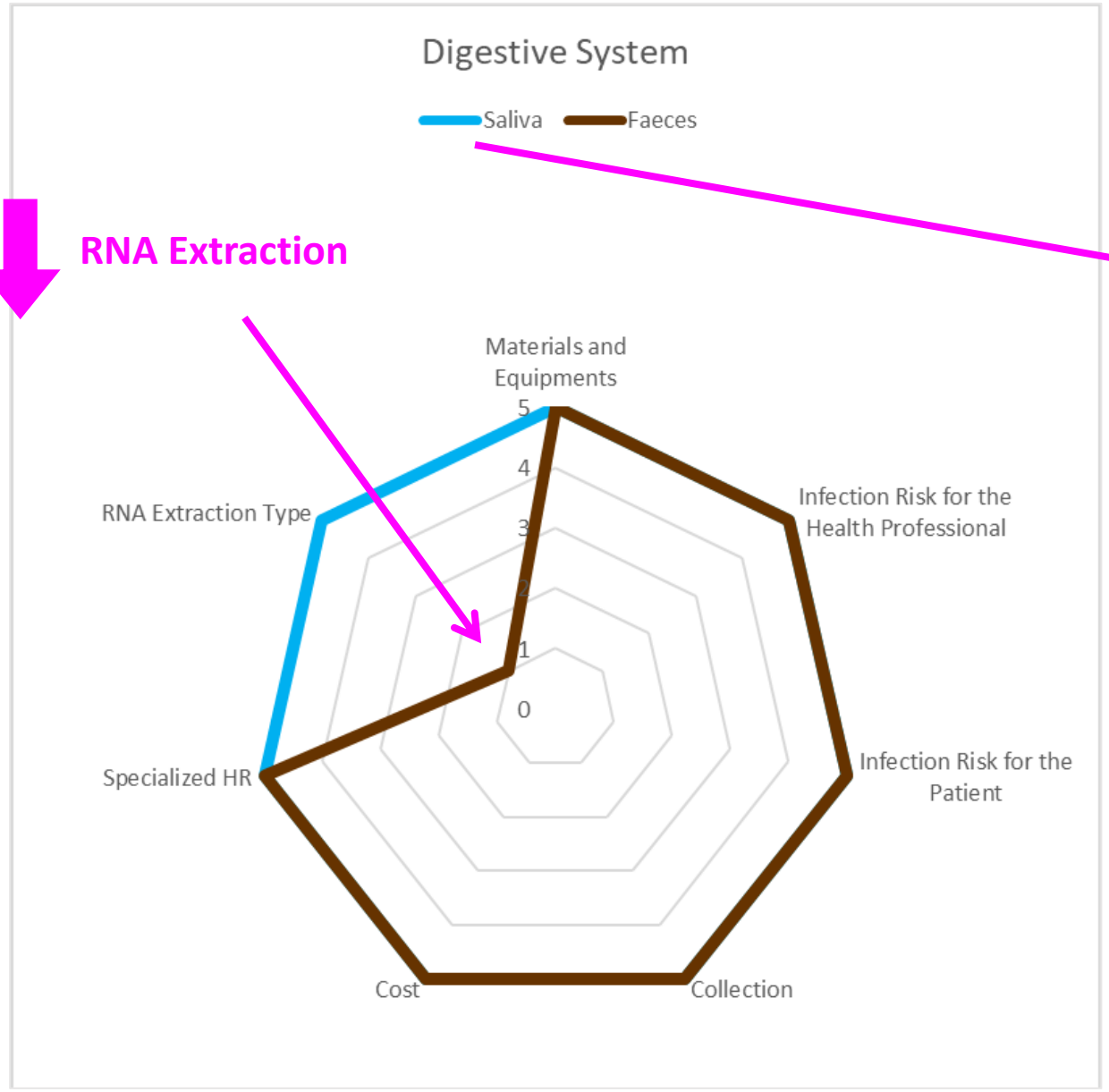
SARS-CoV-2 – Results / Score of the selected criteria according to each studied specimen



TOTAL SCORE: 4.4

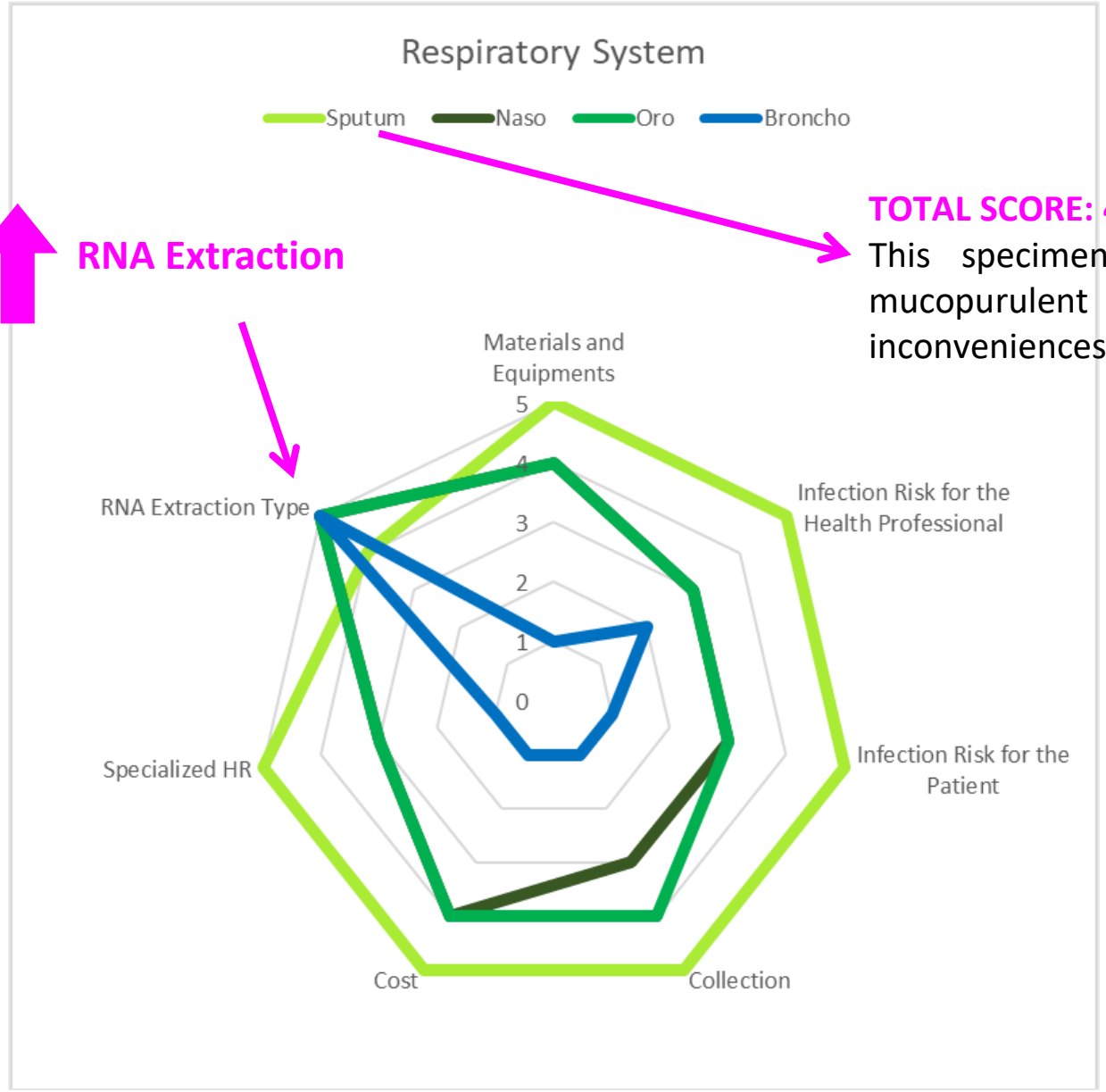
Sputum
Urine
Naso
Oro
Broncho
Saliva
Feaces
Blood

SARS-CoV-2 – Results / Score of the selected criteria according to each studied specimen



Sputum
Urine
Naso
Oro
Broncho
Saliva
Feaces
Blood

SARS-CoV-2 – Results / Score of the selected criteria according to each studied specimen

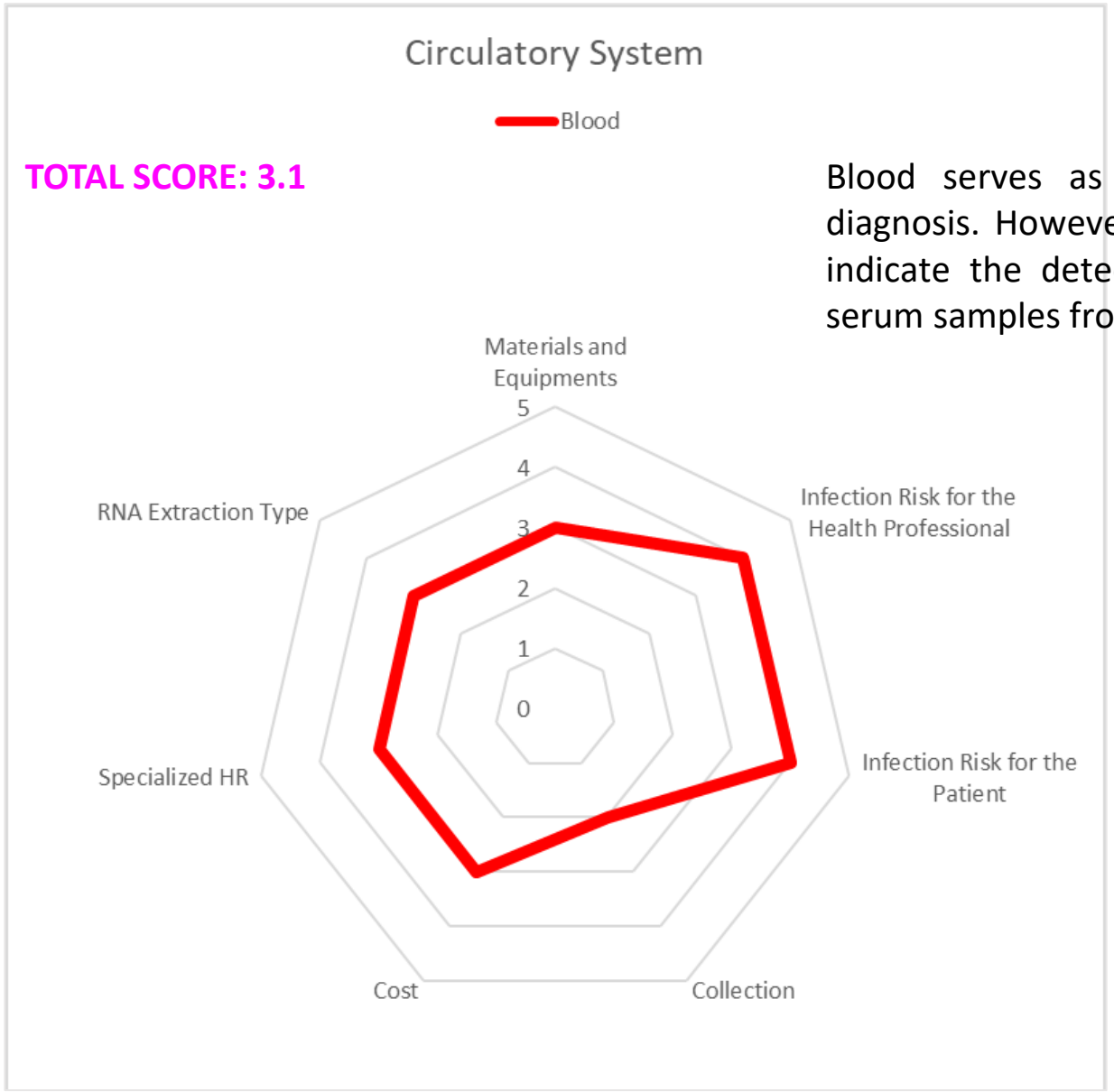


TOTAL SCORE: 4.9

This specimen frequently contains mucoid or mucopurulent material, thus causing some inconveniences for the RNA extraction process

- Sputum
- Urine
- Naso
- Oro
- Broncho
- Saliva
- Feaces
- Blood

SARS-CoV-2 – Results / Score of the selected criteria according to each studied specimen



TOTAL SCORE: 3.1

Blood serves as a viable specimen for COVID-19 diagnosis. However, only a limited number of studies indicate the detectability of viral RNA in plasma or serum samples from COVID-19 patients

Sputum
Urine
Naso
Oro
Broncho
Saliva
Feaces
Blood

SARS-CoV-2 – Conclusions

Saliva 5,0

Sputum 4,9

Faeces

Urine

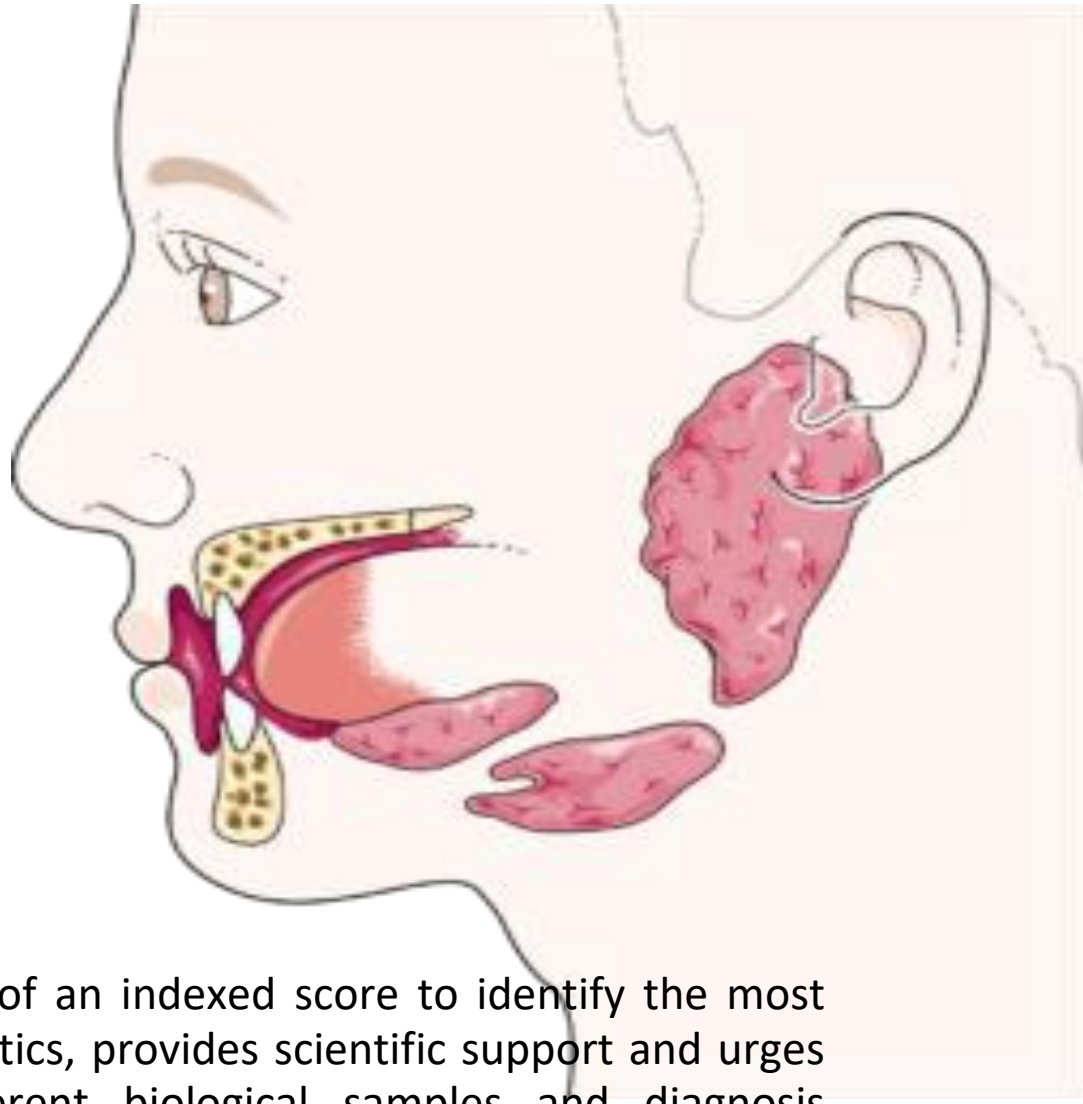
4,4

OPS 3,7

NPS 3,6

Blood 3,1

BAL 1,7



This comprehensive analysis allowed the development of an indexed score to identify the most suitable biological material to perform SARS-CoV-2 diagnostics, provides scientific support and urges the development of further studies focusing on different biological samples and diagnosis methodology, robustly supporting the decision-makers.

Stéphane Derruau et al. COVID-19 and Dentistry in72 Questions: An Overview of theLiterature.J. Clin. Med.2021

SARS-CoV-2 – Team

Ana Marques Ramos, Researcher

Edna Ribeiro, Researcher

Renata Cervantes, PhD Student

Rita Relvas, Environmental Health Student

Iara Fortunato, Biomedical Sciences Student

Corina Luchian, Biomedical Sciences Student

Inês Andrade, Environmental Health Student

Diana Narciso, Environmental Health Student

applied sciences

MDPI

Article

Development of an Indexed Score to Identify the Most Suitable Biological Material to Assess SARS-CoV-2

Marina Almeida-Silva [1,2,*], Renata Cervantes [1], Edna Ribeiro [1] and Ana Marques-Ramos [1]

[1] H&TRC—Health & Technology Research Center, ESTeSL—Escola Superior de Tecnologia da Saúde, Instituto Politécnico de Lisboa, 1990-096 Lisboa, Portugal; r.w.cervantes@gmail.com (R.C.); edna.ribeiro@estesl.ipl.pt (E.R.); ana.ramos@estesl.ipl.pt (A.M.-R.)

[2] OSEAN—Outermost Regions Sustainable Ecosystem for Entrepreneurship and Innovation, 9000-082 Funchal, Portugal

* Correspondence: marina.silva@estesl.ipl.pt

Marina Almeida-Silva | marina.silva@estesl.ipl.pt

H&TRC - Health and Technology Research Center

ISES – 19 to 21 March, Berlin, Germany