

Objectives

The primary objective of this study is to investigate if SARS-CoV-2 detection in saliva is comparable to nasopharyngeal (NP) and oropharyngeal (OP) swabs in both children and adults.

Secondary objectives of this study are:

- a) To compare viral load of SARS-CoV-2 in saliva, NP and OP swabs;
- b) To assess viral load of SARS-CoV-2 in saliva and feces over time, in relation to the course of disease;
- c) To investigate whether viral and bacterial co-infections as well as the composition of the upper respiratory tract microbiome (and mycobiome?) are associated with (severity of) COVID-19;
- d) To investigate transmission and viral load of SARS-CoV-2 in household members, and the course of potential COVID-19 disease in household members;
- e) To investigate whether patients and (potentially asymptomatic) household members develop immunity against SARS-CoV-2.

Design

	Cases (SARS-CoV-2 positive)	Household members of cases	Controls (SARS-CoV-2 negative)
Day 1	Saliva + NP + OP + feces + clinical data	Saliva + questionnaire	Saliva (+ NP + OP?)
Day 3	Saliva + feces + questionnaire/ clinical data	Saliva + questionnaire	
Day 5	Saliva + feces + questionnaire/ clinical data	Saliva + questionnaire	
Day 7	Saliva + feces + questionnaire/ clinical data	Saliva + feces (optional) + questionnaire	
Day 14	Saliva + feces + questionnaire/ clinical data	Saliva + questionnaire	
Day 30	Saliva + feces + questionnaire/ clinical data	Saliva + questionnaire	

Population

- Children and adults admitted to the hospital for suspected COVID-19.
 - Fever + cough/dyspnea (/abdominal symptoms for children)
- Sampling at home and interested in transmission to household members -> limit exclusion of adults to <55 years old
- Sample size: 10 SARS-CoV-2 positive adults & 10 SARS-CoV-2 positive children
 - How many negative controls needed?

Samples

- NP: for microbiome (& virus?) -> RNA protect/glycerol. Store in -80
- OP: for microbiome (& virus?) -> RNA protect/glycerol. Store in -80
- Saliva: for virus, bacterial co-detection, microbiome, antibodies?
 - Needs to be able to be frozen at -20 for at least a week
 - Especially not so great for antibodies? Maybe only collect EDTA sample at day 1 and day 30 -> pick up and freeze immediately and -80.
 - Media? RNA protect/glycerol & EDTA -> need 2 samples?
 - Volumes (1 ml for viral detection + 200 ul for bacterial detection (in total?) + 10 ul for antibody detection)
- <-> estimated total number of saliva samples ~400 -> do we have that number available?

Questionnaires

- 1 baseline
- Follow-up on respiratory symptoms in patient and household members. Daily up to 14 days or at the predefined timepoints?

Analysis

- Bacterial co-detection & Microbiome: compare to healthy controls??
 - Microbiome: PIENTER3?
 - Bacterial co-detection: ?

Plan

- Protocol -> send today to Sjoerd (epidemiologist), Steven, Marlies, Jacqueline ...
- Jacqueline will work on PIF and IC
- Emma will work on other required documents
- Submit Wednesday?