Instructions for saliva sampling using Salivette®

| Preparation | Prepare materials [see also detailed instructions on next page] ✓ Labelling Salivette® (labels for sample IDs, you may protect labels against wet conditions by wrapping tape around labels) ✓ Optional, please note weight of each Salicap® before and after saliva collection. This information is needed for the estimation of the salivary flow rate. |
|---|--|
| Prior collection | Instructions for participants prior saliva collection Do not collect saliva samples ✓ <1h after eating (no chewing gum within past 24h) ✓ <1h after brushing teeth ✓ <18h after smoking, drinking caffeine-containing beverages (coffee, tea, coke), juice, alcohol ✓ <24h after enganging in physical exercise |
| Optional | Recommended procedure in laboratory studies; somewhat difficult to implement in field studies ✓ In general: Rinsing of the oral cavity with water before saliva collection to get rid of food debris, neutralize pH of sample ✓ But: Avoid rinsing or water drinking immediately before collection (wait at least 10 min before collection) |
| CollectionImage: | Instructions for participants on how to collect saliva samples Open the Salivette® by snapping off the stopper an remove the swab (by using your mouth as you drink from the Salivette). Please move the swab gently in your mouth for 1-2 min but do not chew (You may use a timer of your phone to stop the minutes). Please transfer the swab with the absorbed saliva back to the Salivette® (do not touch the swab) and replace the stopper. |
| Storage | Storage (at participant's home or laboratory) ✓ Please store Salivette® at cool temperatures, ideally at -20°C, temporary storage in the refrigerator or freezer are fine (amylase & cortisol are relatively robust against repeated freeze-thaw cycles. Storage in the refrigerator for several days is fine.) |

Detailed instructions for preparing saliva samples

Preparation 1 (optional)



Preparation 2



Storage



Excel file



To note, ...



Further processing

 Note the weight of the Salivette® containing the collected saliva (after saliva collection) by using a documentary file (e.g., Word, Excel)

| Partipant ID | Weight | Session 1 T1 (pre) | Session 1 T2 (post) | Session 2 T1 (pre) | Session 2 T2 (post) | | | |
|-----------------|--------|-----------------------|------------------------|-----------------------|------------------------|--|--|--|
| | pre | | | | | | | |
| | post | | | | | | | |
| | pre | | | | | | | |
| | post | | | | | | | |

Numbering and sorting

- Please put Salivette® into a cryobox in the correct order, starting from the top left, following reading direction, and ending at bottom right
- Please number SaliCaps[®] by writing on the lid of each Salivette[®] (ascending numbering)
- Please label lid of the cyrobox by using study name and number of box and respective samples (e.g., "stress study" Box 1, samples 1-49)

Storage

 Please store Salivette® at cool temperatures, ideally at -20°C or colder (e.g., at -80°C), temporary storage in the refrigerator or freezer are fine (amylase & cortisol are relatively robust against repeated freeze-thaw cycles. Storage in the refrigerator for several days is fine.)

Excel file for analyses

 Please create an excel file, including the followoing columns: number on lid (ascending number), sample code (individual study code on Salivette® as well as analyte(s) of interest (e.g., cortisol, alpha-amylase) and additional comments

| # | code (Salicap) | cortisol | alpha-amylase | XX | comments |
|---|----------------|----------|---------------|----|----------|
| 1 | AB 01 01 A | | | | |
| 2 | AB 01 01 B | | | | |
| 3 | AB 01 02 A | | | | |
| | | | | | 1 |

Shipping [see also our SOP shipping biological samples]

✓ Overnight express, on dry ice or ice pack, please contact lab in advance for scheduling a convenient shipping date

Boxes

- ✓ Salicaps®: 130x130x50 (9x9 grid)
- ✓ Salivette®: 130x130x100 (7x7 grid)*

* We do not recommend Salivettes for the assessment of salivary alpha-amylase.