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Non-invasive sublingual glycocalyx measurements using GlycoCheck (RRID:SCR_018254)

Tracey L Weissgerber¹, Oscar Garcia Valencia²¹Berlin Institute of Health at Charité; ²University of Miami Miller School of Medicine, Jackson Memorial Hospital

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dx.doi.org/10.17504/protocols.io.be5ejg3e**Tracey Weissgerber**
Berlin Institute of Health at Charité

ABSTRACT

This protocol describes procedures for non-invasive measurements of sublingual vascular glycocalyx degradation using the GlycoCheck system (RRID:SCR_018254). This includes procedures for setting up the equipment, performing tests, saving data, shutting down the system, troubleshooting and removing dust from the camera lens.

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KEYWORDS

non-invasive, vascular glycocalyx, GlycoCheck, perfused boundary region, red blood cell filling, P50, glycocalyx, endothelial function, glycocalyx degradation

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GUIDELINES

Never leave or store the camera without a protective lens cover on.

MATERIALS TEXT

Glycocheck system for non-invasive glycocalyx measurements

Disposable tips (lens covers)

Paper towel

Picture of vessels underneath tongue

Mild disinfecting wipes to clean the camera handle

Lint free cotton swabs with long handles to clean lens (occasional use)

SAFETY WARNINGS

Carefully review all safety instructions that come with the camera before use.

Set-up (to be completed before the patient is tested)

- 1 Take out the computer and camera.
- 2 Plug camera into a USB port on the right-hand side of the computer
- 3 Plug Hard Disk Drive into a USB port on the right-hand side of the computer
- 4 Turn on the computer
- 5 Select Glycocheck User
- 6 Wait until "Process Scheduler" status is shown as "Idle" on the taskbar
- 7 While you are waiting:
 - a. Take the storage probe cover off and place in the camera spot of the suitcase
 - b. Open the end of the packaging for a new probe cover
 - c. Gently place the probe cover on the camera, leaving it in the packaging
- 8 Open Glycocheck-New program
- 9 Enter password
- 10 Wait for database synchronization to complete

- 11 On the calibration screen, select "Camera KK"
- 12 Click "Start Calibration"
- 13 Wait for "results: OK" message
- 14 Select "Add new patient" from the drop-down menu
 - a. Enter patient ID
 - b. Under birthday, enter test date
 - c. Under "Last name" enter first 3 letters of the last name
 - d. Under "First name" enter first 3 letters of the first name
 - e. Enter gender
 - f. Click "Add patient" to add the patient to the database
- 15 Click "Admission" to ensure that the patient always appears in the list on the right
- 16 Click on the battery charging signal in the lower right corner of the computer screen. Ensure that "High Performance" is selected

Testing a participant

- 17 Place a paper towel in front of the computer in case of drooling
- 18 Give instructions
 - 18.1 Show picture of blood vessels underneath the tongue – we are looking for big vessels at the back of the tongue, just to the right or left of the center
 - 18.2 Raise tongue slightly, place the camera under tongue just touching the surface (not pressing, no pressure)
 - 18.3 The camera flashes a bright green flashing LED light when turned on. To avoid discomfort, we will ask you to place the camera under your tongue before we turn it on.
 - 18.4 Demonstrate how to hold the camera – with two hands wrapped around base, forearms or arms

supported on desk

18.5 Tell the participant that you will focus the camera by turning the black knob; you will talk to them throughout the protocol to give them instructions as needed

18.6 The participant can use the paper towel if he or she starts drooling.

19 Make sure the correct participant is selected in the "Admissions" List

20 Ask participant to swallow to clear saliva (reduces air bubbles)

21 Wait until participant places camera under tongue

22 Press "Perform selected visit"

22.1 If the participant needs a break, press "Abort"; when the participant is ready to resume testing, press "Cancel"

22.2 Focus the camera once the subject finds a stable image; continue to focus throughout the test as needed, being careful not to move the camera

22.3 If you see a crystalline structure, the camera is focused on the plastic cover and not on the tongue. Turn the black knob a lot until you see vessels; then keep turning until flowing red blood cells are in focus

22.4 If the camera is very out of focus and video is being collected (green bar going up) despite the fact that no vessels are visible and in focus, wait until you have focused the camera; then stop the trial and start over. Do not use data from this trial.

22.5 We need several videos of different vessels, obtained from slightly different locations. Once the green bar goes up by 1/5 to 1/6 in a particular location, ask the participant to move the camera very slightly until a new set of vessels is visible; then hold while the camera records. Repeat this pattern of a stable period of recording followed by a small movement to find new vessels, until recording is completed. If the participant has a difficult time keeping the camera stable, this may not be needed

22.6 If you only see a few very small, looping vessels, encourage the participant to move the camera towards the back and center of the tongue

22.7 Remind the participant to breathe through his or her nose

22.8 If there is an air bubble, or the red air bubble warning light is on, the camera will not collect video (no change in height of green bar). Ask the participant to move the camera slightly until the air bubble is no longer in the image.

22.9 If cells are not flowing, are flowing slowly, or the direction of flow is changing, remind the participant to have the camera gently touching the tongue without pressing into the tongue.

22.10 If the 5:00 timer runs out, click "continue" to give the participant more time to complete the trial. Note that extra time was needed in the trial comments (see instructions below).

23 The program will stop automatically when data collection is complete. Take the camera and place the probe cover back in the protective sleeve.

24 Add your comments in the field that opens once the trial is completed

- a. Time of day
- b. Whether or not the patient is fasted; if not fasted, note time of last meal
- c. Whether or not the patient had caffeine in the six hours prior to the test
- d. Note if extra time was needed
- e. If the data should not be used, state this and list the reason why the data should not be used
- f. Note any other important factors that may affect the outcome of the test – stress, smoking, medications, position of the patient, etc.

25 Click "Next patient" – the program will start calculations after 30 seconds. Two trials can be done back to back by clicking perform new visit before the 30 second mark.

26 Go to "Patient Screen". This button will take a minute to appear if this is the first trial.

27 Wait for data to appear after calculations are complete (5 min). Do not unplug, shut down, remove camera or attempt to start a new trial while calculations are in progress.

28 Once the data appear, click "Home"

29 Perform 3 trials per participant by repeating steps 20-29

Saving the data

30 Go to "Patient Screen" to export summary data calculated by program

30.1 Click "Export Data"

30.2 Save on USB

30.3 Save patient test summary (patientreport.csv)

31 Go to "Measurement Screen" to export detailed data for each trial

31.1 Select the trial that you want to export

31.2 Click "Export data"

31.3 Save on USB

31.4 Adjust filename: add T1 to filename for trial 1, T2 to filename for trial 2, T3 to filename for trial 3

31.5 Repeat steps 31.1 to 31.4 until all trials are saved

Shutting down the camera

32 Never unplug, shut down or detach the camera until all calculations are complete. When all calculations are complete, close the Glycocheck program.

33 Shut down the computer

34 Detach the camera

35 Remove and discard the used probe cover; replace the storage probe cover. Never leave the camera unprotected without a probe cover.

- 36 Wipe the camera handle with a disinfecting wipe.
- 37 Place the camera in the box, carefully coiling the cord so that it fits in the cord space in the suitcase
- 38 Ensure that the computer battery is fully charged for the next trial
- 39 Place the battery in the lower part of the suitcase
- 40 Place the computer in the suitcase, with the tongue picture and patient schedule on top
- 41 Ensure that the camera cord is not sticking out, and will not be damaged when closing the suitcase

Troubleshooting

- 42 Message "Error Occurred Scheduler Service is not running"
 - 42.1 Right click on Process scheduler and select "Start Service"
 - 42.2 If that does not work open task manager and on the "Services" tab look for "gmt-pss" and make sure is running. If it is not then right-click it and select start service.
- 43 If the Glycocheck program gives an error and won't start or won't complete measurements, restart the computer.
- 44 If steps 42 and 43 don't work, email Glycocheck
- 45 Error message: Licsense is expired. Contact GlycoCheck and ask them to update the license.

Cleaning camera lens

46 If you see dark specks on the image, indicating that there is dust on the lens, use the following procedures to clean the lens.

46.1 Remove the protective camera cover

46.2 Remove the camera extension (long, pointed portion of the camera that goes under the participant's tongue)

46.3 Insert a lint free cotton swab with a very long handle into the lens and rub gently to remove dust

46.4 Replace the camera and put on a new lens cover

46.5 Run a test to determine whether the lens is now free of dust and debris. If not, repeat steps 45.1 to 45.4 until the lens is clean. When finished, replace the protective lens cover.