

## Video Article

# Diffuse Reflectance Spectroscopy: Getting the Capillary Refill Test Under One's Thumb

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## Abstract

The capillary refill test was introduced in 1947 to help estimate circulatory status in critically ill patients. Guidelines commonly state that refill should occur within 2 s after releasing 5 s of firm pressure (*e.g.*, by the physician's finger) in the normal healthy supine patient. A slower refill time indicates poor skin perfusion, which can be caused by conditions including sepsis, blood loss, hypoperfusion, and hypothermia. Since its introduction, the clinical usefulness of the test has been debated. Advocates point out its feasibility and simplicity and claim that it can indicate changes in vascular status earlier than changes in vital signs such as heart rate. Critics, on the other hand, stress that the lack of standardization in how the test is performed and the highly subjective nature of the naked eye assessment, as well as the test's susceptibility to ambient factors, markedly lowers the clinical value. The aim of the present work is to describe in detail the course of the refill event and to suggest potentially more objective and exact endpoint values for the capillary refill test using diffuse polarization spectroscopy.

## Video Link

The video component of this article can be found at <https://www.jove.com/video/56737/>

## Introduction

Assessment and triage of the critically ill patient centers on the classical vital signs blood pressure (BP), heart rate (HR), respiratory rate (RR), oxygen saturation, and body temperature<sup>1</sup>. Changes in these parameters appear relatively late in the course of circulatory deterioration. For instance, in hemorrhage, a decrease in BP will not occur until blood loss becomes moderate to severe<sup>2</sup>, and HR increase can also be an insensitive and unspecific marker<sup>3</sup>.

The capillary refill test (CR test) may offer an earlier indication of incipient circulatory collapse, as the refill time is believed to change prior to the vital signs as well as clinical appearance of cold, clammy, and mottled skin<sup>1,4,5</sup>. The capillary refill test is typically performed by application and then release of a firm blanching pressure to the skin with timing (in seconds) of the return of blood to the blanched area. According to guidelines, refill should occur within 2 seconds after release of 5 seconds of firm pressure (*e.g.*, by the physician's finger) in the normal healthy supine patient<sup>6</sup>. The rationale for the test is that a slower refill time would indicate poor skin perfusion, possible caused by one of a number of critical events such as sepsis, blood loss, acute heart failure, or hypothermia.

At present, there is no consensus on a state of the art method for performing the CR test<sup>6,7,8,9,10</sup>. Contentious issues include lack of standardization of the actual blanching maneuver and the dependence on subjective (*i.e.*, naked eye) assessments of the refill endpoint<sup>7,9,11</sup>. Furthermore, there are indications that gender influences CR time<sup>12,13</sup>. The temperature, both ambient and skin, is known to affect the capillary refill time, but to what extent is not clear. Lastly, the use of different measurement sites, peripheral or central, is probably a further cause of variability in results with few studies in this area<sup>14,15</sup>.

In the present work, we used an optical bioengineering system to record the course of return of blood and the subsequent hyperemic response seen during the CR test. The system utilizes diffuse polarization spectroscopy to quantify and describe, in more detail than possible with the naked eye, the time and course of the capillary refill. The system comprises a standard digital camera, fitted with an external light ring with 92 white LEDs, and specially developed software. The lens and the two polarization filters, attached orthogonally in front of the LEDs, block light that has been directly reflected from the skin surface allowing only light that has become depolarized in the tissue to reach the camera. This generates a "sub-epidermal" image of the tissue to a depth of approximately 0.5 mm. The image is divided into its color planes and the red and green content for every pixel is calculated, generating a value which corresponds to the tissue concentration of red blood cells<sup>16</sup>. In video mode, the temporal resolution of the system is 0.02 s.

## Protocol

The study described here followed the local ethic guidelines and was approved by the regional ethical review board in Linköping (permit number 2015/99-31).

### 1. Informed Consent and Screening

1. Obtain informed consent from the subject.
2. Screen according to inclusion/exclusion criteria.  
NOTE: Inclusion criteria were: (i) Healthy adult >18 years of age, (ii) Able to understand written and oral information, and (iii) Provides oral and written consent. Exclusion criteria were: (i) Unable or unwilling to provide informed consent, (ii) Ongoing skin conditions, (iii) Cardiovascular disease, (iv) Medication that can interfere with vascular function. Oral contraceptives are allowed. If the subject uses oral contraceptives this should be noted on the subject's protocol file, (v) Scar free and non-bruised skin of the forehead, (vi) No intake of caffeine or tobacco 2 h prior to the onset of the test, and (vii) No strenuous physical activity at least 2 h prior to onset of the test.

### 2. Acclimatization and Equipment Setup

1. Let the test subject acclimatize for at least 20 min in a supine position prior to the onset of the test.  
NOTE: Correct positioning of the subject and equipment is shown in supplemental figure 1.
2. **Set up the camera recording of the capillary refill tests using the designated software and digital camera (see Table of Materials).**
  1. Connect the USB cable to the camera and the computer and adjust camera settings to video mode.
  2. Turn on the camera and make sure that it connects to the computer.
  3. Start the remote-controlled camera software (see **Table of Materials**).
  4. Turn off ambient light and turn the computer screen away from the measurement area.

### 3. Data Acquisition

1. Click the "life view" (Sic) button and press "Start".
2. Switch between 'video' mode and 'TiVi' mode by clicking the radio buttons (this will not affect the recording, only the on-screen presentation). Ensure that a live video stream of the subject's forehead is visible on the display of the camera and on the computer screen.
3. Adjust the camera to a height of approximately 15 cm directly above the subject's forehead.
4. Make sure that the focus of the camera is set to "AF" (auto focus).
5. Ask the subject to keep his/her head still for the duration of the test and to refrain from talking. Inform the subject that it is OK to keep his/her eyes closed during the test by saying: "Please, place your head on this pillow and keep your head still for the duration of the test. Please, do not talk during the 20 seconds that the test will last. You may keep your eyes closed for the whole time if you find the light too bright".
6. Do not move the camera while recording.
7. In the "life view" window find and click "HD record" to start a continuous recording for 20 s. Note that the duration of the recording (seconds), can be seen on the camera display; the first 5 s of the recording is to acquire the normal capillary blood concentration of the forehead.
8. At the end of the 5 s of baseline recording, apply a firm blanching pressure to the measurement area for 5 s. Use the index finger with a plastic teaspoon between the finger and the forehead for pressure equalization and temperature insulation.
9. Release the pressure and quickly withdraw the examiner's finger and plastic teaspoon from the measurement area.
10. Continue recording for another 10 s to capture the subsequent hyperemic response.
11. Stop recording by clicking "Stop" in the "life view" window.
12. Once the recording has stopped, a window will open for saving and naming the file (in .mov format); choose a folder and name for the video. Do not disconnect the camera from the computer while the video is downloading to the designated folder on the computer.

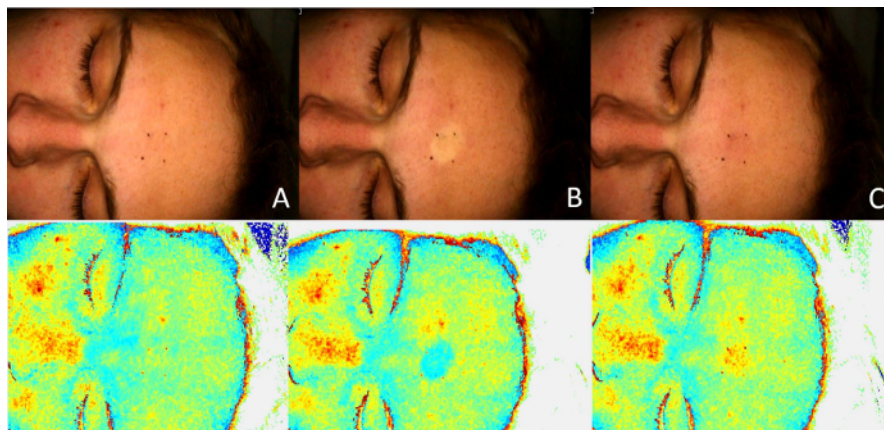
### 4. Data Analysis

1. Proceed to performing image analysis and curve construction with a dedicated analysis software (see **Table of Materials**).
2. Start the analysis software by double-clicking the desktop icon.
3. Once the software has started, on the main screen, select "Movie page."
4. In the "Movie page" window, click "Load movie." Wait for the software to upload and analyze the movie. This may take up to a couple of minutes depending on the size of the video file.
5. Once the upload and analysis of the movie is finished, click "Save as images" and wait for the software to analyze the video.  
NOTE: This will automatically create a new folder on the computer containing the video sequence divided into individual jpeg images.
6. Once the division of the video into individual jpeg images is finished, find and select "Crop images page" in the main screen window.
7. In the "Crop images page", click "First Photo" and navigate to the folder that contains the jpeg photos from the video and select the first photo to be analyzed; the software will automatically select the following photos of that batch.
8. Click the "Edit" menu and choose between circular or rectangular markers for the region of interest.
9. In the "Select Photos" window, find the "Actual photo" box and type in the name of the first photo in the batch where the investigator's finger is fully retracted from the measurement area. To find this photo, look through the folder containing the individual photos from the video.
10. Select a region of interest by clicking and holding the left mouse button and draw a circle or rectangle in the photo window that is within the borders of the blanched area. The selected region of interest is automatically applied to all photos of the batch.
11. Click the "File" menu and choose "Save ROI" to save the location of the selected region of interest for future reference.
12. Click the "Curve tracker" button and wait for the software to analyze the region of interest.

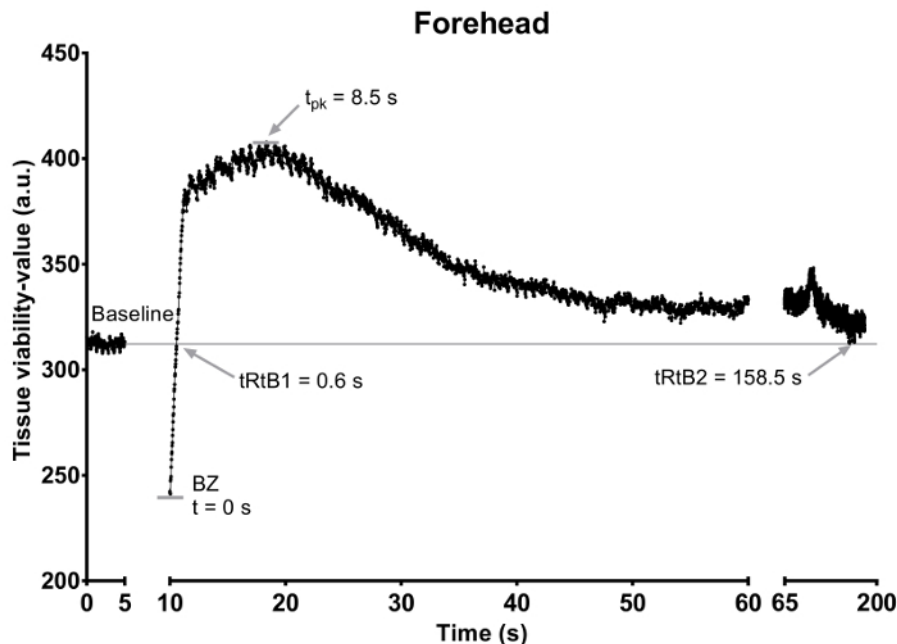
13. Once the curve tracker window opens, the change in red blood cell concentration for the selected region of interest for the duration of the test is displayed as a curve. The on-screen presentation can be changed by clicking the different radio buttons. This will not affect the raw data.
14. Click "Export data" and save the raw data transcript for further analysis.
15. In the raw data transcript, navigate to the column named "Mean intensity TiVi values." Generate three new columns next to this column for "image number," "total time," and "capillary refill time" starting at 1 s, 0 s, and 0 s and with an incrementation of 1 s, 0.02 s, and 0.02 s, respectively.  
NOTE: "Capillary refill time" correlates to the time of the subsequent reaction to the release of the blanching pressure and starts at the mean value that represents the first photo where the investigator's finger is fully retracted from the measurement area.
16. Delete the values between baseline and capillary refill time, as these only contain noise generated by the investigator's index finger and plastic teaspoon during the blanching maneuver.
17. Calculate a mean value for the baseline measurement (first 5 s of the recording).
18. Find the first value that equals or surpasses the calculated mean baseline value and note the time in the "Capillary refill time" column. This time point represents the "time to return to baseline 1."
19. Find the highest mean intensity value of the values generated after pressure release and note the time point in the "Capillary refill time" column. This time point represents the "Time to peak."
20. Find the second value that is less than or equal to the calculated mean value for the baseline measurement and note the time point in the "Capillary refill time" column. This time point represents the "time to return to baseline 2."

## Representative Results

Filming the course of the capillary refilling generates vast amounts of data not possible to obtain by naked eye assessment. We suggest here new endpoints to further improve the usability of the CR test as an early indicator of deterioration in circulatory status. We call these endpoints: "Baseline," "Blood Zero" (or "BZ"), "Time to Return to Baseline 1" (or "tRtB1"), "Time to Peak" or "Tpk." The "Baseline" value is derived by calculating a mean value of all the values obtained during the 5 s baseline measurement. "Blood Zero" is the mean value gained from the first image of the blanched area immediately after the pressure is released. The definition of "tRtB1" is the time, in seconds, after release of the blanching pressure until the value of the blanched area is equal or above the "baseline" value. "Tpk" corresponds to the time at which the highest value is recorded. **Figure 1** shows a selection of the regular photographs and color-coded images from a test performed on the forehead of a healthy male volunteer at room temperature. **Figure 2** shows a representative curve from a capillary refill test from the same test as described above. The measurement time in the depicted test was 3 min, a time not applicable in a clinical situation, but illustrating how long it takes before the value is back to the baseline value.



**Figure 1: Capillary refill response in a healthy volunteer.** A selection of regular photographs (upper row) and correlated color-coded images (lower row) from a capillary refill test performed on the forehead of a healthy male volunteer. (A) shows the forehead prior application of pressure, the blanched area immediately after release of pressure (B), and the hyperemic response (C). [Please click here to view a larger version of this figure.](#)



**Figure 2: Detailed profile of the capillary refill response with suggested endpoints.** The graph shows the course of the capillary refilling, as the change in red blood cell concentration over time, on the forehead of a healthy male volunteer and the new suggested endpoints generated by analysis of diffuse reflectance spectroscopy videos. The "Baseline" value is derived by calculating a mean value of all the values obtained during the 5 s baseline measurement. "Blood Zero" is the mean value gained from the first image of the blanched area immediately after the pressure is released. The definition of "tRtB1" is the time, in seconds, after release of the blanching pressure until the value of the blanched area is equal or above the "baseline" value. "Tpk" corresponds to the time at which the highest value is recorded. [Please click here to view a larger version of this figure.](#)

**Supplemental Figure 1: Measurement setup and positioning of subject.** The figure shows a representative setup for measurement of the CR response on the forehead in a resting, healthy subject. The polarization filters and lead light ring are mounted on a standard digital camera. The camera is attached to a stable tripod with a flexible, 3-way adjustable head allowing for correct positioning. Firm pressure is applied to the measurement area for 5 s by using a standard, plastic teaspoon. Please observe that ambient light should be dimmed to avoid interference with the measurement. [Please click here to download this file.](#)

## Discussion

In order to get the best results with the system, variability caused by environmental factors must be controlled. All ambient light must be turned off. The camera must be positioned in vertical alignment with the measurement area. In order to ensure a constant measurement area, subjects should not move or talk during measurement. For the same reason, the camera is preferably mounted on a stand to avoid movement and to maintain a constant distance to the measurement area. Test subjects should avoid caffeine<sup>17</sup>, tobacco<sup>18</sup>, and hard exercise<sup>19</sup> for at least two h prior to the test and rest for 20 min before the start of measurement, since these factors are known to affect microcirculation. Test subjects should be in a supine position with the measurement site at heart level to avoid positional redistribution of the blood volume. Room temperature and skin temperature should be monitored, as temperature is known to affect the refill time<sup>20,21</sup>.

There are other bioengineering alternatives to naked eye assessment and the presented technique which could be used to measure the CR test. Most of these techniques utilize changes in polarized or unpolarized light after reflection on the skin<sup>22,23,24</sup>. Other techniques measure the change in skin color using a video camera<sup>25</sup>, which is possibly most similar to the clinical situation. Further, indirect measures of blood flow may be achieved by correlating surface temperature to changes in dermal blood flow<sup>26</sup>. These alternative techniques are designed to measure a limited area of the skin or are for use only in one anatomical site (e.g., fingers)<sup>23,24,25</sup>.

With this new system, it is possible to switch between video and still photography and capture a large area, for instance a limb or even the whole body if necessary, with high temporal and spatial resolution. We consequently argue that this is an attractive technique for further physiological and pathophysiological characterization of the capillary refill response.

It should be noted that the camera system and the skin blanching maneuver described and used here are designed for research purposes and are not yet optimized for clinical use. To be fully usable as a method for monitoring critically ill patients, the system needs to be miniaturized and simplified. Ideally, the camera system should be integrated with a device that delivers a standardized blanching pressure and presents a physiologically relevant readout instantaneously. Although we are at an early stage of investigating the basic physiology of the CR response, we believe that most of these challenges can be managed by technological development.

## Disclosures

No financial support from WheelsBridge AB was involved in the conduct of the study. The author JH is employed by the Östergötland County Council but has a royalty agreement with WheelsBridge AB. The senior author CDA has a full-time academic position but also limited involvement in WheelsBridge AB.

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## References

1. Strehlow, M. C. Early identification of shock in critically ill patients. *Emerg Med Clin North Am.* **28** (1), 57-66, vii (2010).
2. Advanced Life Support, G. in *Acute Medical Emergencies*. 67-84 Wiley-Blackwell (2010).
3. Brasel, K. J., Guse, C., Gentilello, L. M., & Nirula, R. Heart rate: is it truly a vital sign? *J Trauma.* **62** (4), 812-817 (2007).
4. De Backer, D., Ortiz, J. A., & Salgado, D. Coupling microcirculation to systemic hemodynamics. *Curr Opin Crit Care.* **16** (3), 250-254 (2010).
5. Mrgan, M., Rytter, D., & Brabrand, M. Capillary refill time is a predictor of short-term mortality for adult patients admitted to a medical department: an observational cohort study. *Emerg Med J.* **31** (12), 954-958 (2014).
6. Advanced Life Support, G. in *Acute Medical Emergencies*. 55-65 Wiley-Blackwell (2010).
7. Lewin, J., & Maconochie, I. Capillary refill time in adults. *Emerg Med J.* **25** (6), 325-326 (2008).
8. Lobos, A. T., & Menon, K. A multidisciplinary survey on capillary refill time: Inconsistent performance and interpretation of a common clinical test. *Pediatr Crit Care Med.* **9** (4), 386-391 (2008).
9. Otieno, H. *et al.* Are bedside features of shock reproducible between different observers? *Arch Dis Child.* **89** (10), 977-979 (2004).
10. Raju, N. V., Maisels, M. J., Kring, E., & Schwarz-Warner, L. Capillary refill time in the hands and feet of normal newborn infants. *Clin Pediatr (Phila).* **38** (3), 139-144 (1999).
11. Klupp, N. L., & Keenan, A. M. An evaluation of the reliability and validity of capillary refill time test. *The Foot.* **17** (1 SRC - GoogleScholar), 15-20 (2007).
12. Gorelick, M. H., Shaw, K. N., & Baker, M. D. Effect of ambient temperature on capillary refill in healthy children. *Pediatrics.* **92** (5), 699-702 (1993).
13. Schriger, D. L., & Baraff, L. Defining normal capillary refill: variation with age, sex, and temperature. *Ann Emerg Med.* **17** (9), 932-935 (1988).
14. Pandey, A., & John, B. M. Capillary refill time. Is it time to fill the gaps? *Med J Armed Forces India.* **69** (1), 97-98 (2013).
15. Strozik, K. S., Pieper, C. H., & Roller, J. Capillary refilling time in newborn babies: normal values. *Arch Dis Child Fetal Neonatal Ed.* **76** (3), F193-196 (1997).
16. Doherty, J. *et al.* Sub-epidermal imaging using polarized light spectroscopy for assessment of skin microcirculation. *Skin Res Technol.* **13** (4), 472-484 (2007).
17. Noguchi, K. *et al.* Effect of caffeine contained in a cup of coffee on microvascular function in healthy subjects. *J Pharmacol Sci.* **127** (2), 217-222 (2015).
18. Reus, W. F., Robson, M. C., Zachary, L., & Heggors, J. P. Acute effects of tobacco smoking on blood flow in the cutaneous micro-circulation. *Br J Plast Surg.* **37** (2), 213-215 (1984).
19. Lenasi, H., & Strucl, M. Effect of regular physical training on cutaneous microvascular reactivity. *Med Sci Sports Exerc.* **36** (4), 606-612 (2004).
20. Anderson, B., Kelly, A. M., Kerr, D., Clooney, M., & Jolley, D. Impact of patient and environmental factors on capillary refill time in adults. *Am J Emerg Med.* **26** (1), 62-65 (2008).
21. John, R. T., Henricson, J., Nilsson, G. E., Wilhelms, D., & Anderson, C. D. Reflectance spectroscopy: to shed new light on the capillary refill test. *J Biophotonics.* (2017).
22. Blaxter, L. L. An automated quasi-continuous capillary refill timing device. *Physiological measurement.* **37** (1 SRC - GoogleScholar), 83-99. (2016).
23. Bordoley, A., Irwin, R., Kalyani, V., MacDonald, C., & Standish, D. *D1GIT: Automated, temperature-calibrated measurement of capillary refill time.* (2012).
24. Kviesis-Kipge, E., Curkste, E., Spigulis, J., & Eihvalde, L. Real-time analysis of skin capillary-refill processes using blue LED. *Proc. of SPIE.* **771523** (2010).
25. Shavit, I., Brant, R., Nijssen-Jordan, C., Galbraith, R., & Johnson, D. W. A novel imaging technique to measure capillary-refill time: improving diagnostic accuracy for dehydration in young children with gastroenteritis. *Pediatrics.* **118** (6), 2402-2408 (2006).
26. Cohen, J. *et al.* *Monitor to Measure Dermal Blood Flow in Critically Ill Patients: Study.* International Scholarly Research Notices, e578316 **578316** (2013 SRC - GoogleScholar) (2013).