Multi-speckle diffuse correlation spectroscopy to measure cerebral blood flow

K. MURALI and HARI M. VARMA

Department of Biosciences and Bioengineering, Indian Institute of Technology - Bombay (IITB), India

Abstract: We present a multi-speckle diffuse correlation spectroscopy (DCS) system for measuring cerebral blood flow in the healthy adult human brain. In contrast to the need for a high frame rate camera to measure the multi-speckle intensity auto-correlation, we employ a low frame rate camera to measure the auto-correlation using the recently introduced multi-step volterra integral method (MVIM). The results are validated by comparison against the blood flow measured using standard DCS system.

© 2020 Optical Society of America under the terms of the OSA Open Access Publishing Agreement

1. Introduction

Blood flow acts as an important biomarker as it is essential for the supply of oxygen to tissues and clearance of metabolic waste products from various parts of the body [1]. Specifically, cerebral blood flow (CBF) is known to be directly linked to the metabolic demand of oxygen supply [2,3], to neuro-vascular coupling [4,5] and metabolic response to functional stimuli [6,7]. Thus it is essential to study blood flow in general and CBF in specific as it is an indicator for well-being of the brain and can have important implications in the areas of neuroscience and rehabilitation engineering. Optical modalities usually employed to measure blood flow are laser doppler flowmetry (LDF), laser speckle contrast imaging (LSCI) and diffuse correlation spectroscopy (DCS). While LDF [8,9] and LSCI [10–12] are used to quantify surface blood flow (<1mm deep), DCS is employed to measure deep tissue blood flow [13,14].

DCS utilizes the speckle statistics of the scattered light measured at several spatially separated distances from the source using avalanche photodiodes (APD) or photo-multiplier tubes (PMT) [3,13,14]. One of the limitations of DCS is that it essentially collects the speckle data by independent single mode fibres, thus confining to a small detection site. To increase the signal to noise ratio (SNR) of the system, it is essential to collect multi-speckles with multiple detectors allowing parallel simultaneous detection, which would increase the cost of the system by several folds [15]. As an alternative to APD/PMT, speckle contrast based methods as in Ref. [16–19], combines the array detection as in LSCI and deep tissue imaging capability of DCS. Among these, speckle contrast optical spectroscopy (SCOS) uses multi-distance or multi-exposure speckle contrast to directly fit for deep tissue blood flow using correlation diffusion equation (CDE) [13,14].

With the advent of high-speed cameras, direct measurement of the intensity auto-correlation for quantifying surface and deep tissue blood flow were reported. In Ref. [20], the intensity auto-correlation is measured using a high frame rate camera (around 20000 frames per second (fps)) to quantify surface blood flow in mice brain and was used for selecting appropriate models for electric field auto-correlation in various types of tissues/ blood vessels. A microfluidic channel based platelet function testing system has been proposed in Ref. [21], where a camera of 1250 fps has been employed to distinguish different concentration of platelets from measured the intensity auto-correlation. In Ref. [22], the researches have employed a high frame rate camera (9250 fps) to measure deep tissue blood flow in mouse brain by measuring the speckle de-correlation time at a maximum source-detector (SD) separation of 3.2 mm. Here, the SD separation was limited to smaller range due to the fact that, for a larger SD separation, the auto-correlation

#401702 https://doi.org/10.1364/BOE.401702
Journal © 2020 Received 1 Jul 2020; revised 5 Oct 2020; accepted 6 Oct 2020; published 27 Oct 2020
When the tissue is illuminated with a coherent light source, the scattered light will form a speckle pattern due to the mutual interference at the detector [10,11]. The speckle contrast \( \kappa(r,T) \) and the normalized electric field auto-correlation \( (g_1(r,\tau)) \), associated with the time varying speckle pattern is related to each other as given by the following relation [28],

\[
\kappa^2(r,T) = \frac{2\beta}{T} \int_0^T (1 - \frac{\tau}{T}) |g_1(r,\tau)|^2 \, d\tau.
\]  

(1)

Here, \( r \) is the SD separation, \( T \) is the exposure time of the detector, \( \tau \) is the correlation delay time and \( \beta \) is the experimental constant accounting for the collection optics. The objective of MVIM is to recover \( g_1(r,\tau) \), for every \( \tau \), from the measurement \( \kappa(r,T) \), for every \( r \) and exposure times \( T \). A detailed analysis of MVIM algorithm is given in Refs. [26,29] and hence will be avoided for brevity. Nevertheless, we briefly present the essential features of MVIM as follows. The recovery of \( g_1(r,\tau) \) from \( \kappa(r,T) \), can be posed as a regularized least square minimization problem, where the function \( f(x) = \|Ax - b\|^2 + \lambda \|x - x_0\|^2 \) is minimized. Here \( A \) is the kernel matrix obtained by discretizing Eq. (1), \( x \) is defined to be \( g_1(r,\tau) \) and \( b \) is the multi exposure speckle contrast data \( \kappa(r,T) \). \( \lambda \) and \( x_0 \) are the pre-defined regularization parameter and apriori function respectively. The solution to the above minimization problem is given by \( x = (A^TA + \lambda I)^{-1}(A^Tb + \lambda x_0) \). Here, iterative scheme is employed to recover \( g_1 \), by using prior information based on the recovered \( g_1 \) from the previous iteration. The above mentioned MVIM algorithm can also be employed to recover the normalized intensity auto-correlation \( g_2(r,\tau) \), by discretizing the equation, \( (\kappa^2(r,T) + 1) = \frac{2}{T} \int_0^T (1 - \frac{\tau}{T})g_2(r,\tau)d\tau \), which is obtained from Eq. (1) using Siegert’s relation [14,28].

The Eq. (1) can be modified to relate changes in intensity auto-correlation \( \Delta g_2 \) and changes in speckle contrast \( \Delta \kappa \), as

\[
\Delta \kappa^2 = \kappa^2_p(T) - \kappa^2_b(T) = \frac{2}{T} \int_0^T (1 - \frac{\tau}{T})\Delta g_2 d\tau.
\]

Here, \( \kappa_b \) and \( \kappa_p \) denote...
the speckle contrast at baseline and perturbed blood flow respectively. Using MVIM, \( \Delta g_2(r, \tau) \) can be recovered from \( \Delta k^2(r, \tau) \) measurements, which can subsequently be related to the blood flow using the Taylor series expansion: 
\[
g_2(D_0^B + D_p^B) = g_2(D_0^B) + D_p^B \frac{\partial g_2}{\partial D_B}. \tag{2}
\]
Here, the \( D_0^B \) and \( D_p^B \) denote the baseline and the perturbed particle diffusion co-efficient, which are related to the blood flow. The Jacobian, \( J \), can be analytically expressed using the semi-infinite solution of CDE as,
\[
J \equiv \frac{\partial g_2}{\partial D_B} = \beta r_1 r_b b \Gamma \left( \frac{r_1 e^{-\sqrt{a + b D_B} \tau + r_1 e^{-\sqrt{a + b D_B} \tau r_1 + r_1 e^{-\sqrt{a + b D_B} \tau (r_1 + r_b)}}}}{\sqrt{a + b D_B} \tau (r_1 e^{-\sqrt{a \tau r_1} + r_1 e^{-\sqrt{a \tau}})^2} \right). \tag{2}
\]
Here, \( a = 3 \mu_a \mu_r \) and \( b = 6 a k_0^2 (\mu'_r)^2 \). The quantities \( \mu_a \), \( \mu_r \) and \( k_0 \) represent absorption coefficient, scattering coefficient and wave number respectively and \( \alpha \) represents the fraction of moving particles in the medium. The \( r_1 \) and \( r_b \) can be obtained by using the method of images as given in Ref. [14].

2.2. Experimental method

The experimental setup for the M-DCS system to measure cerebral blood flow is depicted in Fig. 1. A 785 nm laser source (L785P090, Thorlabs, USA) was focused to form a pointed source (using appropriate beam shaping optics that includes anamorphic prism, aspheric lens, focusing lens and a mirror to form a focused source of diameter approximately 1 mm) on the adult human forehead at FP1 (in the standard 10-20 EEG system), with a power of 15 mW at the skin surface as depicted in Fig. 1(b). A sCMOS camera (Prime BSI Photometric) was used to capture the scattered intensity at 2 cm away from the source at FP1 position. The detection was done by 50 \( \times \) 50 pixels in an area around 0.3 \( \text{mm}^2 \) (0.5mm \( \times \) 0.5mm), with an appropriate f-number to match the pixel to speckle ratio. The f-number of the objective lens (Navitar Zoom 7000) was calculated by matching the pixel size to speckle size, \( \rho_s \), using the relation \( \rho_s = 2.44 \lambda (1 + M f/\#) \) [11] which gives a value of \( f/\# = 2.5 \). The data was captured at five exposure times (10, 28, 50, 112, 215 microseconds) with 50 frames each exposure. The speckle contrast was computed as a ratio of the standard deviation to the mean of the intensity data detected by 125000 pixels (50 \( \times \) 50 \( \times \) 50) at a given exposure time. The temporal resolution of the system at this spatial resolution is 3.4 s, as it was the total time taken to capture the above-mentioned multi-exposure data. Concurrently, a DCS probe, consisting of single-mode fibre connected to an APD (SPCM-AQRH-14, Excelitas technologies) and a hardware correlator (correlator.com), was used to measure the intensity auto-correlation [13,14].

2.3. In vivo experimental protocol

Participants

Three healthy adults with mean age of 30.4±4.9 volunteered for the experimental validation. All participants provided informed consent before the study and the study was approved by the Institute ethical committee.

Baseline measurements

The participants were asked to lie in the supine position and the laser was focused to FP1 area above the frontal cortex, as in standard EEG system, and the scattered intensity data at multiple exposures was collected at 2 cm away from the source. The multi-exposure speckle contrast was computed from the measured intensity speckles and is fed to MVIM algorithm to recover \( g_2 \). Concurrently, a DCS probe was placed at the nearby region of interest (ROI), but with a same SD separation of 2 cm to measure the intensity auto-correlation. Both M-DCS and standard DCS data was collected for five trials on each participant.
Voluntary apnea task
The data acquisition follows the same procedure as done in baseline measurement with participants in supine position. Initially, the baseline readings were acquired for 1 minute. Subsequently, measurements were acquired for the cases where the participants were asked to hold their breath for 30 seconds after which they were asked to breathe normally for 2 minutes and it was considered as one trial. The study was carried out on three subjects with three trials per subject. Before the start of the study, one practise session was performed by all the participants.

Quantitative number processing task
The same data acquisition procedure as done in baseline measurement and voluntary apnea test was adopted here. After the baseline reading was acquired for 1 minute, a randomly generated three digit number (between 500 and 600) was given as an auditory cue and the participant was asked to subtract the number "seven" serially from the given three digit number for 1 minute. After one minute, a bell sound was given as an indication for the subject to stop the mental calculations. A post task reading was acquired for 1 minute, while continuing the measurements. The above procedure is denoted as one trial and we conduct three trials per participant and the whole process was repeated for three different participants. There was one practice trial before starting the experiment.

2.4. Data analysis and algorithm: implementation details
The multi-exposure speckle contrast was computed, as a ratio of standard deviation to the mean intensity, using 50 frames (with each frame of size 50 × 50 pixels) for given exposure and for 5 such exposures with appropriate noise corrections. MVIM algorithm was employed to calculate $g_1$ (or $g_2$) for the given SD separation of 2 cm and for $\tau$ ranging from $10^{-10}$s to 0.22s equally divided in logarithmic scale with 250 values. For comparing the $g_1$ obtained using M-DCS and standard DCS for the baseline blood flow, the experiment was repeated for 5 trials to compute the mean and the standard deviation at each $\tau$. Here, the prior values based on the solution of CDE was used for recovering $g_1$. The $g_1$ measured using both M-DCS and standard DCS was fitted against the semi-infinite solution of CDE model to quantify the blood flow $aD_B$.

To show that M-DCS works independent of the prior values, we have used four different types of priors and compared the results with each other. The different priors used were: (a) $x_0^0$ which is the semi-infinite medium solution of CDE [14]. Note that, for using CDE we need...
values of tissue optical properties such as \(\mu_a\) and \(\mu'_a\); (b) An arbitrary exponential model based prior \(x_0^0 = 1 + u\exp(-\tau)\) was used to recover \(g_2\), where \(u\) and \(v\) are the co-efficients of the fit. In this model, explicit values on tissue optical properties are not needed. Here, for the first iteration, the prior \(x_0^0 = 1 + \beta\) (based on the fact that \(g_2 \rightarrow (1 + \beta)\) at lower values of \(\tau\) was used and for the subsequent iterations, prior \(x_0^0\) was used to fit the result of the recovered \(g_2\) from the previous iteration; (c) The prior \(x_0^0\) was taken as the third type of prior to perform a comparative study to check the need of exponential model based priors in recovering \(g_2\) and rCBF; and (d) For recovering \(\Delta g_2\) from changes in speckle contrast, we have used prior of the form \(x_0^0 = u(\exp(-\tau v) - \exp(-w\tau))\), where \(u\), \(v\) and \(w\) are the co-efficients of the fit. In this case, for the first iteration, no prior (or \(x_0 = 0\)) was used and \(x_0^0\) was used as prior for subsequent iterations. The comparative study between recovered \(\Delta g_2\) with prior \(x_0^0\) and without prior (i.e. \(x_0 = 0\)) has also been performed, to check if M-DCS can work without using any prior values of \(x_0\).

For the voluntary apnea test, the field auto-correlation \(g_1\) was obtained using M-DCS and was fitted for semi-infinite medium solution of CDE to obtain the particle diffusion co-efficient \(D_b\) and subsequently relative cerebral blood flow (rCBF) \(= D_b^0 / D_b^0\). Here, the prior \(x_0^0\) was used. The capability of M-DCS to recover the intensity auto-correlation \(g_2\) is demonstrated in the quantitative number processing task. Here, the prior \(x_0^0\) was used and the recovered \(g_2\) was fitted to the semi-infinite solution of CDE to obtain the rCBF. By using the prior \(x_0^0\), we show that M-DCS does not require any explicit values about tissue optical properties (\(\mu_a\) and \(\mu'_a\)) for recovering \(g_2\). In addition, we also recover the change in intensity auto-correlation \(\Delta g_2\) associated with change in speckle contrast by the proposed M-DCS system. Here, for the first iteration, prior used is \(x_0 = 0\) (or no prior) and for subsequent iterations, \(x_0^0\) is used as prior. The rCBF was quantified by using jacobian, \(J\), calculated by using equation (2), wherein the absorption and scattering coefficients were assumed to be constants as 0.12 cm\(^{-1}\) and 12 cm\(^{-1}\) respectively [14,19]. The baseline blood flow \(D_b^0\) was assumed to be 2 × 10\(^{-5}\) cm\(^2\)/s and \(\tau\) was chosen at 4 \(\mu s\) for the calculation of Jacobian \(J\) and \(\Delta g_2\). The mean speckle contrast of first 17 s of the baseline was taken as \(\kappa_b\). We also compare the cases of recovering \(g_2\) from \(\kappa(T)\), by using two different priors (i.e. \(x_0^0\) and \(x_0^0\)), to show that no explicit values of tissue optical and dynamic properties (\(\mu_a\), \(\mu'_a\), and \(D_b^0\)) are required for M-DCS. The regularization parameter was chosen using L-curve approach [26,30] for all methods without iteration (when \(x_0^0\) was used as prior) and was linearly varied from 0.5 to 30 times the initial regularization parameter (determined by L-curve approach) for the methods with iteration. The maximum number of iterations were fixed to be 5 for all calculations in this paper.

3. Results

3.1. Baseline measurements

The field auto-correlation \((g_1)\) measured by M-DCS and standard DCS during baseline blood flow in the adult human head at a SD separation of 2 cm for 5 trials is shown in Fig. 2. The standard deviation (\(\sigma\)) of M-DCS measurements is also shown, where it is relatively smaller at lower values of \(\tau\), when compared to the standard deviation obtained using standard DCS measurements. We infer that this is due to the presence of CDE model as prior information in case of M-DCS. To verify it, the results were fitted for semi-infinite solution of CDE model \((g_1^0)\) and are shown in the inset. The corresponding standard deviation of the fitted \(g_1\) called \(\sigma_f\) is also plotted in the inset and it can be seen that \(\sigma_f\) of M-DCS and standard DCS are in reasonable agreement with each other. The mean \(D_b\) values (for a given subject) obtained for M-DCS was 7.42 × 10\(^{-9}\) cm\(^2\)/s and for standard DCS was 7.20 × 10\(^{-9}\) cm\(^2\)/s, which are comparable to one another.
3.2. Optimizing the number of pixels

The number of pixels needed to compute speckle contrast is determined by the overall signal to noise (SNR) of the camera pixels. The percentage error $E$ is the relative error between blood flow calculated by M-DCS against standard DCS. $E$ is plotted as a function of number of pixels used to compute speckle contrast, as shown in Fig. 3. It can be seen that around 8000 to 10000 pixels are needed to get $E$ less than 0.1%. The requirement of larger number of pixels (for smaller $E$) will demand more frames to be acquired, which in turn reduces the temporal resolution. Nevertheless, for this study, we have used a high density of 125000 pixels for computing the blood flow.

3.3. Voluntary apnea test and quantitative number processing task

The results of voluntary apnea task and quantitative number processing task obtained by M-DCS by recovering auto-correlation is given in Fig. 4. For the voluntary apnea task, the mean rCBF for three subjects with three trials each (nine trials in total) is plotted in Fig. 4(a). Figure 4(b) shows two representative $g_1$ curves for a given trial, plotted at two data points: one at ’a’ in the baseline and other at ’b’, after holding the breathe. The $g_1$ curve shifts to the left, with an increase in blood flow index of around 8% (on the given trial shown here), after holding the breathe for 30 s. The inset plot in Fig. 4(b), shows the standard deviation over five $g_1$ curves in a given trial during the baseline (near point ’a’) and after the task (near point ’b’). The changes in rCBF obtained for voluntary apnea test are comparable to that of results obtained in Ref. [19], where the change in
rCBF was around 12 to 15% and to that of Ref. [31] obtained using MRI, wherein similar trends of increase in total blood flow can be observed, after holding the breathe.

By using the proposed M-DCS system, the intensity auto-correlation was recovered for the quantitative number processing task and the corresponding rCBF is shown in Fig. 4(c) and (d). The mean and standard deviation of the rCBF for the nine trials (on three subjects) is shown in Fig. 4(c). The results shows an elevated blood flow during the task and recedes to the baseline after the task. The $g_2$ curves in a given trial for two data points, one in baseline and one during the task is shown in Fig. 4(d). It can be seen that the curve shifts to the left, with an increase in blood flow index of around 14% (on the given trial shown here). The inset plot in Fig. 4(d), shows the standard deviation over five $g_2$ curves in the given trial. The changes in rCBF obtained for quantitative number processing, also called serial subtraction task, are comparable to that of the results obtained in Ref. [32], where functional near infrared spectroscopy was used to measure changes in oxy and deoxy Haemoglobin (HbO and HbR) concentrations. The changes in HbD (difference in oxy and deoxy Haemoglobin concentrations) is highly correlated with rCBF [33] and similar trends on increase in HbD during the task and decrease in HbD after the task is observed in Ref. [32], which are comparable to rCBF obtained by M-DCS. To show that the problem of recovering both $g_1$ and $g_2$ can be implemented using MVIM, in Fig. 4(e) we have shown that both the $g_1$ and $g_2$ recovered using M-DCS (for a given trail during the baseline). By using the Siegert' relation, $g_2$ recovered by M-DCS is converted to $g_1$ and is plotted in Fig. 4(e) for comparison.

The above results use priors (either CDE or arbitrary exponential model $x_0^g$) to iteratively recover the auto-correlation from speckle contrast data. We have compared the results of $g_2$ recovered in quantitative number processing task (at the representative points 'a' and 'b' indicated in Fig. 4(c)), obtained in two cases: with and without using arbitrary exponential prior (i.e. by using $x_0^b$ and $x_0^g$ as prior respectively) in Fig. 4(f). Although in both the cases, $g_2$ is comparable, in case of not using exponential model based priors iteratively, we can see ripples at larger values of $\tau$ due to the presence of noise in speckle contrast data. This is due to the inherent ill-posed nature of the problem [26,27] and can be minimized by using aprior information (on $g_1$ or $g_2$, as seen Fig. 4(d)) and also by employing dense speckle contrast data (dense internms of exposure times). Nevertheless, the recovered $g_2$ without using exponential model based prior are reliable to measure the changes in blood flow rather than absolute blood flow. Figure 4(g) shows the correlation plot between mean rCBF (of the nine trials), with and without exponential model based priors. With a correlation co-efficient of $R = 0.86$, the results are in reasonable agreement with each other. This indicates that, even without any exponential model based priors, M-DCS can pickup changes in blood flow. We would like to reassert here, that regularization is essential for M-DCS due to the fact that recovering field/intensity auto-correlation from multi exposure speckle contrast data is an ill-posed problem [27].

The changes in $g_2$ was measured by the M-DCS system from the changes in speckle contrast, as explained in section 2.1. We compute $\Delta g_2 = g_2(D_2^B) - g_2(D_0^B)$ using the M-DCS system with multi exposure speckle contrast data. The recovered $\Delta g_2$ curves corresponding to the two representative points (in quantitative number processing task, as the one indicated in Fig. 4(c)) is shown in Fig. 5(a). During the task $\Delta g_2$ will decrease as shown in Fig. 5(a), which can be clearly deduced from Fig. 4(d). The Jacobian J was computed at $\tau = 4\mu s$ and the corresponding rCBF is shown in Fig. 5(b). It can be seen that rCBF obtained using $\Delta g_2$ is comparable to that of rCBF obtained by $g_2$ as shown in Fig. 5(b). In Fig. 5(a), we have shown the results of M-DCS based $\Delta g_2$ recovered with and without using prior values of $x_0$ (i.e by using $x_0^d$ and $x_0 = 0$ as priors respectively). We can see that $\Delta g_2$ curves are comparable to one another, at earlier values of $\tau$. This shows that we do not need any prior values of $x_0$ to measure changes in blood flow (not absolute blood flow) using M-DCS.
Fig. 4. The mean and standard deviation of rCBF during voluntary apnea test and quantitative number processing task for nine trials is shown in (a) and (c) respectively. The standard deviation is represented only on the upper side for better visualization of the plot to indicate the relative changes in mean rCBF. The solid vertical line in (a) and (c) indicate the beginning and end of the task. The data points 'a' and 'b' indicate the two representative points in the time-series: one during the baseline and the other after holding the breathe (in voluntary apnea task) or during the task (in quantitative number processing task). The representative $g_1$ and $g_2$ computed by M-DCS system for both the tasks at the above mentioned data points are plotted in (b) and (d) respectively. The shift in $g_1$ and $g_2$ towards the left indicates the increase in blood flow. (e) Comparison of $g_1$ and $g_2$ obtained using M-DCS for a given trail during baseline. The $g_1$ obtained from M-DCS and $g_1$ obtained via $g_2$ (obtained by using M-DCS) through Siegerts’ relation are comparable to one another. (f) The results of M-DCS based $g_2$, recovered by using exponential model as prior (i.e. $x_b^0$) is compared to that of the $g_2$ recovered without using exponential model as prior (i.e.$x_b^0$). It can be seen that curve shifts to the left in both the cases indicating that both the cases can pick up relative changes in the blood flow. The correlation plot between mean rCBF (of the nine trials) between the two cases (i.e. $rCBF_{NP}$, where no exponential prior was used and $rCBF_P$, where exponential prior was used) is shown in (g), indicating that the rCBF between both the cases are reasonably well correlated (Correlation coefficient R was 0.86, with slope of 1.05 and intercept of 5.28).
Fig. 5. (a) The representative $\Delta g_2$ curves at two time points 'a' and 'b' (for quantitative number processing task, as shown in Fig. 4(c)). It can be seen that as $\Delta g_2$ decreases, there is an increase in the blood flow. The $\Delta g_2$ curves obtained with and without using priors are comparable to one another, showing that M-DCS does not need any prior values of $x_0$ for measuring changes in blood flow. (b) Comparison of rCBF obtained by using $\Delta g_2$ to that of rCBF obtained by using $g_2$, indicating that rCBF is comparable to one another.

4. Discussion

A camera-based DCS system, termed as M-DCS, to measure deep tissue blood flow is presented. M-DCS utilizes multi-exposure multi-distance speckle contrast data to measure field / intensity auto-correlation using the recently introduced MVIM algorithm. We have successfully demonstrated M-DCS in measuring cerebral blood flow in the prefrontal cortex of adult human brain. In order to demonstrate M-DCS, we have measured the intensity / field auto-correlation associated with the blood flow in pre-frontal cortex of the healthy human volunteers as shown in Fig. 2. We validated this measurement by comparing it against a concurrently obtained auto-correlation data from the same region of interest using a standard DCS probe. A good agreement of the blood flow index ($\alpha D_B$) obtained by the two methods was observed as shown in inset of Fig. 2.

DCS system is quite often employed to measure blood flow changes rather than absolute blood flow. In order to initiate blood flow changes in pre-frontal cortex, we adopted two functional activation tasks namely, voluntary apnea task and quantitative number processing task. To show that both $g_1$ and $g_2$ can be measured using M-DCS, we have used one task as an example to illustrate the same. The $g_1$ and $g_2$ curves measured by M-DCS during the above experiments are plotted in Fig. 4(b) and Fig. 4(d) respectively, whereas Fig. 4(a) and Fig. 4(c) shows the mean and standard deviation of rCBF and are comparable to that of the results shown in previously published literatures [19, 31, 32]. We have reformulated MVIM algorithm to accommodate the changes in multi-exposure speckle contrast as given in section 2.1, to recover the changes in intensity auto-correlation ($\Delta g_2$). The rCBF obtained using $\Delta g_2$, as shown in Fig. 5(b) was also in good agreement with the previously reported data.

The major drawback of employing a multi-speckle DCS using cameras is the requirement of very high frame rate. The requirement of high frame rate increases as one attempts to probe deeper due to the fact that intensity speckles de-correlates faster. This problem is circumvented in M-DCS using the MVIM algorithm, where fastly de-correlating speckles can be quantified by using multi-exposure speckle contrast data, captured with a low frame rate camera. Additionally, this makes M-DCS a low cost, multi-speckle alternative to conventional DCS system.

The major limitation of M-DCS system is associated with MVIM algorithm. We have recently shown in Ref. [27] that the measurement of blood flow index by using standard DCS system and speckle contrast based systems (eg: SCOS, diffuse speckle contrast analysis (DSCA) [17] and M-DCS) are equivalent. However, it was also proved that recovery of field / intensity auto-correlation from multi-exposure speckle contrast is ill-posed and hence necessitates the
use of regularized inversion. In other words, the noise in the multi-exposure speckle contrast data will be amplified by the MVIM algorithm, while attempting to recover \( g_1 \) (or \( g_2 \)), if proper regularization scheme is not adopted. Any prior information used in the regularization scheme will make MVIM less ill-posed resulting in better recovery of \( g_1 \) or \( g_2 \). Hence we have studied the effect of apriori information in the M-DCS measurements as shown in Fig. 4(f) & (g) and Fig. 5(a).

Another limitation of the system is its current temporal resolution of 3.4 s. One of the reasons for low temporal resolution is the inability of the camera to adapt for the faster changes in exposure time through inbuilt camera software. This, in future, can be controlled by either hardware triggering or by controlling the duration of input light (through spatial light modulators or acousto-optic modulator modulators), as used in Ref. [12], resulting in better temporal resolution. Also, currently we use 125000 pixels for calculation of speckle contrast in a spatio-temporal manner. We have optimized the number of pixels needed for optimal estimate of blood flow to be around 10000 pixels. This high density data can be achieved by using a better objective wherein a single image of 100 x 100 pixels can be employed in the same ROI. With the above mentioned modifications, the temporal resolution of the M-DCS system can be enhanced. Currently, we have shown the working of the system in the human forehead to quantify the blood flow in prefrontal cortex. It can be extended to other parts of the brain, however, hair present may act as a hindering factor. To overcome this problem, we can use cameras coupled to optical fibers, as used Refs. [34,35].

**Funding**

Indian Institute of Technology Bombay (Seed grant); Department of Science and Technology, Ministry of Science and Technology, India (SERB–Early career research award); Department of Biotechnology, Ministry of Science and Technology, India (Ramalingaswamy Fellowship-2016).

**Acknowledgement**

Portions of this work were presented at the Biophotonics Congress: Biomedical Optics 2020 conference titled ‘A low frame rate camera based Diffuse Correlation Spectroscopy (DCS) system to measure blood flow in human adult brain’ (STu4D.7).

**Disclosures**

The authors declare that there are no conflicts of interest related to this article.

**References**

1. A. Zauner, W. P. Daugherty, M. R. Bullock, and D. S. Warner, “Brain oxygenation and energy metabolism: part i-biological function and pathophysiology,” Neurosurgery 51(2), 289–302 (2002).
2. K. Uludag, D. J. Dubowitz, E. J. Yoder, K. Restom, T. T. Liu, and R. B. Buxton, “Coupling of cerebral blood flow and oxygen consumption during physiological activation and deactivation measured with fMRI,” NeuroImage 23(1), 148–155 (2004).
3. T. Durduran and A. G. Yodh, “Diffuse correlation spectroscopy for non-invasive, micro-vascular cerebral blood flow measurement,” NeuroImage 85, 51–63 (2014).
4. A. Viltjener and U. Dirnagl, “Coupling of brain activity and cerebral blood flow: basis of functional neuroimaging,” Cerebrovasc. Brain Metab. Rev. 7, 240–276 (1995).
5. A. Devor, S. Sakadžić, V. J. Srivivasan, M. A. Yaseen, K. Nizar, P. A. Saisan, P. Tian, A. M. Dale, S. A. Vinogradov, M. A. Franceschini, and D. A. Boas, “Frontiers in optical imaging of cerebral blood flow and metabolism,” J. Cereb. Blood Flow Metab. 32(7), 1259–1276 (2012).
6. C. Cheung, J. P. Culver, K. Takahashi, J. H. Greenberg, and A. Yodh, “In vivo cerebrovascular measurement combining diffuse near-infrared absorption and correlation spectroscopies,” Phys. Med. Biol. 46(8), 2053–2065 (2001).
7. V. Quaresima, S. Bisconti, and M. Ferrari, “A brief review on the use of functional near-infrared spectroscopy (FNIRS) for language imaging studies in human newborns and adults,” Brain and language 121(2), 79–89 (2012).
8. V. Rajan, B. Varghese, T. G. van Leeuwen, and W. Steenbergen, “Review of methodological developments in laser doppler flowmetry,” Lasers Med. Sci. 24(2), 269–283 (2009).
9. C. Riva, B. Ross, and G. B. Benedek, “Laser Doppler measurements of blood flow in capillary tubes and retinal arteries,” Invest. Ophthalmol. Vis. Sci. 11, 936–944 (1972).
10. J. Briers and A. Fercher, “Retinal blood-flow visualization by means of laser speckle photography,” Invest. Ophthalmol. Vis. Sci. 22, 255–259 (1982).
11. D. A. Boas and A. K. Dunn, “Laser speckle contrast imaging in biomedical optics,” J. Biomed. Opt. 15(1), 011109 (2010).
12. A. B. Parthasarathy, W. J. Tom, A. Gopal, X. Zhang, and A. K. Dunn, “Robust flow measurement with multi-exposure speckle imaging,” Opt. Express 16(3), 1975–1989 (2008).
13. D. A. Boas, L. Campbell, and A. G. Yodh, “Scattering and imaging with diffusing temporal field correlations,” Phys. Rev. Lett. 75(9), 1855–1858 (1995).
14. T. Durduran, R. Choe, W. Baker, and A. G. Yodh, “Diffuse optics for tissue monitoring and tomography,” Rep. Prog. Phys. 73(7), 076701 (2010).
15. G. Dietische, M. Ninkc, C. Ortolf, J. Li, F. Jaillon, and T. Gisler, “Fiber-based multispeckle detection for time-resolved diffusing-wave spectroscopy: characterization and application to blood flow detection in deep tissue,” Appl. Opt. 46(35), 8506–8514 (2007).
16. C. P. Valdès, H. M. Varma, A. K. Kristoffersen, T. Dragojević, J. P. Culver, and T. Durduran, “Speckle contrast optical spectroscopy, a non-invasive, diffuse optical method for measuring microvascular blood flow in tissue,” Biomed. Opt. Express 5(8), 2769–2784 (2014).
17. R. Bi, J. Dong, and K. Lee, “Deep tissue flowmetry based on diffuse speckle contrast analysis,” Opt. Lett. 38(9), 1401–1403 (2013).
18. C. Huang, D. Irwin, Y. Lin, Y. Shang, L. He, W. Kong, J. Luo, and G. Yu, “Speckle contrast diffuse correlation tomography of complex turbid medium flow,” Med. Phys. 42(7), 4000–4006 (2015).
19. T. Dragojević, J. L. Hoffmann, D. Tamborini, D. Portaluppi, M. Buttavafa, J. P. Culver, F. Villa, and T. Durduran, “Compact, multi-exposure speckle optical spectroscopy (scos) device for measuring deep tissue blood flow,” Biomed. Opt. Express 9(1), 322–334 (2018).
20. D. D. Postnov, J. Tang, S. E. Erdener, K. Kılıç, and D. A. Boas, Dynamic laser speckle imaging, BioRxiv p. 626515 (2019).
21. H.-J. Jeon, M. M. Qureshi, S. Y. Lee, J. D. Badadhe, H. Cho, and E. Chung, “Laser speckle decorrelation time-based platelet function testing in microfluidic devices,” Sci. Rep. 9(1), 16514 (2019).
22. M. M. Qureshi, J. Brake, H.-J. Jeon, H. Ruan, Y. Liu, A. M. Safi, T. J. Eom, C. Yang, and E. Chung, “In vivo study of optical speckle decorrelation time across depths in the mouse brain,” Biomed. Opt. Express 8(11), 4855–4864 (2017).
23. W. Zhou, O. Kholiqov, S. P. Chong, and V. J. Srinivasan, “Highly parallel, interferometric diffusing wave spectroscopy for monitoring cerebral blood flow dynamics,” Optica 5(5), 518–527 (2018).
24. J. D. Johansson, D. Portaluppi, M. Buttavafa, and F. Villa, “A multipixel diffuse correlation spectroscopy system based on a single photon avalanche diode array,” J. Biophotonics 12(11), e201900091 (2019).
25. W. Liu, R. Qian, S. Xu, P. C. Konda, and R. Horstmeyer, “Fast sensitive diffuse correlation spectroscopy with a spad array,” in Optical Tomography and Spectroscopy, (Optical Society of America, 2020), pp. SM3D–3.
26. K. Murali, A. Nadakumaran, T. Durduran, and H. M. Varma, “Recovery of the diffuse correlation spectroscopy data-type from speckle contrast measurements: towards low-cost, deep-tissue blood flow measurements,” Biomed. Opt. Express 10(10), 5395–5413 (2019).
27. K. Murali, A. Nadakumaran, and H. M. Varma, “On the equivalence of speckle contrast-based and diffuse correlation spectroscopy methods in measuring in vivo blood flow,” Opt. Lett. 45(14), 3993 (2020).
28. R. Bandypadhyay, A. Gittings, S. Suh, P. Dixon, and D. J. Durian, “Speckle-visibility spectroscopy: A tool to study time-varying dynamics,” Rev. Sci. Instrum. 76(9), 093110 (2005).
29. C. Andrade, N. B. Franco, and S. McKee, “Convergence of linear multistep methods for volterra first kind equations with k(0) = 0,” Computing 27(3), 189–204 (1981).
30. P. C. Hansen and D. P. O’Leary, “The use of the l-curve in the regularization of discrete ill-posed problems,” SIAM J. Sci. Comput. 14(6), 1487–1503 (1993).
31. Z. B. Rodgers, V. Jain, E. K. Englund, M. C. Langham, and F. W. Wehrli, “High temporal resolution mri quantification of global cerebral metabolic rate of oxygen consumption in response to apneic challenge,” J. Cereb. Blood Flow Metab. 33(10), 1514–1522 (2013).
32. C. Hock, F. Müller-Spahn, Schuh-Hofer, M. Hofmann, U. Durnagl, and A. Villringer, “Age dependency of changes in cerebral hemoglobin oxygenation during brain activation: a near-infrared spectroscopy study,” J. Cereb. Blood Flow Metab. 15(6), 1103–1108 (1995).
33. M. Tsuji, A. Duplessis, G. Taylor, R. Crocker, and J. J. Volpe, “Near infrared spectroscopy detects cerebral ischemia during hypotension in piglets,” Pediatr. Res. 44(4), 591–595 (1998).
34. R. Bi, Y. Du, G. Singh, C. J.-H. Ho, S. Zhang, A. B. E. Attia, J. Li, and M. Olivo, “Fast pulsatile blood flow measurement in deep tissue through a multimode detection fiber,” J. Biomed. Opt. 25(5), 055003 (2020).
35. K. M. Bergonzi, T. M. Burns-Yocum, J. R. Burnstead, E. M. Buckley, P. C. Mannion, C. H. Tracy, E. Mennerick, S. L. Ferradat, H. Dehghani, A. E. Eggebercrecht, and J. P. Culver, “Lightweight scmos-based high-density diffuse optical tomography,” Neurophotonics 5(3), 035006 (2018).