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Confocal Retinal Imaging Using a Digital Light Projector with a Near Infrared VCSEL Source

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Abstract

A custom near infrared VCSEL source has been implemented in a confocal non-mydratic retinal camera, the Digital Light Ophthalmoscope (DLO). The use of near infrared light improves patient comfort, avoids pupil constriction, penetrates the deeper retina, and does not mask visual stimuli. The DLO performs confocal imaging by synchronizing a sequence of lines displayed with a digital micromirror device to the rolling shutter exposure of a 2D CMOS camera. Real-time software adjustments enable multiply scattered light imaging, which rapidly and cost-effectively emphasizes drusen and other scattering disruptions in the deeper retina. A separate 5.1" LCD display provides customizable visible stimuli for vision experiments with simultaneous near infrared imaging.

Keywords

VCSEL; rolling shutter; DLP; retina; confocal; CMOS; multiply scattered light; Aeon Imaging

1. INTRODUCTION

The Digital Light Ophthalmoscope (DLO) uses a confocal imaging technique introduced and developed by Aeon Imaging LLC.¹⁻⁵ A digital light projector (DLP) is configured to rapidly project a sequence of adjacent illumination lines across the field of view. Backscattered light from the target is not descanned, but imaged directly onto a monochrome complementary metal oxide semiconductor (CMOS) sensor with a rolling shutter method of detection. In this configuration, the rolling shutter acts as a flexible electronic aperture, which can be finely adjusted in position and width through software in real-time.⁵⁻⁸ By temporally and spatially synchronizing the illumination lines to the rolling shutter exposure, the DLO acquires a line-confocal image of the entire field of view every frame. To image the retina, the illumination and detection pathways are combined using a beamsplitter, shown schematically in Figure 1.

The DLO has been previously used for non-mydratic retinal imaging for diabetic retinopathy screening.²⁻³ For this application, a DLP LightCrafter 4500 (Texas Instruments Inc., Dallas TX) was configured to display up to 48 fixed line patterns using the internal green and red LED illumination. To visualize the retinal vasculature at high contrast while simultaneously reducing pupil constriction and patient discomfort, the DLP's broad green

LED spectrum was long pass filtered at 570 nm to obtain a 580 ± 10 nm output. This spectral range has been shown to provide strong hemoglobin and oxygenated hemoglobin absorption⁹, providing similar images to those obtained at shorter (500–570 nm) wavelengths. Images taken with alternating red and yellow illumination can be combined to provide a pseudo-color image with adjustable color balance to the operator in real-time, or to an image reviewer in post-processing, as shown in Figure 2.

The use of red (630 nm) and yellow (570–590 nm) illumination permits an indirect measurement of blood oxygen saturation in the retinal vessels and arteries, which can help to determine the health and proper functioning of the retina.¹⁰ A small-field version of the DLO has been presented that uses adaptive optics to visualize retinal photoreceptor cones and capillaries and performs two-wavelength oxygenation measurements of the microcirculation.¹¹ Separately, a DLO has been presented that short-pass filters the broad green LED to provide 535–555 nm excitation for autofluorescence imaging.¹² Images acquired from each of these DLO configurations are provided in Figure 3.

A natural extension of the DLO technology is to operate the DLP with a video input instead of the relatively fixed, preloaded patterns.¹³ In this configuration, the DLP parses an RGB video input with 8 bits/channel into 24 monochrome bit planes, each of which is displayed sequentially in time. The DLP LightCrafter 4500 is capable of accepting 120 Hz video input, providing an exposure time of 347 μ sec per bit plane. Custom OpenGL software was used to generate a sequence of adjacent lines; on the PC, the lines have a 0–7 bit-shifted intensity for each RGB color channel, which is translated to a sequence of 24 monochrome bit planes. The primary advantage of using a 120 Hz video input is the ability to change the illumination geometry in real-time via software. For example, stimulus crosses, dots, text, etc. can be used as texture masks in OpenGL, which then appear as black-on-red stimuli to the subject, as previously done using the Rodenstock SLO (scanning laser ophthalmoscope). The black on red stimuli provide the advantage that scattered light from a bright stimulus does not lead to artefactual detection of visual targets by neighboring regions of healthier retina. Due to the absence of light in the stimulus region, the live image stream shows the precise location of the stimuli on the retina without behavioral calibration, which enables a more accurate assessment of visual function, as shown in Figure 4.

A secondary advantage to driving the DLP with a video input is the ability to adjust the width and position of the illumination lines in real-time. Coupled with the control of the width and position of the rolling shutter detection aperture, this permits further flexibility in tuning the system for optimizing the light return, image contrast, or frame rate. For example, the DLO can acquire images at 15 Hz with up to 192 lines displayed per frame. The number of lines per frame, corresponding line width, camera pixel clock and timing delay can all be adjusted in real-time to vary the frame rate, e.g. 72 lines for 40 Hz imaging, or 48 lines for 60 Hz imaging.

The use of an external video input for simultaneous imaging illumination and stimuli permits a variety of visual function tests such as static and kinetic perimetry and task-based tests such as reading, with a single illumination source. However, it is not particularly effective at providing multi-colored and multi-intensity stimuli, due to the perceived bright

background illumination needed for imaging. To enable a wider range of visual function tests, a custom near infrared (NIR) illumination source was built and integrated into a DLO, with visual stimuli provided by a separate 5.1" LCD screen.

The advantages of NIR illumination for retinal imaging are well-known from prior work with scanning laser ophthalmoscopes (SLOs).⁹ NIR light is barely visible to the subject, which improves patient comfort and avoids pupil constriction, and is safe to use at higher powers than shorter wavelength light. Hemoglobin and oxygenated hemoglobin absorption is lower in the NIR than in the 500–590 nm range, which reduces the contrast of the retinal vessels and arteries; however, NIR light penetrates deeper into the retina, providing greater visibility of choroidal vessels, and better visualization of scattering defects using multiply scattered light imaging.^{6,14}

Multiply scattered light imaging is particularly beneficial for detecting disruptions in the deeper retinal layers that are normally obscured by strong superficial scattering. The Sarks¹⁵ model of the development of age-related macular degeneration (AMD), shown schematically as a cross-section of the retina in Figure 5, shows the location of drusen deposits and hyperpigmentation in the deeper retinal pigment epithelium (RPE) layer. These can be seen in optical coherence tomography (OCT) B-scan cross-sections of the retina, shown in Figure 6. While OCT provides a clear view of scattering disruptions in the RPE layer, it must trade-off its B-scan density with the time to acquire a full 3D scan across the field of view. If too large a spacing is taken between B-scans (>60 μm), smaller drusen and pigmentary changes can be missed. By comparison, multiply scattered light imaging immediately identifies scattering disruptions across the full field of view, but with less precise depth characterization.

The DLO is built on the use of the CMOS rolling shutter as a flexible electronic aperture. Its predecessor, the Laser Scanning Digital Camera (LSDC), uses a NIR vertical cavity surface emitting laser (VCSEL) illumination source and either a polygon or galvanometer scanning mirror to scan a line across the field of view. As with the DLO, the illumination line is spatially and temporally synchronized to the rolling shutter, which acts as a spatial filter.^{5–7} On both the DLO and LSDC, the timing synchronization can be adjusted through software to offset the position of the confocal aperture with respect to the illumination line, which permits the operator to switch from confocal to multiply scattered light imaging in real-time. Due to the ease of switching from confocal to multiply scattered light mode with the rolling shutter, the DLO and LSDC are well-suited to rapidly and cost-effectively detect early signs of AMD.

2. METHODOLOGY

The DLP LightCrafter (Texas Instruments, Dallas, TX) has a digital micromirror device (DMD) with a native resolution of 608×684 micromirrors that is used to spatially modulate the illumination source. The micromirrors are packed in a diamond-shaped geometry, each of which can be tilted $\pm 12^\circ$ to direct the illumination to either an output or a beam dump. After removing the DLP LightCrafter's visible LED optical module, it can be replaced using a telecentric or non-telecentric design architecture, with unique considerations that apply for

coherent (laser) and non-coherent (LED, lamp) light. Details regarding the operation of the DMD, design considerations, and advantages/disadvantages of each approach are provided in application notes.^{16–18}

In this work, a custom projector was built with a telecentric optical design to couple directly into the DLO and ensure that the magnification does not change when correcting for a subject's refractive error. A low divergence 860nm VCSEL (RPMC Lasers Inc., O'Fallon MO) with a maximum output power of 750mW was used as the illumination source. A VCSEL was a reliable source used for NIR imaging with visual stimuli in the LSDC⁸, and the current VCSEL was selected for the DLO due to its relatively uniform and circularly symmetric output profile and low speckle noise. As compared to a typical high-power LED, the VCSEL has a smaller etendue and greater spatial coherence; this allows the VCSEL to remain collimated over greater distances, avoids the need for a custom total-internal-reflectance (TIR) prism, facilitates the addition of coherent sources at other wavelengths, and eliminates losses from pupil shaping. Due to its relatively uniform field, a separate integrating element, such as a fly's eye lens, integrating rod, fiber, or liquid light guide was not required and was omitted for this source to save on space and cost, and avoid unnecessary power loss. For safety, the VCSEL output is modulated using a signal derived from the camera and DLP output triggers. The VCSEL will only switch on during an active frame exposure. If the DLP output trigger count at the end of a frame exposure is incorrect, the microcontroller will retry on up to 5 frames before switching off the VCSEL and requiring a hardware reset.

When using a VCSEL or other coherent source in a DMD-based projector, diffraction will occur due to the regular spacing of the micromirrors, producing a set of diffracted output beams with an angular spacing that is dependent on the micromirror geometry, illumination wavelength and incident angle. DMD diffraction has been characterized in the design of laser-based custom projection systems used for wavelength tuning,¹⁹ tunable fiber lasers,²⁰ quantitative phase imaging,²¹ and fluorescent microscopy.²² It is generally desired to couple as much of the illumination power into the zero order diffracted output (the specular reflection) of the micromirror when tilted into an ON state. To achieve this, the illumination angle of incidence was carefully selected so that a higher diffraction order is aligned with the zero order diffracted output, resulting in a blazed configuration.¹⁸

The VCSEL was collimated with a 75 mm achromat lens to over-fill the DMD and use the central, most uniform, portion of the beam. Kinematic mirror mounts provided fine adjustment of the incident angle, nominally set to 38 deg from the flat DMD surface, to align the 4th diffraction order with the specular output. With all DMD mirrors on, a 20% efficiency was achieved. A tilted 30 mm achromat collection lens is used to remove keystone distortion and focuses the output to a pupil stop, which shapes the illumination pupil as well as blocks all other diffraction orders. The illumination is then expanded by 5x and coupled into the DLO with a Maxwellian view, shown in Figure 7. A polarizing beamsplitter is used to remove unwanted reflections from optical components. The illumination field size is 3.115×3.115 mm at the DMD surface; due to the pixel addressing format, this corresponds to 288×576 micromirror pixels on the DLP3000 DMD. The field is magnified to an approx. 1024×1024 pixel region of interest (5.427×5.427mm) at the rolling shutter CMOS detector

(IDS Imaging Development Systems GmbH, Obersulm, Germany) using two lenses in the detection pathway. Non-mydratic imaging is performed at 50 Hz using 48 pre-loaded illumination lines and a typical time-averaged power at the cornea of 220 μ W. As with the earlier DLO systems, the image contrast can be increased by adding illumination lines and reducing the frame rate; images with up to 192 lines have been acquired at 12 Hz.

The stimulus channel was designed to be diffraction limited over the visible wavelength range with a 1.6mm pupil and with swappable ocular lenses that provide 0.5 and 1 arc min/pixel resolution. With 1920 $\times$ 1080 pixels, the stimulus field of view with 1 arc min/pixel was 25 $\times$ 18 deg. and a maximum of 132 cd/m² luminance was achieved. The LCD screen is controlled through software as an external monitor, capable of red, green and blue output with 8-bits per channel. Custom calibration software is used to align the LCD stimuli field to the imaging field of view. The stimulus display has been controlled using two software libraries commonly used by the vision research community: the PsychoPy API, accessible via Python software development, and Psychtoolbox, accessible via MATLAB (The Mathworks, Inc. Torrance, CA).

To improve ease of use, a patient response button and a microphone with audio recording were added to the DLO. By controlling these components with the DLO's microcontroller, accurate button-press timing and audio responses are made available directly in software (Matlab or Python) via a platform independent serial port.

3. RESULTS

As compared to imaging with visible wavelengths, the implementation of the custom NIR illumination module provides a significant gain in the detected signal to noise ratio, since pupil constriction and patient discomfort typically limits the visible illumination power to 30–50 μ W. Furthermore, the low etendue of the VCSEL source permits a very small (<1mm) illumination pupil, which reduces the extent of corneal reflections at the sensor and permits a larger pupil collection area. A small illumination pupil can be particularly beneficial when imaging older and diseased eyes, when the pupil may be constricted or may be partially obscured by lens opacities. As expected, the use of 860 nm illumination was highly scattering, penetrated the deeper retina, and qualitatively appeared to have similar vessel contrast as seen in images acquired with red (630 nm) illumination. Confocal and multiply scattered light images were acquired with the NIR and visible wavelength DLO systems of 5 subjects aged 29–67 yr. As shown in Figures 8–9, the superficial light return shown in the confocal images will often mask scattering disruptions such as drusen from the deeper layers, which are revealed in the multiply scattered light images.

To test the functionality of the LCD stimuli and audio recording used to measure responses, a single-line reading test with simultaneous NIR imaging was performed on 9 normal subjects. The subjects were instructed to read aloud a sentence with an average of 60 characters and 12 words in 20/60 Helvetica font. The sentences were presented at various contrast levels using Psychtoolbox, with and without added spatial noise and sinusoidal text motion. The reading speed of the sentences was evaluated by calculating the duration of the recorded speech. In this experiment, the subjects did not perceive the NIR background

illumination. As expected, reading speed decreased significantly with contrast of the letters against the background, both with and without noise ($p < 0.005$). For the rapid dynamic noise used, text motion did not increase reading speed, and in fact was detrimental.

4. CONCLUSIONS

A custom VCSEL-based NIR illumination module has been developed for the Digital Light Ophthalmoscope (DLO). With a lower etendue than LEDs, minimal speckle, and uniform field, the VCSEL provides a high power efficiency without the need of a TIR prism or integrating element. When over-filling the DMD and operating in a blazed condition, a 20% power through-put was attained. When integrated into the DLO for retinal imaging, the high spatial coherence of the VCSEL source permits a small ($< 1\text{mm}$) illumination pupil, which facilitates imaging in older and diseased eyes and allows for a greater region of the pupil to be used for detection when the pupil area is split between input and output regions. The deeper penetration of the NIR illumination, combined with the all-electronic adjustment of the rolling shutter confocal aperture, permits rapid and cost-effective visualization of scattering disruptions such as drusen and hyperpigmentation that are commonly seen in AMD and pre-AMD patients.

With the use of NIR illumination for retinal imaging, a separate LCD was added to the DLO to provide RGB color stimuli for vision research experiments. By independently controlling the LCD as an external monitor, custom stimuli and test protocols can be created using open source software libraries such as Psychtoolbox and PsychoPy, with simultaneous NIR imaging.

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Figure 1.

Left. Optical schematic of the Digital Light Ophthalmoscope (DLO). The retina is illuminated by a sequence of line patterns generated by the digital light projector. Light return is not descanned, but imaged directly onto a 2D CMOS rolling shutter sensor. Conjugate pupil planes in the illumination and detection pathways provide pupil separation to eliminate corneal reflections and timing synchronization set the position of the illumination line relative to the sensor pixel row being exposed. Right. Photograph of the DLO used for diabetic retinopathy screening.

Figure 2.

Images of an undilated 36 year old Caucasian diabetic male acquired at Joslin Diabetes Center in Boston, MA. (A) DLO red-green pseudo-color image; (B) DLO RGB pseudo-color image; (C) Magnified and cropped Optos image; (D) Central ETDRS field Zeiss FF4 fundus photo (taken after dilation).

Figure 3.

Left. Central macula images of a dilated normal 29 year old African male subject acquired with an adaptive optics implementation of the DLO using red (630 nm) illumination.¹¹ The photoreceptor cone mosaic (A1) and retinal capillaries (A2) are visible, 100 µm scale bar. Right. The DLO was configured for simultaneous confocal and autofluorescence mydriatic imaging by filtering the green illumination LED to 535–555 nm.¹² The confocal images are registered and averaged (B1). The registration coordinates are then applied to the very dim autofluorescence image frames and averaged (B2), showing typical patchy hypofluorescence and hyperfluorescence in a patient with age-related macular degeneration (AMD).

Figure 4.

The DLO configured for low vision uses a DLP LightCrafter 4500 to generate illumination lines for confocal imaging, as well as custom stimuli that appear as black shapes on the typically red illumination background. To provide real-time updates to the stimuli, e.g. for guiding a subject's fixation with the mouse, the DLP is configured to convert a 120Hz RGB 24-bit video input to 24 monochrome bit planes that are displayed sequentially in time. Since the stimuli are formed by the absence of light, they are visible in real-time on the retinal image display. Left. Single image frame with text stimuli, which can be used to monitor reading performance. Center. Single image frame during a static perimetry test, with a fixation cross and Goldmann III target. The visualization of the target indicates which part of the retina was stimulated, even with unstable fixation. Right. Registration coordinates are plotted in color to indicate the direction of a subject's gaze over time (proceeding blue to red), as previously done with Aeon Imaging's Laser Scanning Digital Camera⁸. Image registration and averaging is often used to improve signal to noise.

Figure 5.

Schematic cross-section of the retina, redrawn from the Sarks¹⁵ model of the development of AMD (left to right), from the earliest stages to end stage atrophy. The important damage in eyes with AMD is beneath the highly reflective vitreo-retinal interface, and therefore obscured when imaging the directly backscattered light. Multiply scattered light helps visualize the debris and fluid in the deeper layers.

Figure 6.

A. En face retinal image of an undilated 67 year old male taken using the Heidelberg Spectralis SLO. Green horizontal line shows the location of the OCT scan in B, with two red lines denoting the mean thickness range B. SD-OCT B-scan (cropped), showing drusen and hyperreflective foci. Due to time considerations, a coarse B-scan step size is sometimes used, which can miss the detection of small and medium sized drusen that are located between scans. C. and D. 20 deg SLO retinal images of a patient with AMD, with NIR (790 nm) confocal (left) showing drusen as reflective and multiply scattered light imaging at 633 nm, emphasizing hyperpigmentation (dark clumps).¹⁴

Figure 7.

Schematic optical diagram of the DLO with a custom NIR VCSEL source. The VCSEL is collimated and illuminates the DMD in a blazed configuration. The output is focused by a tilted lens to eliminate keystone distortion and is shaped at a conjugate pupil plane. The illumination is then magnified and relayed to the eye. Backscattered light is magnified and imaged directly onto the CMOS rolling shutter sensor. An LCD panel is coupled into the system using a dichroic beamsplitter, providing 24-bit RGB stimuli under independent control for vision experiments.

Figure 8.

Near infrared (860nm) DLO retinal images of an undilated 50 year old female subject with dominant drusen. Left. Confocal image, emphasizing the superficial layers of the retina. Right. Multiply scattered light image, showing widespread drusen (white bumps). The transition from the left to right modes of operation is completely electronic and achieved through software in real-time.

Figure 9.

DLO retinal images of an undilated 67 year old male subject with early lens changes, the same subject as in Figure 6A–B. A. 15 deg confocal image of the central macula using Aeon’s custom 860nm illumination module, showing more superficial detail than seen in fundus cameras. B. Multiply scattered light image of the same retinal location, showing drusen (small bumps with shadows) and hyperpigmentation (dark spot) that cannot be seen in the superficial confocal image. C. 25 deg. confocal image with red 630nm light. D. Multiply scattered light image of the same retinal location, with arrow indicating an area of hyperpigmentation. The transition from A. to B. and C. to D. modes of operation is completely electronic and achieved through software in real-time.