Optical probe based on double-clad optical fiber for fluorescence spectroscopy

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Abstract: We report an optical probe based on a single double-clad fiber (DCF), which is suitable for fluorescence spectroscopy. The excitation light is delivered through the small diameter core of the DCF and the excited fluorescence light is collected by the large diameter inner cladding of the same fiber. To retrieve the signal beam from the inner cladding, we utilize a DCF coupler that couples only the light beams in the inner claddings of two different pieces of DCF. It was found that the separation of the channel for the excited beam from the channel for the excitation beam in the same piece of fiber could diminish the autofluorescence background noise generated by the fiber itself, while maintaining all the benefits of a single-fiber probe system. The usefulness of the DCF probe and the performance of the DCF coupler are then reported by presenting the fluorescence spectrum of a fresh gingko leaf and comparing it with the spectrum taken with conventional methods. The fabrication process of the DCF fiber and the inner cladding mode coupler for it are also presented.

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1. Introduction

Fluorescence spectroscopy has become an attractive modality for non-invasive and in vivo examinations, and has been widely used in such diverse fields as biomedicine and chemical engineering [1-3]. A fluorescence spectroscopy system is typically composed of a light source, a spectrometer, and a fiber-optic probe to allow for both light delivery and data collection. A versatile interface between the instrument and the tissue under test, provided by various fiber-optic probes, enables effective detection of fluorescence light excited by the excitation laser beam delivered through the probe.

A variety of fiber-optic probes for fluorescence spectroscopy have been reported and well summarized in Ref. [4]. Generally, commercially available silica fibers have been employed in the fabrication of fiber probes, and fiber-optic probes for fluorescence spectroscopy can be categorized into two types: a single-fiber type and a multi-fiber type. A single-fiber type probe uses a single fiber both to deliver the excitation beam and to collect the excited fluorescence signal beam. Conversely, multiple fibers are used for the same purpose in the multi-fiber type probe.

A single-fiber probe has advantages of small probe diameter, simple configuration, and low factory cost over the multi-fiber probe. However, the application of single-fiber probes has been limited by the autofluorescence background signal inherent in the fiber itself [4, 5]. The light beam passing through a fiber excites the fiber material and generates a counterpropagating fluorescence light, referred to as the fluorescence background signal. In general, the fiber core induces stronger fluorescence than the fiber cladding due to the doping material. Then, when the fiber is long and/or the sample has a very weak fluorescence, the background signal can in fact dominate the measurement, thereby making it difficult to discern the fluorescence signal coming from the sample of interest. The multi-fiber probe does not have the problem regarding the fluorescence background signal, as the fiber(s) for collecting the fluorescence signal are different from the fibers used for delivering the excitation beam [3, 4].

In order to avoid the generation of Raman and fluorescence background signals, fiber probes using a hollow waveguide (HW) [6] or an air-core photonic crystal fiber (PCF) [7] have been introduced. In both cases, the excitation beams were guided through the air-core, thus, the background signals originating from the core material were not inherently generated. However, the HW [6] case had disadvantages of high bending loss, an exceedingly small NA, and a large diameter. Specifically, since the probe was a single-fiber type, a bulk type beam splitter has typically been used to retrieve Raman or fluorescence signals. For the PCF case [7], even though the excitation beam could be guided through the air-core of the PCF, the excited signal was collected by three multi-mode fibers surrounding the PCF, thereby incurring all the disadvantages of the multi-fiber type probe. Furthermore, the exorbitant price of the PCF also limited its widespread application. In addition, the air holes of the HF and PCF should be prohibited from directly contacting liquids, otherwise they could be damaged due to capillary action.
In this paper, we report the fiber probe that is constructed in a single-fiber type but operates as a multi-fiber probe; thus having all the merits of both types of probes. By utilizing an optical fiber having double claddings (double-clad fiber; DCF), we could deliver an excitation beam through the small diameter conventional core of a fiber and subsequently collect the excited fluorescence light signal using the large diameter inner cladding of the same fiber. To retrieve a signal beam in the DCF cladding that was not affected by the background beam in the core, we designed and fabricated a special fiber coupler that coupled the optical powers of the cladding modes of two different DCF pieces. Here, the usefulness of the proposed DCF probe and the performance of the fabricated DCF coupler are reported by presenting an all-fiber fluorescence system. The system performance is then compared with the ones of conventional systems. The fabrication process of the DCF fiber and the inner cladding mode coupler are also presented.

2. Method

2.1 Double-clad fiber (DCF)

A double-clad fiber (DCF), usually having two concentric cladding layers around a single core, was originally developed for high-power fiber lasers [8] and used for fiber gratings [9]. One of the cladding layers of the DCF, usually the inner one, generally acts as a multi-mode waveguide; thereby allowing another channel in addition to the conventional core channel. Recently, double-clad PCFs were used in two-photon fluorescence and second harmonic generation microscopy to enhance the signal collection efficiencies [10, 11]. Applications of DCFs in three-dimensional imaging endoscopy and combustion gas thermometry, based on absorption spectroscopy, were also reported [12, 13]. However, in most applications, dichroic mirrors or bulk type beamsplitters have been used for accessing the beams in DCF channels. As such, even though a system could benefit from the DCF, there were inherent problems not only in system complexity but also in optical alignment due to the use of bulk-optic based devices. Therefore, it is necessary to devise the coupler that can retrieve only the beam in the signal channel, the inner cladding channel in this case, for maximizing the benefits of DCF-based probes.

For our experiment, we fabricated a DCF by drawing the preform of a conventional single mode fiber with a conventional fiber drawing tower. However, the acrylate coating on the fiber was made with a low-index polymer (Luvantix, PC375) instead of the high-index coating generally used for a conventional single mode fiber. The low-index coating on the conventional cladding enables the cladding layer to guide modes. In other words, with the low-index coating, the cladding layer becomes a large diameter multimode waveguide referred to as the inner cladding layer. However, since the low-index coating acts as the cladding of the inner cladding waveguide, it could also be referred to as the outer cladding layer, thus enabling us to call this fiber a double-clad fiber (DCF). Hence, the fabricated DCF has two concentric waveguides; one is the single mode core at the center and the other is the multimode inner cladding around the core. Due to this unique structure, in a fluorescence spectroscopy system, we can deliver the excitation laser beam through the small single mode core of the DFC and then collect the excited fluorescence signal beam by using the large inner cladding of the same fiber.

The DCF used in this experiment had a step-index profile with the diameters of the core, the inner cladding, and the outer cladding being 7.4 μm, 125 μm, and 180 μm, respectively. The numerical apertures (NAs) of the core and the inner cladding were measured to be 0.12 and 0.44. The propagation loss of the core mode was 0.08 dB/m, but the inner cladding had a rather large loss of 0.66 dB/m at a wavelength of 635 nm. The high propagation loss of the inner cladding mode is mainly due to absorption caused by the polymer material itself. However, since a short total system length could be maintained, propagation loss due to the inner cladding did not create any severe problems.
2.2 System configuration

Figure 1 shows the optical system for fluorescence spectroscopy, which is based on the proposed single-DCF probe. The excitation beam coming from an Argon-ion laser is delivered through the small single mode core of a DCF, and the fluorescence signal beam excited by the sample is subsequently collected by the large multimode inner cladding of the same DCF.

To launch the excitation laser beam (488 nm, Stabilite 2017, Spectra-Physics) into the core of the DCF, the laser beam was first coupled to a 3.0 m-long single mode fiber (SMF). The SMF had the same core diameter as the DCF and was fusion spliced with the DCF. A neutral-density filter (ND) was used to adjust the intensity of the input beam, and a bandpass filter (BP) (488 ± 10 nm) was inserted to remove the background noise of the laser. To ensure that the laser beam was launched into only the core mode of the DCF, a mode stripper was used. About a 3 cm-length of the low-index coating of the DCF was removed and immersed into an index-matching oil. In this way, it could be ascertained that the excitation beam was guided to the sample through only the core of the DCF. Even though the beam passed the fiber coupler before reaching the sample, it was not appreciably affected by the coupler because the DCF coupler was specially designed to couple only the cladding modes, not the core modes.

The fluorescence signals excited at the sample were collected by the inner cladding layer of the DCF. As previously mentioned, the inner cladding had a diameter as large as 125 μm and was concentric with the core. At the DCF coupler, the beam in the inner cladding of port P2 was managed to be coupled to the inner cladding of port P4; thus, only the excited fluorescence signals, not the beam in the core contaminated by the fiber background, were directed to the detection arm of the system. Before coupling the fluorescence signals into a 50 μm-core-diameter multimode fiber (MMF), a 500 nm longpass filter (LP) was attached to block the fundamental excitation light that was elastically scattered back from the sample. Finally, the spectrum was measured with a spectrometer (S2000-FLG, Ocean Optics, Inc., Dunedin, Florida). The fiber lengths of the probe arm, the source arm, and the detection arm of the DCF coupler were approximately 0.7 m, 1.0 m, and 1.0 m, respectively.

2.3 Cladding mode coupler

A fiber coupler is a device that couples the core mode of a fiber with the core mode of another fiber in general. However, since the excited fluorescence signal is designed to be guided through the inner cladding of a DCF, for the proposed system it is necessary to have a coupler that couples the optical power between the inner claddings of two different pieces of DCF. A conventional bulk-optic based beamsplitter can be used to retrieve the signal beam as in conventional systems; however, a system based on this type of splitter might lose most of the benefits of using the DCF probe. Therefore, a 2x2 DCF cladding mode coupler was
devised and fabricated using a side-polishing method [14]. A piece of DCF was inserted and glued into a slot made on a silica block; the slot was a V groove curved with a curvature radius of 0.5 m. Then, the silica block embedding the fiber piece was roughly polished on a brass plate using alumina powder with about a 5 μm particle size. Fine polishing was made using cerium oxide powder with less than a 1 μm particle size on a polyurethane plate. By mating two silica blocks having the side-polished DCFs, the DCF coupler could be constructed. By adjusting the polishing depth, we could control the coupling ratio. In our experiment, the polishing depth was controlled by monitoring the coupling ratio in real time. We measured the coupling depth to be about 35 μm and the coupling length was about 6 mm. Even with the polishing, the thickness of the remnant inner cladding was larger than 20 μm, which was thick enough to confine the core mode within the core part of the DCF. Therefore, there no appreciable amount of mode coupling between the core and the inner cladding or between the cores was expected.

![Fig. 2. Schematic diagram of the DCF coupler. The entire optical beam coming along the core of P1 port is delivered into the core of P2 port, and vice versa. However, a part of the beam coming from P2 port but along the inner cladding of the first fiber F1 is coupled to the inner cladding of the second fiber F2 and directed to P4 port.](image)

Figure 2 shows the schematic diagram of the implemented DCF coupler. The entire beam coming from P1 port along the core of F1 fiber is delivered into the core mode of the same fiber at P2 port without being affected by the coupler. The same thing happens with the beam coming from P2 along the core. However, a part of the beam coming from P2 but along the inner cladding layer can be coupled to the second fiber F2 and be directed to P4 with the help of the proposed DCF coupler. Hence, the DCF coupler enables exclusive access to the light beam in the inner cladding of the DCF probe. Note that P3 port has no function in this experiment, thus is blocked by dipping its end into an index-matching oil.

![Fig. 3. Coupling efficiency of the DCF fiber coupler measured as a function of the wavelength. The left-hand and right-hand inset figures are the near field images taken at the through port and cross port of the DCF coupler, respectively.](image)
Figure 3 shows the coupling efficiency of the DCF coupler measured in a wavelength range of 600–800 nm. First, a white-light source was launched into P2 port of the DCF coupler shown in Fig. 1, and then the output spectrum at P4 port was measured. The same measurement was repeated with a piece of DCF of the same length but without using the coupler. The ratio between the two measurements was taken as the coupling efficiency of the coupler. The figure shows that the coupling efficiency is around 10–15% and slightly increases with the wavelength. The inset figures are the near field images taken at the two output ports of the DCF coupler. The through port in the left has a bright spot at the center, corresponding to the core mode, while the cross port in the right has no appreciable core mode. This measurement confirms that the mode coupling between the core modes of two DCFs is not appreciable. Instead of side-polishing, by using a fusion method we could also fabricate a DCF coupler. Our first try gave a better coupling efficiency but the fluorescence background signals were a little more severe. The detailed spectral properties of both types of DCF couplers are currently under investigation. We are also trying to get more reliable DCFs.

2.4 Experiment

To test the performance of the proposed system, the fluorescence spectra of fresh gingko leaves were measured. The gingko leaf is known to have fluorescence emissions at the red and far-red wavelength regions, which are remote from the background spectrum of the fused silica under 488 nm excitation. The DCF probe was used as an optical probe and the measurement was made with the setup shown in Fig. 1. The tip of the fiber probe was placed such that it slightly touched the upper surface of a gingko leaf that was fixed on a slide glass.

Fig. 4. Normalized fluorescence spectra of a fresh gingko leaf under 488 nm excitation, obtained using (a) the proposed DCF probe, (b) the single MMF probe in the configuration of (d), and (c) the direct-pump configuration in (e). BP, bandpass filter; LP, longpass filter.
Figure 4(a) shows the fluorescence spectrum of the gingko leaf obtained using the proposed DCF probe. We can clearly see the two peaks located around at 685 nm and 740 nm wavelengths, which are the inherent peaks of Chlorophyll \(a\) of the antenna system of photosystem II [15]. However, we also observed some small protuberance near the 500 nm wavelength, which was not related with the sample under test. In order to explore the origin of this signal, the same measurement was made by building two additional conventional systems; one was a single MMF (multi-mode fiber) probe system and the other was a direct-pump configuration system. As shown in Fig. 4(d), the MMF probe system was composed of a single MMF and a conventional bulk-optic beamsplitter. The MMF probe was made of a standard silica MMF having a 3.0 m length and a 50 \(\mu\)m core diameter. The MMF and the DCF were drawn from the preforms having the same properties and using the same Drawing Tower. In the direct-pump configuration, as shown in Fig. 4(e), the excitation laser beam was delivered through free space, and the excited fluorescence signal was collected by a single MMF. A 0.7 mm-diameter laser beam was illuminated on a sample at an angle of 30° and the MMF was placed 3 mm above it.

Figure 4(b) shows the fluorescence spectrum obtained with the single MMF probe system of Fig. 4(d). The spectrum had an unwanted strong spectral band, or background signal, in the wavelength range of 500–530 nm, which was even stronger than the Chlorophyll fluorescence of the gingko leaf. Since the background signal was too strong, the scale of the plot was increased by a magnitude of 4. However, the background signal was hardly found in the spectrum of Fig. 4(c) taken with the direct-pump configuration of Fig. 4(e). Therefore, we could conclude that the abnormal protuberance signal in Fig. 4(a) was not the fluorescence emission of the gingko leaf, but the distinctly suppressed silica background of the fibers.

As an excitation beam, the Argon-ion laser beam was adjusted to have a 10 mW total power at the sample for the direct-pump configuration case, and 5 mW powers for the other two cases. The spot size of the laser beam was around 0.7 mm. The spectrum was taken with an acquisition time of 1.0 s and was normalized to the spectral intensity at a wavelength of 685 nm. All spectra in this paper were corrected with regard to the wavelength-dependent signal detection efficiency of the system using a standard Tungsten Halogen lamp (LS-1-CAL, Ocean Optics, Inc., Dunedin, Florida), and the dark current noise of the detector was subtracted before the spectrum measurements were taken.

3. Discussion

The silica background signal, dominantly occurring around the wavelengths of 500–530 nm, was greatly suppressed with the proposed DCF probe [Fig. 4(a)], especially when compared with the conventional MMF probe case [Fig. 4(b)]. However, it was still appreciable compared with the direct-pump case [Fig. 4(c)]; further investigation into the origin of the silica background signal for the DCF probe is necessary for subsequent improvements in the performance of the proposed system, especially as neither the DCF nor DCF coupler have been sufficiently well established to permit precise analysis. Nevertheless, these preliminary investigations lead us to think the confinement of the excited background light is at the core of the DCF fiber. In a standard SMF, the beam guided in a core mode cannot be coupled to a cladding mode. However, the background light excited at the core can propagate in most directions. The conventional core of the DCF has a low NA, thus it cannot strongly constrain the excited light within the core. Therefore, when the light excited at the core of the F1 fiber in Fig. 2 is directed into the reverse direction, some parts of it can be coupled to the F2 fiber by the DCF coupler through the inner cladding layers, which appear in the background at port P4.

The incompleteness of the DCF and the DCF coupler might also be the origin of the silica background signals. Commercially available all-silica DCFs are usually designed for fiber lasers, not for optical communications, which means that the core of the fiber is specially doped (usually with Yb). The doping material for laser generation has a much
stronger background and significant absorption at certain wavelength regions; this feature makes the fluorescence signal deteriorate. The high price of commercial DCFs also work as a barrier to its widespread use. In addition, we could obtain a larger NA for the inner cladding mode, because the polymer material of the outer cladding can have a much lower refractive index than the silica itself. The fiber with a large NA has correspondingly high signal collection efficiency. Due to these reasons, we designed and fabricated a DCF that had a low-index polymer outer cladding. However, polymers are not as good waveguide materials as the silica-based materials. Thus, we are currently attempting to fabricate an all-silica DCF that has no lasing dopants and single core mode operation even at short excitation wavelengths.

Of course, a more appropriate DCF coupler is also under preparation, which includes the coupler based on a fusion process. Instead of side-polishing, by fusion splicing two DCFs along their sides, a DCF inner cladding mode coupler was also made. A preliminary test of this coupler gave better coupling efficiency; however, the silica background signal was a little higher. A more detailed investigation of the spectral properties of DCF couplers is still underway. Even though the leaf fluorescence spectrum measured by the proposed DCF probe [Fig. 4(a)] still had an appreciable fiber background, the ratio of the signal to the background was at least 50 times larger than that measured by a conventional MMF probe [Fig. 4(b)].

The spectral shape difference noticed in Figs. 4(a)–4(c) can be attributed to the multilayer structure of the leaf and the effective detection depth of the probe. A gingko leaf has a multilayer structure consisting of a waxy coating at the outer surface, a layer of epidermal cells, followed by a spongy mesophyll. The parameters of a fiber-optic probe, including the fiber diameter, NA, probe-sample spacing distance, and illumination-collection fiber separation distance can all have a significant influence on a probe’s fluorescence signal sensitivity. In other words, different probe configurations have different effective detection depths of fluorophore layers in turbid media. This influence of fiber-optic probe parameters on the detection depth has been previously investigated [16, 17]. The DCF probe proposed and used in our experiment can be considered a compact multiple-fiber probe that has a concentric illumination-collection-fiber structure and the smallest possible illumination-collection fiber separation distance. Hence, even though the same sample was used in obtaining the spectra of Fig. 4, the exact locations of the sample where the measurements were made might differ from case to case, which could also explain differences in the shapes of the obtained fluorescence spectra.

Fig. 5. The fluorescence intensities measured in terms of the distance from the probe tip to a sample. The measurement was made at a wavelength of 685 nm with the proposed DCF probe (open circles) and a conventional single MMF probe (open triangles). The solid lines are the fitting curves made using exponential functions.

The variation of the fluorescence signal intensity at a wavelength of 685 nm and with respect to the distance from the probe tip to the sample was investigated for both the DCF and
the MMF probes. The measurements were all made using the same gingko leaf, and the results are depicted in Fig. 5. The data points were curve fitted with an exponential function of \( I = a + b \exp(-d/\tau) \) for each case. In both cases, the figure shows that the fluorescence intensities decrease with the distance, and the decaying constants for the DCF and the MMF probes are \( \tau_{\text{DCF}} = 0.944 \text{ mm} \) and \( \tau_{\text{MMF}} = 1.009 \text{ mm} \), respectively. This measurement concludes that the proposed DCF probe provides a similar working distance as a conventional single MMF probe.

4. Conclusion

A fluorescence spectroscopy system based on a single double-clad fiber (DCF) probe and a special coupler for it was designed, established, and evaluated. By making a low-index polymer coating on a conventional single mode fiber (SMF), we could construct a double channel fiber that uses the conventional cladding of an SMF as the detection channel for the excited fluorescence signals, in addition to the conventional core as the excitation channel. An excitation laser beam was delivered through the small core of the DCF and the excited fluorescence signal was subsequently collected by the large diameter inner cladding of the same fiber. To retrieve only the signal beam in the inner cladding layer, a special DCF coupler was fabricated by using the side-polishing method, which allowed exclusive access to the excited fluorescence signal.

The performance of the fluorescence spectroscopy system composed of the DCF probe and the DCF coupler was confirmed by taking the fluorescence spectrum of a gingko leaf and comparing it with the performances of conventional systems. When a 488 nm Argon-ion laser was used as the excitation source, the proposed system gave well defined inherent Chlorophyll fluorescence peaks at 685 nm and 740 nm. Even though the proposed system had a non-negligible amount of silica background signals around the wavelengths of 500–530 nm, it was much smaller than the ones obtained with the conventional system using a single multimode fiber (MMF) as the probe.

A DCF-based system for fluorescence spectroscopy could enable wide applications, if the coupling efficiency and the reliability of the DCF coupler could be improved a little more; disease diagnosis in a narrow duct is a good example of practical applications. A similar configuration or scheme could be also used for reflectance spectroscopy to cope with the problem of significant specular reflection at the end surface of the probe tip.

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