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# In Vivo Sized-Fiber Spectroscopy

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## ABSTRACT

Sized-fiber array spectroscopy describes a device and method for measuring absorption and reduced scattering properties of tissue. The device consists of two fibers with different diameters that are used to measure the amount of light back-scattered into each fiber. Only one fiber emits and collects light at a time. Recent innovations allow for a wide spectral emission and collection of diffuse reflectance. Experimental results are presented for a device with these fiber sizes that demonstrate the sensitivity of the device in measuring samples of tissue.

**Keywords:** Reflectance, Optical Biopsy

## 1. INTRODUCTION

In many fields of medicine, the determination of optical properties of a tissue can be valuable information. Simple and rapid measurements of the optical properties can be ascertained by the sized-fiber device descibed in this paper. The device is based on the fact, that in general, tissues with different scattering and absorption properties will scatter different numbers of photons back into a fiber. If only a single fiber is used, two samples with very different optical properties can backscatter the same number of photons. To distinguish the samples, a second measurement must be made using a different size fiber.

This paper begins with a description of the sized-fiber device, consisting of two sizes of bifurcated fiber, and construction of the device. Following will be a description and results of the experiments which measure the reflectance of *in vitro* and *in vivo* porcine tissue.

The sized-fiber device was adapted to use white light measurements instead of a limited wavelength source from a laser, literally giving a spectrum of information. This was made possible by a special bifurcated fiber, which I shall refer to as a Y-fiber. A two fiber bundle was coupled into a single fiber making a "Y" shaped junction with the three fibers. The two fiber bundle was butted onto the face of the single fiber with a core diameter twice the diameter of the fibers in the bundle, as shown in figure 1. In the two fiber bundle, the function of one fiber is a light input fiber from a lamp; the other is the output (backscatttered photons) signal coming back from the large fiber. This has the benefit of reducing reflection noise by separating the lamp light from the back-scattered light from the tissue. The sized-fiber device has two Y-fibers with different core diameters, thus the descriptive name, sized-fiber spectroscopy. The two Y-fibers are joined together and inserted through a needle to measure diffuse reflectance in tissue.

## 2. MATERIALS AND METHODS

The construction of a single Y-fiber begins with the preparation of the three fibers; two fibers, having a core diameter $\leq 50\%$ than the diameter of the larger fiber core, and cut to 2 m in length. For these experiments, 100 $\mu$m and 200 $\mu$m fibers compose one Y-fiber, and the other Y-fiber by 300 $\mu$m and 600 $\mu$m fibers. The fibers are perpendicularly polished and 1 cm of the fiber jacket is removed. The use of glass-glass (core-cladding) fibers ensures that the cladding remains intact to the end of all fibers. The two smaller fibers are mounted side by side in an SMA connector. The large fiber is also mounted in an SMA connector. An SMA coupler connects the fibers together without the use of index matching oil or gel.

The Y-fibers must then be joined. The 200 and 600 $\mu$m fibers are bound together at the free fiber end with UV cured epoxy(191–M, Dymax corp, Torrington, CT). Prior to epoxying the ends of the fiber, the two fibers are

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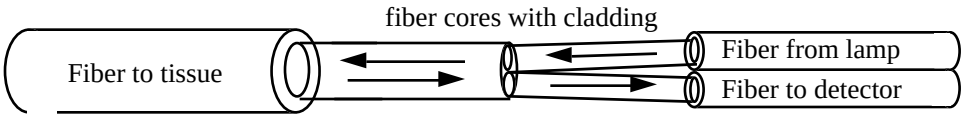


**Figure 1.** Three fibers are coupled together to form a Y-shaped junction. Light is guided from the lamp to the tissue; diffusely relected light from the tissue is guided back to the detecor, minimizing noise. The arrows indicate the direction of light travel which affect the measurements. With the noise minimized, very small reflecances ($\sim$0.1%) are detectable.

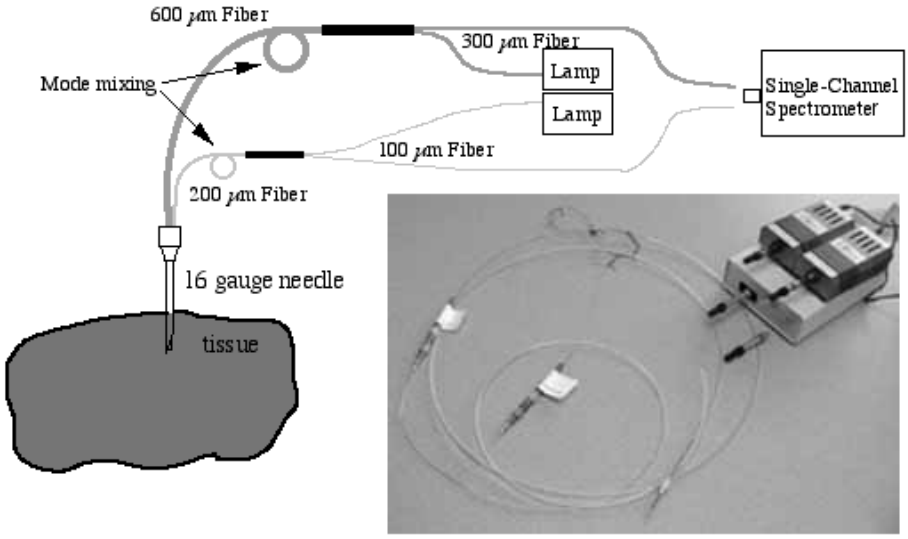


**Figure 2.** This is a schematic diagram for the sized-fiber measurements. Only a single spectrometer was available; the two Y-fibers share a single spectrometer. SMA fiber mounts are used to ease the switching of the fibers between measurements.

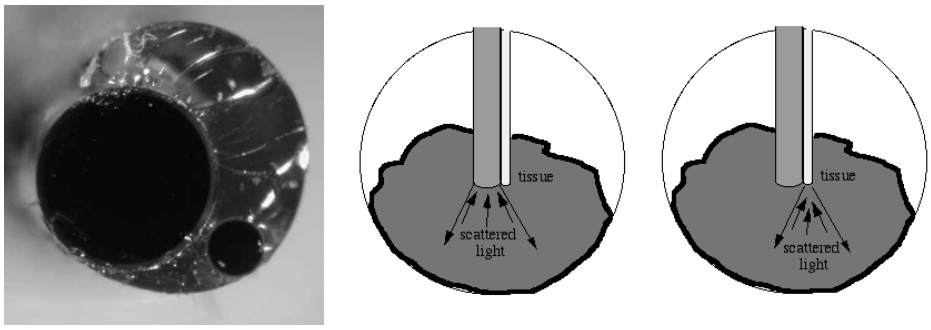


**Figure 3.** The far left picture shows the proximate location of the fiber faces fixed in epoxy which are inserted through a needle. The next two figures show how each fiber measures tissue independently.

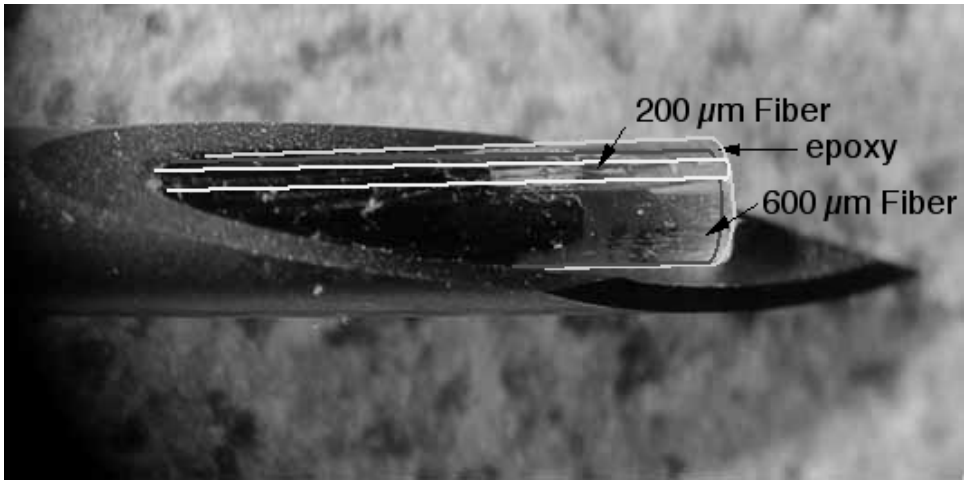


**Figure 4.** This is a picture of the needle probe with the fibers outlined for clarity. The fibers are fixed together in epoxy (also outlined)so that the faces are in the same plane. Tubing fixed to the fibers prevent the fibers from going any farther into the needle than shown. This gives consistent placement of the fibers in relation to the needle.

threaded through a piece of tubing which extends 1.5 m in length. The fibers are epoxied to this tubing at the probe end of the fibers. The purpose of the tubing is two-fold, to act as a depth stop for the fibers within the needle and to reduce any stresses on the fibers. An epoxy mold is made from two pieces of of teflon tubing with one fitting inside the other. The fibers are inserted into the smaller diameter ($\leq$ 800 $\mu$m inner diameter) 4 cm piece of tubing which is sliced lengthwise on one side of the tube. The mold tubing should come in contact with the 1.5 m tubing so that epoxy will bind it to the fibers. Care is taken to align the fiber faces such that they lie in the same plane, before the other piece of tubing, 5 cm in length, is sleaved over the sliced tubing, which completes the epoxy mold. The epoxy is inserted with a needle into the open end of the tube molding. The epoxy should overfill the 5 cm tube and run into the 1.5 m tubing which is positioned 3.2 cm from the end of the fibers. The 3.2 cm distance was chosen as the appropriate depth stop distance in a 1 inch 16 guage needle (figure 4). The epoxy is then cured and the outer tubing is removed from the smaller tubing. The slicing of the inner tube greatly reduces the risk of breaking a fiber, saving much aggravation. Once the inner tube is peeled away any excess epoxy can be cut away from the face of the fiber faces. If necessary, the fibers can then be polished together in the epoxy.

Finally, the two fiber bundles of 300 and 100 $\mu$m fibers are threaded through 1.5 m of teflon tubing for reinforcement and SMA connectors are fixed to the four loose fiber ends. The 300 $\mu$m fiber bundle is coupled to the 600 $\mu$m fiber and likewise the 100 $\mu$m fibers to the 200 $\mu$m fiber. Mode mixing loops are put into the 200 and 600 $\mu$m fibers with the 1/2 m of exposed fiber between the reinforcement tubing and the SMA connectors. Mode mixing produces an exit half-angle of 16.7° in air for both the 200 $\mu$m and 600 $\mu$m fibers.

The experimental set up uses two tungsten-halogen lamps (LS-1, Ocean Optics, Inc.) to illuminate the sample and a spectrometer (S2000, Ocean Optics, Inc.) to measure the light backscattered into the fiber. The experimental appartatus is shown in figure 2. A 100 and 300 $\mu$m fiber are each connected to a lamp. The remaining 100 and 300 $\mu$m fiber connect to the spectrometer, one at a time, depending on which size fiber is making a measurement. A 100 $\mu$m fiber connected to the spectrometer corresponds to a 200 $\mu$m fiber reflectance; and so, only the lamp with the 100 $\mu$m fiber is on. Likewise, a 300 $\mu$m fiber connected to the spectrometer corresponds to the 600 $\mu$m fiber reflectance in which case, the lamp with the 300 $\mu$m fiber is used. A 1 inch 16 gauge needle is filled with distilled water and then the epoxied fibers are inserted into the needle as far as allowed by the tubing depth stop. The needle and fibers are inserted into the tissue to be be measured. The water is used to aid in coupling of light between the tissue and the fiber. Measurements are recorded for both fiber sizes in air and water inside of a black box for subsequent normalization of further measurements in terms of percent reflectance. The Fresnel reflectance for the

glass/air and glass/water interfaces gives

$$R_{air} = \left(\frac{n_{air} - n_{fiber\,core}}{n_{air} + n_{fiber\,core}}\right)^2 = 3.5\%$$

and

$$R_{water} = \left(\frac{n_{water} - n_{fiber\,core}}{n_{water} + n_{fiber\,core}}\right)^2 = 0.22\%$$

respectively. The data is normalized using the 0.22% Fresnel reflectance signal in water,

$$R_{tissue} = \frac{S_{tissue} * 0.22\%}{S_{water}}$$

where $S_{tissue} and S_{water}$ are the raw signals of tissue and water respectively. The normalized reflectance is the same if $S_{water}$ is replaced by $S_{air}$ and 0.22% by 3.5% so that

$$R_{tissue} = \frac{S_{tissue} * 3.5\%}{S_{air}}$$

The Fresnel reflection for the fibers in tissue is not subtracted away. Initially a Fresnel reflection of 0.22% for water was expected, since water is used to couple light from the fiber into the tissue. However, the raw signals for *in vivo* tissue are smaller than from water thus a negative reflectance would result if a 0.22% is subtracted from $R_{tissue}$ to account for Fresnel reflections. Data is recorded in 0.75 nm increments over the range of 400–1000 nm. Note, data is presented over the range of 500–800 nm since the noise dominates outside this range.

## 3. RESULTS

The sized-fiber device has been used to measure porcine tissues *in vitro* and *in vivo*. The device gives consistent measurements of tissues for both size of fibers. Figure 5 shows the typical variation for *in vitro* reflectance spectra for kidney. The two locations are expected to be different, since the kidney is not homogeneous. In figure 6, the reflectance from the 600 $\mu$m Y-fiber is plotted against the reflectance of the 200 $\mu$m Y-fiber for the 5 measurements in each location at a wavelength of 500 nm. Also, *in vivo* reflectances of the kidney are included on figure 6 which are notably smaller than the *in vitro* kidney. This was to be expected since more blood is present in live tissue. Moreover, the *in vitro* kidney was removed nearly a week prior to the time of measurement and may have dehydrated changing the index of refraction for the tissue. However, the sized-fiber technique demonstrates a high degree of sensitivity in measuring small reflectance signals of the *in vivo* tissue.

Furthermore, the sensitivity of the device during *in vivo* measurements demonstrate that the spectra noticibly changes as the needle and fibers pass from one tissue to the next. Figure 7 shows typical spectra of different tissues with both fiber sizes. The signal in real time remains quite constant, then will quickly change as the device passes form one tissue to the next. This indicates the the device returns information about the tissue that is immediately at the probe face, though the effect has not been fully quantified.

## 4. DISCUSSION

Although the spectra of tissue are fascinating, the sized-fiber method still lacks a simple algorithm to convert the reflectance measurements reported here into more useful information of the absorption and reduced scattering properties of the tissue.

Previous methods of sized-fiber spectroscopy did not achieve the high degree of sensitivity shown here. Using Y-fibers has eliminated problems associated with optical alignment and have enhanced the ruggedness of the device. Requiring only a couple white light lamps, some optical fibers bundled together, and a detector or two, means the device is quite compact. The Y-fiber greatly reduces the cost of this technology by eliminating the need for many pricey optical components and equipment such as lasers, beam splitters, polarizers, optical choppers, and lock-in amplifiers used in previous methods of sized-fiber spectroscopy. Yet, replacing all the aforementioned equipment with a couple Y-fibers, greatly improves the signal to noise ratio making the *in vivo* measurements possible. The Y-fiber device is simple, rugged, compact, relatively inexpensive, and sensitive.

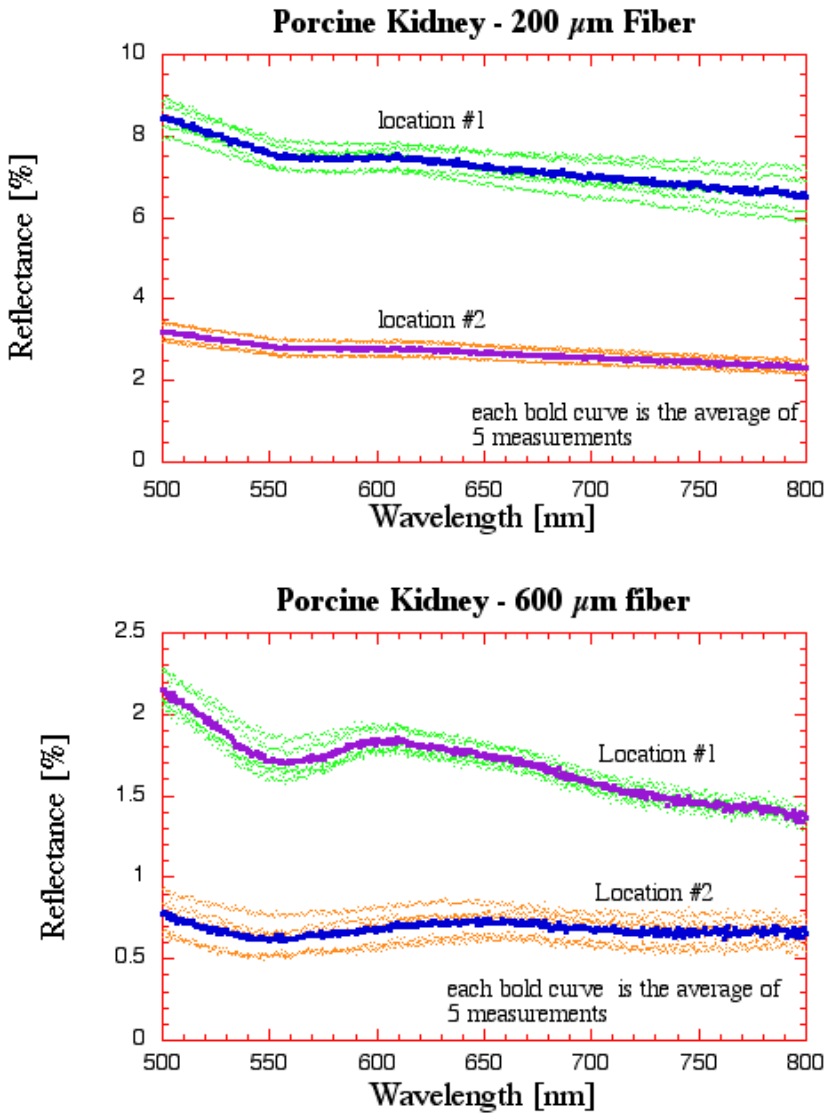


**Figure 5.** These typical *in vitro* spectra of kidney using a 200 $\mu$m and 600 $\mu$m fiber in a porcine kidney at two locations. Repeated experiments at each location show the measurement to measurement variation of signal. Since the kidney is not a homogeneous organ, it is not expected that the spectra be the same throughout the organ, and significant differences in reflectance were measured at the different locations.

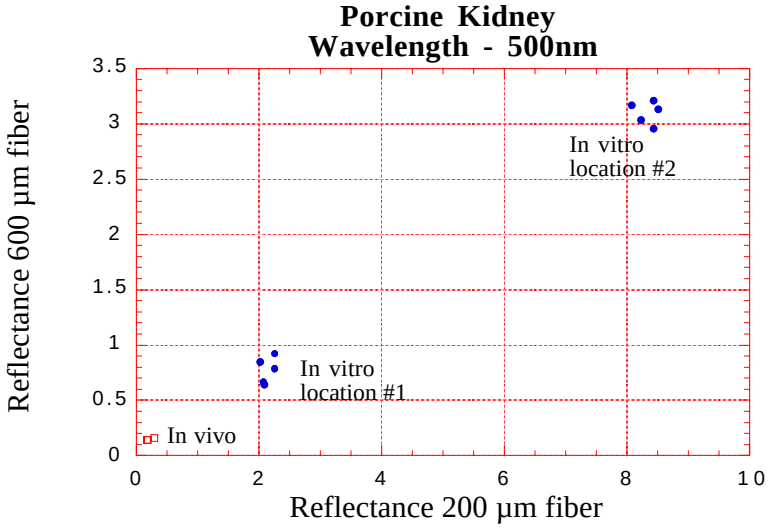


**Figure 6.** The measured reflectance of a 600 $\mu$m fiber versus a 200 $\mu$m fiber for *in vitro* measurements in two locations as well as for *in vivo* measurements of kidney. Expectedly, the *in vivo* reflectance is much smaller due to the presence of blood and other possible factors such as hydration levels.

## 5. ACKNOWLEDGMENTS

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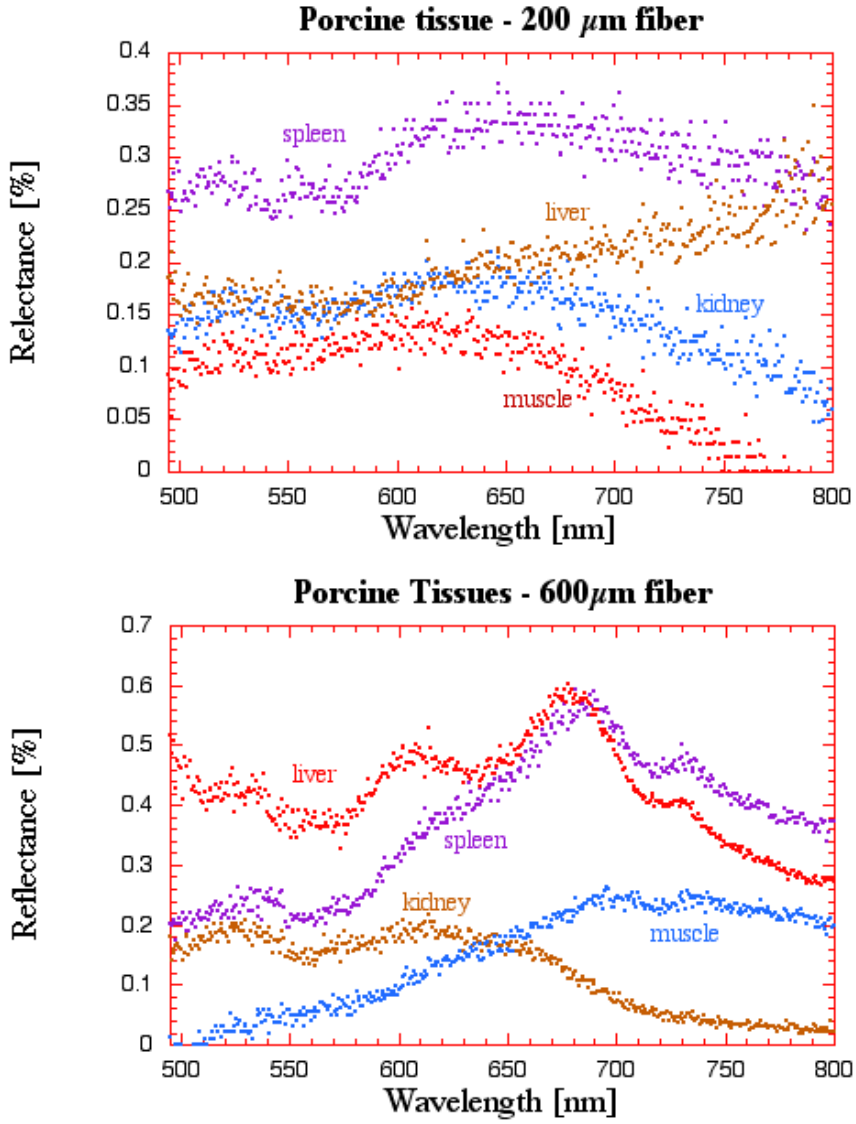


**Figure 7.** These typical *in vivo* spectra of tissues are stable as the fibers are held in place.