

Surgical imaging catheter for confocal microendoscopy with advanced contrast delivery and focus systems

Anthony A. Tanbakuchi^a, Andrew R. Rouse^b, Josh A. Udovich^a and Arthur F. Gmitro^{a,b}

^aOptical Sciences Center, University of Arizona, Tucson, Arizona;

^bDepartment of Radiology, University of Arizona, Tucson, Arizona

ABSTRACT

We present a laparoscope for fluorescence confocal microendoscopy specifically designed for microscopic imaging during diagnostic laparoscopic surgery. The catheter consists of a disposable rigid distal tip which houses a flexible microendoscope and dye channel. The laparoscopic tip is a small disposable polycarbonate sheath containing two inner lumens with a glass window on the distal end. The sheath outer diameter suitable for use in a 5mm trocar. The smaller inner lumen provides a channel for delivering fluorescent contrast agents to the tissue through a 200um hole in the glass window. On the proximal end, the smaller lumen is coupled to a computer controlled fluid delivery system that controls the amount of contrast agent dispensed onto the tissue down to a fraction of a micro liter. The main lumen houses the microendoscope. The microendoscope incorporates a computer-controlled focus mechanism that can quickly and accurately focus while correcting for hysteresis. This fluorescence confocal micro-laparoscope will be tested in a small-scale clinical trial on women undergoing oophorectomy in the near future.

Keywords: Confocal, microendoscope, Imaging catheter, Laparoscopic surgery, Ovarian cancer

1. OVARIAN CANCER

In 2005 the National Cancer Institute estimated that over 16,200 women died of ovarian cancer. During the same year, more than 22,200 were newly diagnosed with the disease. For those diagnosed, only 30 to 45 percent will survive five years.^{1,2} The survival rate for ovarian cancer is poor because the disease frequently has no obvious signs until it has reached an advanced stage. Only 19 percent of all cases are diagnosed during the early localized stages of the disease; most of the population are diagnosed during the later stages when the treatment is expensive and generally unsuccessful due to the high rate of recurrence.

For the female population at large, most are at a low risk of acquiring ovarian cancer. It is estimated that approximately 1 in 58 newborn females (1.7 percent lifetime risk) will develop ovarian cancer in their lifetime.³ However, the lifetime risk for the disease increases by two fold if the individual has one first or second-degree relative with the disease. With two first-degree relatives the risk increases by 25x (a 40 percent lifetime risk).³⁻⁶

For the subgroups of women with increased risk, few options exist to allow early detection of the disease. The NIH 1994 consensus stated that there is no single acceptable screening test for ovarian cancer and no evidence that combining the available screening tests — CA125, transvaginal ultrasound, and pelvic exam — has an acceptable sensitivity and specificity.⁷ Ovarian cancer is thought to metastasize early in the course of the disease and many experts believe that a different biology occurs during Stage I cancer as compared to the later stages. In Stage I, it is thought that the cancer often metastasizes before a lesion in the ovary becomes grossly visible. Without a viable method for early detection of the disease, most women at high risk are provided the option of prophylactic oophorectomy. Preemptive oophorectomies have the negative side effects of sterility and loss of natural hormone production. Hormone replacement is typically not an option for this subgroup since they are also at an increased risk for breast cancer and hormone replacement can further increase this risk.

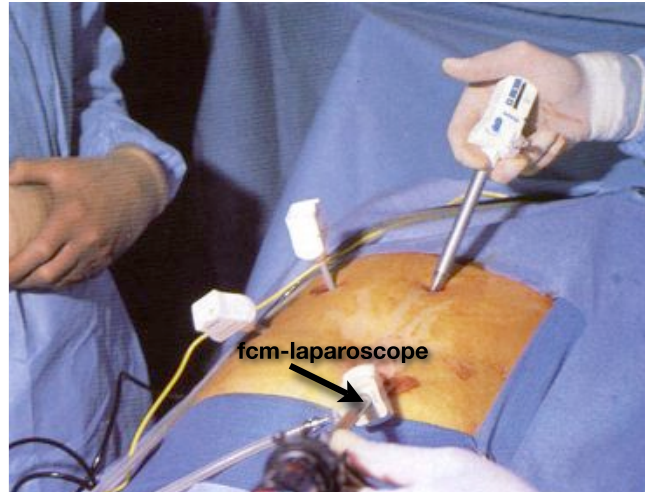


Figure 1. Illustration of how a fluorescence confocal micro-laparoscope would be used during a minimally invasive surgery to microscopically image the ovaries in real-time. (photo: Zucker⁸)

1.1. Diagnostic tool for the early detection of ovarian cancer

To improve the early detection of ovarian cancer for women at high risk, we propose a screening tool with the capability of diagnosing microscopic cancers or areas of abnormality in the ovary using minimally invasive surgical access. To accomplish this, we have developed a fluorescence confocal micro-laparoscopic — the *fcm-laparoscope*. A surgeon can use this imaging tool during a laparoscopic procedure (as illustrated in Figure 1) to inspect the ovaries via real-time in-vivo optical biopsy.

1.2. Screening tool requirements

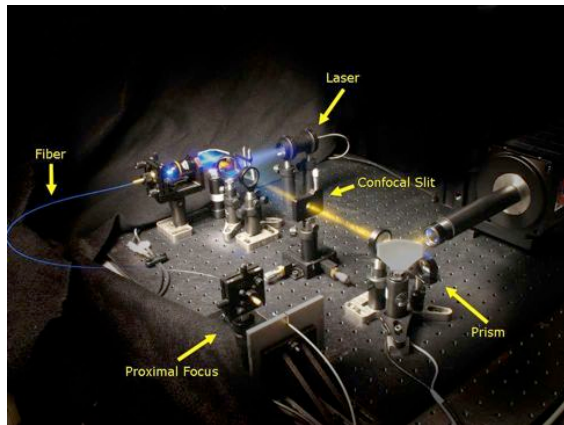
To create an effective screening tool for ovarian cancer we have concluded the following five requirements : (1) The device should provide real-time in-vivo confocal microscopic images, which have been previously shown to be effective in differentiating normal from abnormal ovarian tissue.⁹ (2) The device components in contact with the patient should be disposable and readily sterilizable. (3) The device should be similar in form to other laparoscopes and compatible with trocar's commonly used during oophorectomies that are capable of accepting devices 5mm or smaller.⁸ (4) The device should be able to quickly focus onto the ovaries to minimize the procedure time. (5) The device should have an integrated dye delivery system that is capable of tightly controlling the amount of contrast agent delivered to the field of view. The volumes must be controlled due to the cytotoxicity of contrast agents.

In the following sections the fcm-laparoscope is described in conjunction with a discussion of how each of the above requirements has been satisfied.

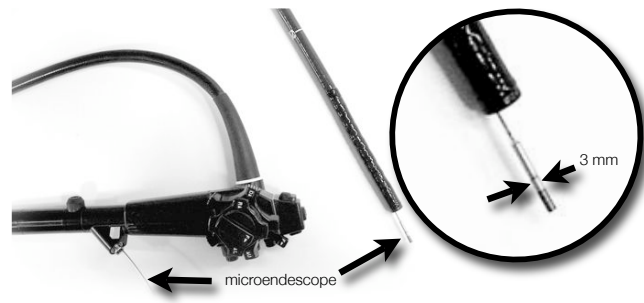
2. EXISTING WORK

We have previously shown that a confocal microendoscope with a fiber-optic imaging bundle can provide a flexible imaging tool for accessing small spaces in the body.⁹⁻¹¹ This instrument is clinically feasible for performing optical biopsies through it's miniaturized microendoscope catheter (Figure 2b) and external scanning system. The system developed in our lab has a $430\mu\text{m}$ field of view with a lateral resolution (x and y) of $3.0\mu\text{m}$ and axial resolution (z) of $30\mu\text{m}$. In vitro and in vivo images from human tissue specimens and animal models have been shown to provide adequate performance for microscopic evaluation of cells.¹¹ Figure 3 illustrates the distinct difference

Further author information: (Send correspondence to A.A.T.)
 A.A.T.: E-mail: kuchi@u.arizona.edu, Telephone: 1 520 626 4500
 A.R.R.: E-mail: rouse@u.arizona.edu, Telephone: 1 520 626 8352
 J.A.U.: E-mail: judovich@optics.arizona.edu, Telephone: 1 520 626 8352
 A.F.G.: E-mail: gmitro@radiology.arizona.edu, Telephone: 1 520 626 4720

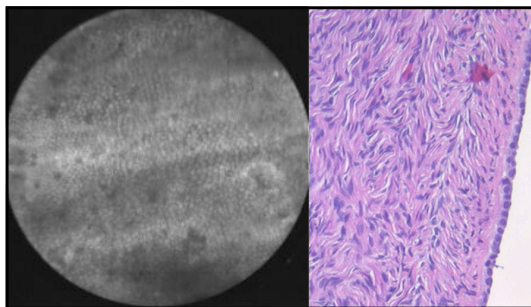


(a) Mobile slit-scanning confocal system.

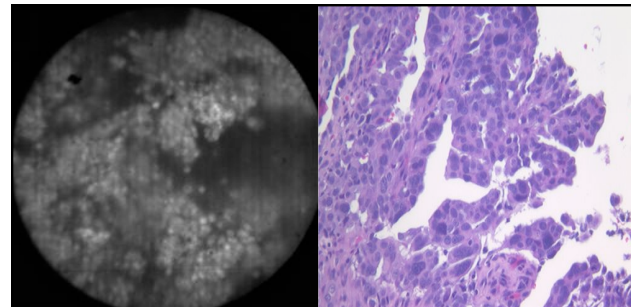


(b) Microendoscope (3mm diameter) fed through the therapeutic port of a colonoscope. Proximal end connects to scanning system at left.

Figure 2. Existing slit-scanning confocal microendoscope system.



(a) Normal ovary.



(b) Abnormal ovary.

Figure 3. Confocal microendoscope images illustrating the ability to differentiate between (a) normal and (b) abnormal ovarian tissue. Fresh ex-vivo tissue was imaged using a topical application of the contrast agent acridine orange. Left images are from the confocal microendoscope, right images are standard H&E stained histology slides.

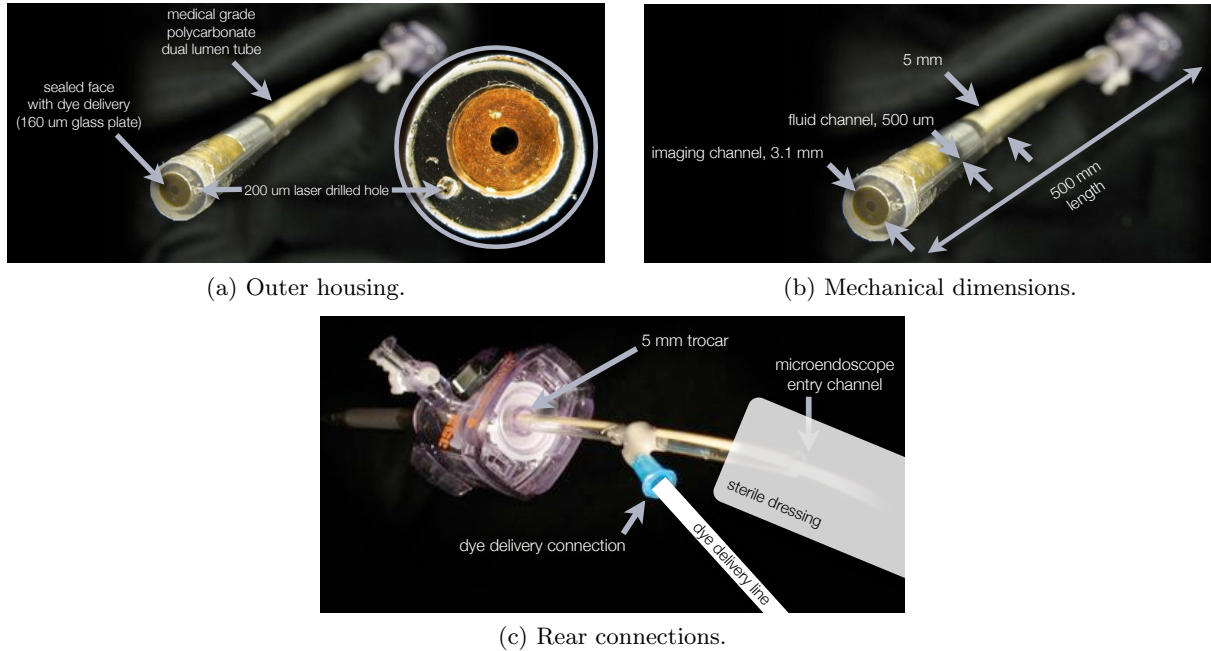


Figure 4. Physical characteristics of the fcm-laparoscope.

between normal and abnormal ovary.* To date, we have built a database of fluorescence confocal microendoscope images of ovaries from 38 subjects that include 239 normal images and 103 images from histologically verified ovarian cancer. From these images, it has become apparent that identification of pathology is possible.

In addition to providing adequate microscopic imaging performance, the system runs in real-time and has multi-spectral capabilities. Its slit-scanning design allows high-speed live video display to the surgeon at 30 frames per second. We have shown that a multi-spectral confocal microendoscope for in-vivo use has the potential to provide expanded diagnostic information to the surgeon. Such a system could be used in conjunction with multiple targeted contrast agents to increase the diagnostic information.¹² We have most recently developed a new generation of this instrument illustrated in Figure 2a that is mobile, compact, and suitable for clinical trials.[†]

3. FLUORESCENCE CONFOCAL MICRO-LAPAROSCOPE

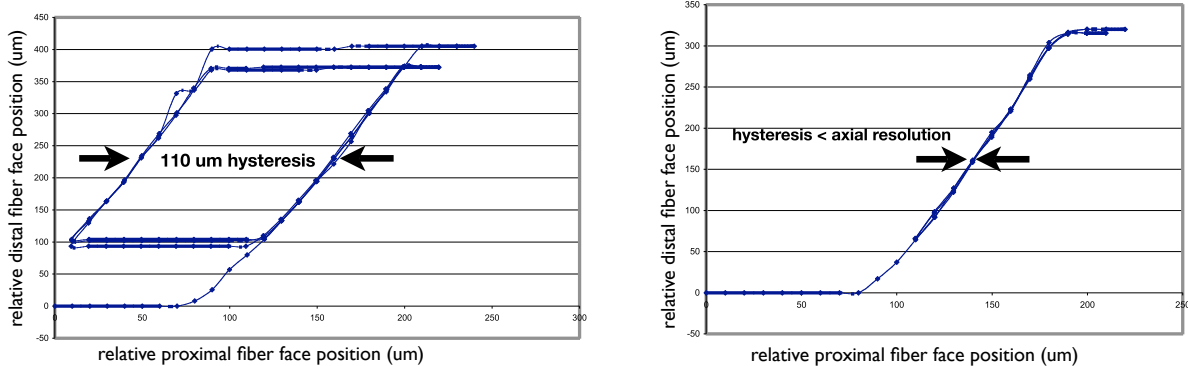
To make our confocal microendoscope amenable to laparoscopic inspection of the ovaries, we have further developed this device into a fluorescence confocal micro-laparoscope with fast focus and dye delivery systems. In the following subsections we describe the new device.

3.1. Mechanical specifications

To make our device feasible for use during clinical studies, we have engineered it so the patient comes into contact only with a rigid medical grade polycarbonate sheath that contains the existing confocal-microendoscope and dye delivery systems. This approach has three key advantages: (1) The existing confocal microendoscope can be used without redesign. (2) Only the outer polycarbonate housing needs to be sterilized. (3) The sheath is inexpensive and disposable.

*All tissue used in this research were collected in accordance with protocols approved by the Institutional Review Board of the University of Arizona.

[†]For further details on our new clinical system, see related paper in the same proceedings entitled *Design of an in-vivo multi-spectral confocal microendoscope for clinical trials*, paper number 6082-05.



(a) Inherent hysteresis of manual focus microendoscope. (b) Results with computer controlled focusing. Hysteresis is effectively corrected.

Figure 5. Focusing system performance.

The sheath is illustrated in Figure 4. The polycarbonate sheath is a rigid dual lumen tube that contains (1) a main imaging channel that accepts the 3mm microendoscope and (2) a fluid delivery channel. The 3.1mm imaging channel is fully sealed from the patient on the distal face by a $160\mu\text{m}$ glass cover slip and a sterile dressing on the proximal end. The $500\mu\text{m}$ diameter dye delivery channel has access to the tissue via a $200\mu\text{m}$ hole — this allows delivery of the contrast agent at the edge of the microendoscope’s field of view and the wicking action at the cover slip-tissue interface delivers dye over the imaging area. The polycarbonate sheath is 500mm in length and just under 5mm in diameter.

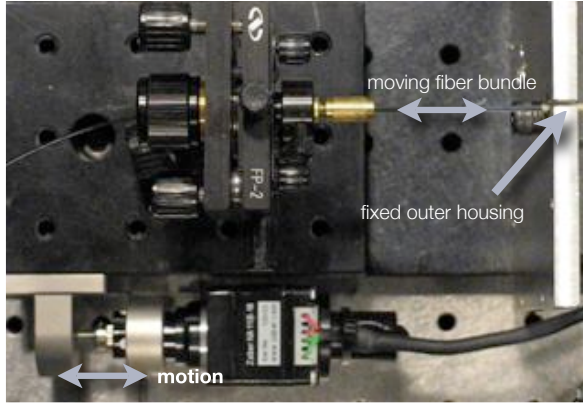
The polycarbonate sheath with glass face and dye connection is most suitably sterilized using ETO sterilization. Autoclaving is not a suitable technique because the high temperatures involved can soften the polycarbonate. Newer sterilization techniques such as STERRAD 50 are also not recommended because of concerns that the sterilizing chemical agents may not adequately invade the small lumen. However, twelve hours of ETO treatment sterilizes the device without any negative side effects.

3.2. Focus system

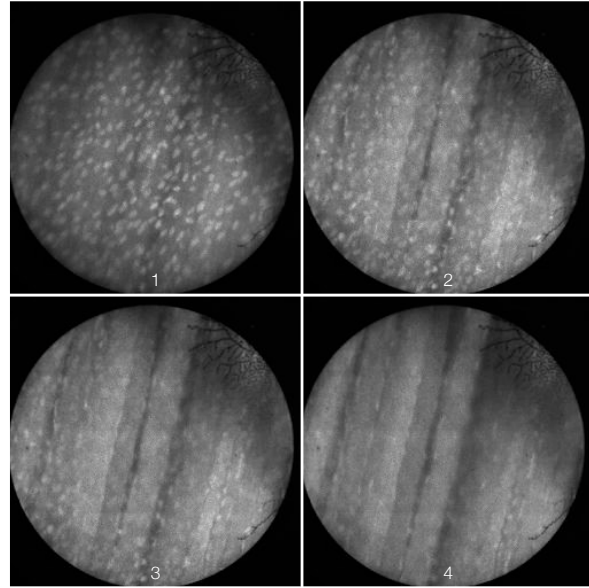
Our confocal microendoscope catheter has the ability to focus at various depths in tissue via a mechanism that is similar to a bicycle break cable. The microendoscope has a coherent fiber bundle encased in a outer tubing. At the distal end a small objective lens is mounted to the fixed outer tubing and the coherent fiber bundle is free to move inside. The tissue is placed in contact with the cover slip and as the distal fiber face position is translated a corresponding change in focus is achieved. Focus and axial depth scanning are achieved by moving the proximal end of the coherent fiber bundle — achieved originally via a manual micrometer — that results in movement of the distal fiber face.

The inherent problems with the manual micrometer system are: (1) it is slow and (2) movement of the proximal fiber does not directly relate to movement of the distal face of the fiber. Figure 5a illustrates the inherent hysteresis in the manual focusing system. On the x-axis, the proximal fiber face position is plotted against the distal fiber face position. It is clear that as the proximal fiber is moved, an inherent repeatable $110\mu\text{m}$ hysteresis must be overcome before any proximal movement is achieved.

To correct the two problems with the manual focus system, we developed a computer-controlled high speed positioning system shown in Figure 6a. By programming the motor to move the proximal fiber such that the distal face always approaches the desired position in the same direction, with a movement at least as large as the inherent hysteresis, we were able to achieve positioning accuracy that was an order of magnitude better than our system’s axial resolution. Figure 5b illustrates the repeatability of the new focusing configuration. The new motorized system allows focusing in increments as fine or as coarse as desired in less than a second. Focusing in $50\mu\text{m}$ increments from the epithelial layer into a muscle layer in a mouse peritoneal wall is illustrated in Figure 6b.



(a) High speed computerized focus motor that overcomes the inherent microendoscope hysteresis.



(b) Image sequence illustrating focus accuracy. Sequence shows the process of incrementally focusing from the epithelial layer down to the muscle layer of sample tissue in $50\mu\text{m}$ increments.

Figure 6. Focusing system.

3.3. Dye delivery system

The images in Figure 3 were stained with acridine orange, which is a vital nucleic acid fluorescent dye that intercalates with DNA and RNA. This contrast agent provides the selective binding and fluorescent emission signal generating useful image contrast. Fluorescent confocal imaging relies on contrast agents provide useful images for diagnosis. Cocktails of targeted contrast agents with multiple excitation wavelengths labeled to different components of the cell are an exciting application for the multi-spectral imaging of the confocal microendoscope. However, in-vivo use of contrast agents is limited by problems of cytotoxicity, potential mutagenicity and tumorigenicity. We do not believe that cytotoxicity is a major concern because imaging can be no more cytotoxic than tissue extraction biopsy, if the dye is delivered to the localized imaging site. However, questions concerning the mutagenic potential of the specific contrast agents used must be addressed and we have animal studies under way to study these issues for the dyes we are using.

To provide the localized dye delivery, we have developed a system to deliver controlled quantities of dye to the micro-laparoscope's field of view. This is accomplished by using a computer controlled dye pump that can dispense quantities of fluid in units of 100 nano liters or more. The dye is pushed down the polycarbonate dye channel and out the $200\mu\text{m}$ fluid delivery hole at the distal tip. Figure 7 shows an example of acridine orange being delivered via the operator actuating a foot switch. The process of controlled dye delivery happens in approximately one second.

When the distal tip of the fcm-laparoscope is in contact with tissue the ejected dye is drawn across the dye-tissue interface in to the field of view. Delivering a fixed single quantity of dye onto the peritoneal wall of a mouse in-vivo (just after the mouse was euthanized) is illustrated in the image sequences of Figure 8. In the initial image no dye has been delivered and the image is essentially black since there is no fluorescence signal. Once the dye has been delivered image contrast instantly develops and then stabilizes in a fraction of a second. The final image is at approximately one second after dye delivery when the epithelial layer is brought into focus.

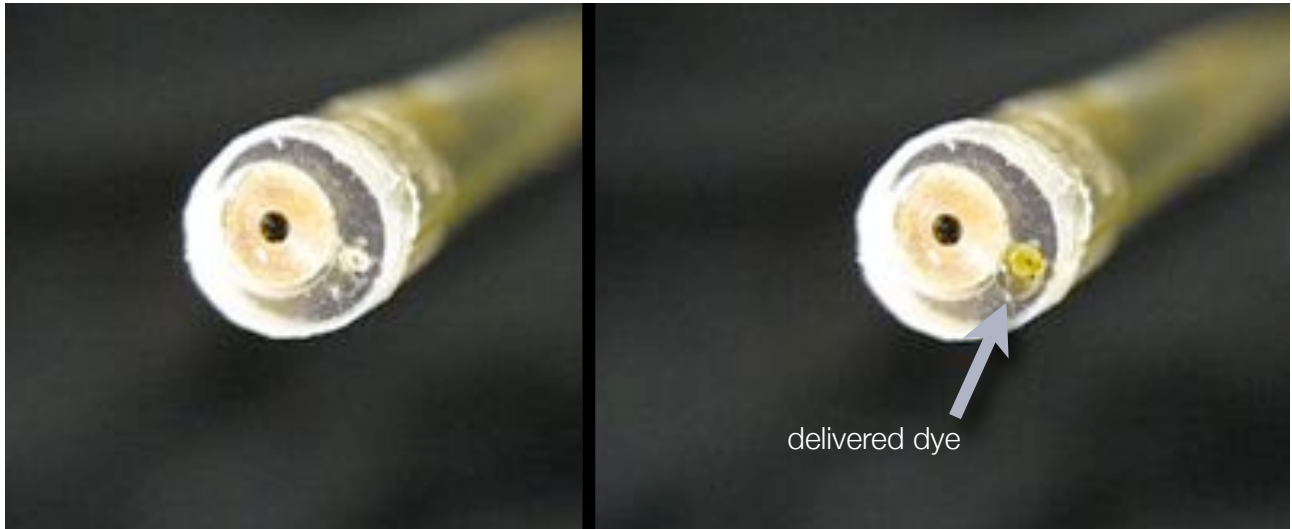


Figure 7. Laparoscope tip before and after dye ejection.

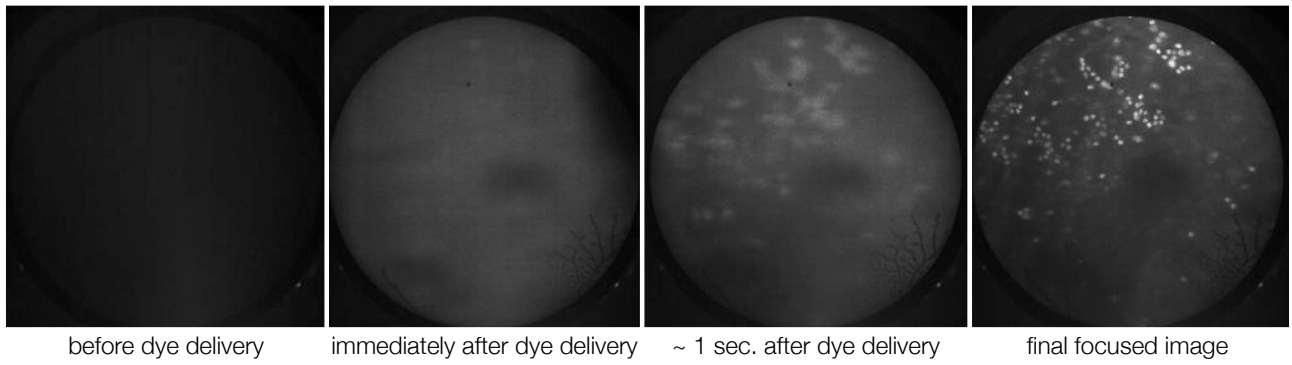


Figure 8. Dye delivery time sequence of a mouse's peritoneal wall. The sequence is approximately three seconds in length.

4. CONCLUSIONS AND FUTURE WORK

We have presented a fluorescence confocal micro-laparoscope that is capable of imaging tissue at the microscopic level. Although this tool has initially been targeted towards the use in screening high risk groups for ovarian cancer, it is widely applicable to the inspection of any laparoscopically accessible tissue surface where cancers develop. This tool is particularly well suited for laparoscopic surgery for 4 reasons: (1) It provides fluorescent confocal images with a $430\mu\text{m}$ field of view, $3\mu\text{m}$ transverse resolution, and $30\mu\text{m}$ axial resolution in real time at 30 frames per second. (2) The outer housing is disposable and sterilizable. (3) The system can be rapidly and accurately focused. (4) The contrast agents delivered locally in controlled volumes.

In the following year, this device will be tested in a small scale in-vivo human trial imaging the ovary. The study will consist of 10 women scheduled for oophorectomies. Prior to the removal of the ovaries, the fcm-laparoscope will be inserted into one of the 5mm trocar ports to access the ovaries. To inspect the surface of the ovaries they will be grasped with an atraumatic grasper and rotated such that the surface may be fully visualized with our system. The results of this human trial should help to validate our device design.

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