# WHITE PAPER: LASER LIGHT APPLICATIONS FOR EARLY CANCER DIAGNOSIS

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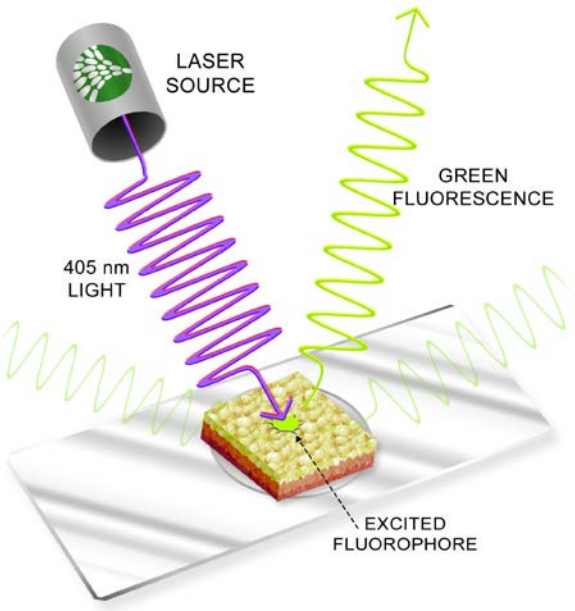
## Laser Light Applications for Early Cancer Diagnosis by Andrew Graveley

In recent years, the field of medical optics has been exploring a new approach to surgical endoscopic illumination using lasers, revealing many novel benefits. While halogen and xenon lamps (standard endoscopic light sources) may be sufficient for larger endoscopes, the demand for more extensive imaging through the body is driving endoscopes to smaller sizes. As cameras continue to reduce in size and improve image quality, the diameter of the illumination fiber must also be reduced. In this case, traditional light sources are inferior in their ability to output light through small fibers, while laser light can maintain sufficient light output through fibers nearing the size of a strand of hair. Furthermore, the use of a compound laser light source grants the ability to drastically alter the spectral response through the control of individual lasers without additional changes to the optical system. Such versatility has catalyzed the design and development of endoscopic laser light sources.

Of the many medical applications for endoscopes, cancer diagnosis is among the most prominent.




**Above: Green Fluorescence modeled in tissue with a 405nm excitation source**

The prognosis of cancer treatments is highly dependent on the stage at which the disease is diagnosed, thus prompting doctors to take proactive measures to find neoplasms before they mature or metastasize. While early stages are significantly easier to treat, they have traditionally been more difficult to diagnose. Visible light endoscopy, where a broad spectrum of 'white' light is used to illuminate the subject, is often limited to diagnosing later stages of carcinogenesis. Unfortunately, simply improving the image quality of current endoscopes will not suffice to improve the diagnosis; in reality this fault lies in the properties of the visible light spectrum and how it interacts on a small scale with human tissue. The differences in the altered tissue anatomy of neoplasms are too minimal for effective demarcation of the malignant tissue when examined under visible light. [1, 2] Such 'non-visible' growths require random-biopsies within the organ to be diagnosed internally, as a last recourse. This brute-force procedure creates additional cost to the patient, extended procedural time, and increased risk of inaccurate diagnosis due to large margins of error. [2]

**Selective Band Imaging**

These issues are overcome by the use of alternative optical techniques to visualize organs and tissue. Carcinogenic regions are known to form dense vasculature to supply the forming tumors with nutrients; one useful technique exploits this characteristic by enhancing the optical contrast of superficial vasculature. Selective band imaging (SBI) is a method in which tissue is illuminated with two narrow bandwidths of light wavelengths that correspond to the peaks in the absorption spectra of hemoglobin. This gives the appearance of darkened blood vessels and capillaries as they absorb the bandwidths of light used. The dark vasculature contrasts the lighter, more reflective surrounding tissue, allowing areas such as carcinomas to be revealed by a sort of high contrast imaging. The laser-based Hyperion technology developed by Nathaniel Group, now Ushio America, Inc., allows users to implement this technique as an optional SBI mode in addition to the standard white light mode. With this technology, the user can simply select the mode controlled by the firmware, and neither the user nor the manufacturer is required to change the optical hardware on the camera or light source. Using the available controls, the light source can be switched from standard imaging to diagnosis enhancing SBI. Additional benefits include less power consumption, less heat dissipated by the light source, and less heat concentrated in the optical components of the illumination device for equal or greater light output.

In a different approach, use of only the near-UV wavelengths is the basis of fluorescent imaging systems that can reveal visible and non-visible early stage cancers alike.[2] Fluorescence occurs when a molecule known as a *fluorophore* is excited by a certain wavelength of light before releasing the absorbed energy at a higher wavelength. The frequency of excitation light is specific to the fluorophore, as it is based on the chemical bonds that *resonate*, so to speak, with an optimal wavelength. Fluorescence imaging systems use near-UV light to excite exogenous fluorophores that unevenly accumulate in different tissues. The near-UV excitation wavelength must be filtered out before reaching the camera so only the longer emission light from the fluorophores is shown. This mechanism, which reveals an image based on a molecular concentration associated with abnormal tissue, is known as exogenous fluorescence diagnosis (EFD).

**Autofluorescence Imaging (AFI)**

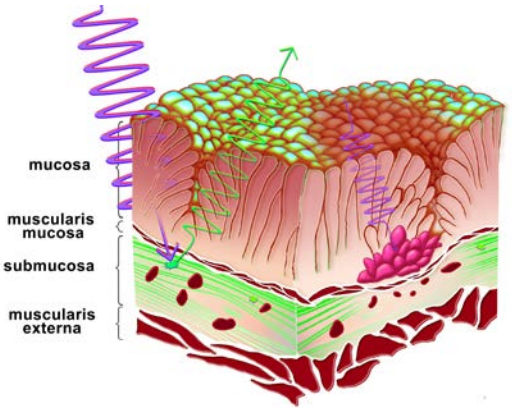
Under normal circumstances, tissue walls of the respiratory tract, digestive tract, and other organs will fluoresce under near-UV light.[3, 4] Consequently, several common molecules endogenous to epithelial tissue have been identified as fluorophores under near-UV light including collagen, elastin, keratin, NADH, and protoporphyrin.[3, 5] Research by Z. Huang and colleagues has shown that the primary source of fluorescence in the gastric wall is the high density of collagen in the submucosal layer, outputting green luminance when excited with a near-UV light source. In contrast, carcinomas in these tissues are known to show up dark in fluorescent imaging, allowing the cancerous cells to be demarcated.[4] The mechanism for this change is primarily anatomical; the cancer does not significantly alter the concentration of fluorophores, but rather the superficial mucosa layer of the epithelium thickens, thereby dampening the light transmission to and from the fluorescent collagen in the deeper submucosa. Fluorescence in cancerous tissue is further reduced through the light absorption from hemaglobin as the cancer increases localized bloodflow.[4, 6] In clinical application, autofluorescence poses minimal side effects to the patient, while significantly improving the diagnostic sensitivity when compared to white light endoscopy.[3,6,7,8] In a 2005

study by the Academic Medical Center of the Netherlands endoscopies were performed on 60 patients with Barrett's esophagus, a condition that can predispose to esophageal cancer.[7] During their procedures, white light endoscopies only revealed 63% of all early cancer diagnoses confirmed histologically. However, using AFI, they were able to catch 91% of the total early cancer diagnoses in patients with Barrett's esophagus. A later 2010 study of 71 patients also reported that autofluorescence imaging at 405nm provided better diagnostic precision for cancer and dysplasia than white light and other state of the art imaging technologies.[8] Nathaniel Group, now Ushio America, Inc., has developed a patent pending technology that utilizes this optimal wavelength for fluorescence imaging and can be selected by the user, without additional optical filtration.

With so much research in support of autofluorescence, it is surprising that it has not become a standard method for prescreening applicable cancers. The major limitation of AFI is its inability to distinguish abnormal tissue from areas of inflammed tissue, leading to low specificity and high rates of false positive diagnoses.[7] While AFI's high sensitivity and low specificity is acceptable from a safety standpoint, so many false positive diagnoses will lead to unnecessary costs in time and lab tests, as well as distress for the patient. Fortunately, with a simple technique AFI can be supplemented with the use of SBI to reduce the rate of false positives. In a study in which 20 patients with Barrett's esophagus were screened for high-grade dysplasia, physicians checked the results of the fluorescent imaging with selective band imaging, where lesions with normal border patterns and vasculature were considered to be inflammed tissue.[7] This reduced the false positive rate from 40% to 10%, of which a majority of the remaining false positives were low-grade displasia.[7] For a physician using the Hyperion videoendoscopic system, this comparison only requires the user to select the desired wavelength to examine a suspicous lesion using autofluorescence and then selective band imaging.

**A Promising Drug for Fluorescence**

To expand the fluorescence capabilities of medical diagnosis, many pharmaceuticals are on the market and under development to improve the diagnosis of disease. Cysview® (hexaminolevulinate hydrochloride), Hexvix™, Levulan®, and Metvix® (MAL) are some of the drugs approved worldwide for the use of EFD.[9, 10] These are an assortment of drugs derived from the endogenous molecule, 5-aminolevulinic acid (ALA).




**Above: Mechanixm of autofluorescence demarcation. Healty tissue fluoresces from the collagen found in the submucosa, while the enlarged mucosal layer on cancerous tissue reduces the light transmission and thus the fluorescence.**

As found in normal metabolic processes, ALA is a metabolic rate-limiting precursor to protoporphyrin IX (PpIX) that is fluorescent under near-UV light.[3] When ALA is administered either orally or topically, within 3-14 hours PpIX accumulates in carcinomas or other abnormal tissue in significantly higher concentration relative to surrounding healthy tissue.[9,11] This accumulation leads to the fluoresent positive identification of carcinomas or otherwise diseased tissue.

A major benefit of ALA over current drug therapies is its ability to metabolize within 48 hours, meaning a decrease in the time a patient must avoid light exposure in order to prevent skin irritation or damage. [10] In non-muscle invasive bladder cancer, an endoscopic surgical procedure known as a transurethral resection is a preferred treatment as it is minimally invasive, though it has shown to be less than reliable. As bladder cancer has an indistinct appearance under white light, portions of cancerous tissue are often overlooked by surgeons, leading to high recurrence rates. [11] However, in conjunction with ALA fluorescence diagnosis, the visual aid of tissue fluorescence during transurethral resection has been shown to reduce recurrence rates by 20%. [12] Similar improvements in other endoscopic procedures have been reported when implementing fluorescence diagnosis. Cervical premalignancies can be better identified and more accurately diagnosed, as tissue for biopsy can be better selected by fluorescence demarcation. [11]

**Choosing Between Exogenous and Autofluorescence**

While pharmaceuticals can be used for both diagnostics and treatment, it may not always be the best option when diagnosis is the preliminary concern. AFI and ALA-induced EFD both have very high sensitivity, with slight compromises in specificity. [7, 13, 4, 14] Considering the severe side effects of photosensitivity, added cost of a pharmaceutical, and prolonged prodcedures that are inherent with the use of EFD, autofluorescence may often be preferred for cancer screening. Although the side effect of photosensitivity may be mitigated with a topical application, this will complicate the procedure, and in some cases this may reduce the accuracy of the diagnosis. [13] While it was previously believed that AFI alone was not sufficient to provide a good image, with recent advances in endoscopic technology such as the improved light output of the Hyperion, additional fluorophores are no longer necessary.

**Porphyrin Derivatives, and Photodynamic Therapy**

Hematoporphyrin, known for its fluorescent properties, was the first ever chemical used for photodynamic therapy (PDT) when it was discovered to concentrate in cancerous tissue in the mouth. [15] This prompted investigation into the use of this fluorophore for demarcation of neoplasms and carcinomas. It was subsequently discovered that this chemical has photosensitizing effects to cells under high concentration. More recently, similar porphyrinoids such as Photofrin® (porphimer sodium) have been developed to more selectively accumulate at high enough concentrations to become phototoxic to the targeted carcinomas, without posing a risk to healthy tissue. Such advantages have made PDT increasingly popular as a targeted treatment for specific cancers or other diseases. Various drugs of this class have been approved for clinical use worldwide for PDT, and more advanced classes are currently under development. Photofrin, though currently the most popular drug for PDT, has the tradeoffs of requiring a high dose of light; slow onset of effectiveness, meaning it must be administered a day or more before treatment; and a clearance time of over four weeks, requiring patients to avoid bright light for an extended period of time. Although it is an effective, and especially prefered treatment for some cases, the improvements of newer drugs will make PDT more suitable for patients.

| Drug Family | Name | Dose (mg/kg) | irradiation λ | J/cm2 | Onset (hrs) | Clearance | Targeting |
|---|---|---|---|---|---|---|---|
| Porphyrin Precursor | Levulan® (ALA) | topical | 400-600 | 10 | 14-18 | 4 weeks | skin*1, esophageal, endobronchial, uterine, and brain cancer. |
| porphyrinoid | Photofrin® | 2 | 630 | 130-300 | 24-50 | 4-8 weeks | Barrett's esophagus*1, esophageal*1,endobronchial*1, bladder*2, cervical*2, and lung*2 cancers |
| | Foscan | 0.15 | 652 | 5-25 | 24-96 | 4-6 weeks | nasopharyngeal*, bile duct*, lung, esophageal, gastric, prostate, skin, and head & neck cancers |
| | Photochlor® | .15 | 665 | 150 | 24-48 | | Barrett's esophagus*, esophageal*, basal cell*, and lung* cancers |
| | Laserphyrin® | .5-3.5 | 664 | 150 | 4 | 3-7 days | Lung*2, head & neck*, and liver* cancers |
| | Visudyne® | .3 | 690 | 50 | 3-5 | 24 hrs | Subfoveal CNV*2, psoriasis*, and skin* cancers |
| Purpurin | Purlytin® | 1.2 | 660 | 200 | 24 | | Metastatic breast adenocarcinoma*, basal cell carcinoma*, and Kaposi's sarcoma |
| texaphyrin | Antrin® | .6-7.2 | 732 | 150 | 3 | | cervical*2, prostate*2, breast*, skin*, Kaposi's sarcoma*, and brain cancers |
| | Tookad® | 2-4 | 763 | 360 | .5 | 1.3 hrs | Prostate cancer* |
| | Photosens® | .5-.8 | 672 | 150 | 24-72 | | squamous cell*, breast*, oropharyngeal*, lung*, and larynx* cancers |

* in clincial trial, *1 approved in US, *2approved outside US [9],[10]

**Table: Drugs either currently approved or undergoing clinical trials for the use of photodynamic diagnosis, compared by important parameters of their effectiveness and side effects. The irradiation is the optimal light source wavelength to activate the drug, J/cm² is the energy of the light, onset is the time the drug must be administered before therapy, and clearance is the time it takes to be removed from the patient's system - for which the patient must avoid light exposure. Currently, the most commonly used drugs have compatible irradiation specifications for use with the Hyperion, including Levulan® and Photofrin®. Clearance data is not available for all drugs that have not completed clinical trials.**

**Implementing new technologies**

Nathaniel Group, now Ushio America, Inc., has pioneered the technology of endoscopic laser light sources in their Hyperion product line. They are among the first to deliver a high performance illuminator capable of multiple imaging modalities with improved power output and without the use of optical filters. The Hyperion empowers users to select the imaging mode, whether it is near-UV fluorescence, selective band imaging, or standard white light. The ability to easily switch between complementary SBI and fluorescence imaging modalities will soon allow doctors to get more accurate results from endoscopic cancer screening, and optimize cancer prevention. With the development of new drugs for photodynamic therapy, the same light sources used for diagnostic imaging will even be adapted for future cancer treatment thanks to the versatility of laser light.

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