

Photonic early warning systems for cancer, biothreats

The Center for Photonics Technology (CPT) is using light in two separate efforts to detect germs and disease so that prevention or early treatment can save lives. In an effort to diagnose early-stage cancer – before it shows up on X-ray and other imaging methods – the Center has had promising results with its method of using laser spectroscopy to detect cancer in blood samples. The Center is also developing optical biosensors that can detect traces of the bacteria and viruses that cause diseases such as tularemia, anthrax, and plague.

Non-invasive cancer detection

Recent experiments using 61 human blood samples and more than 500 animal blood samples show that laser spectroscopy of serum may provide a reliable non-invasive cancer diagnosis technique, according to Anbo Wang, director of CPT.

The technique involves sending laser photons through a sample of serum and measuring the spectrum of the exiting beam. Raman scattering is created when the laser photons bounce off molecules in the sample, transferring energy to the sample molecules, and causing them to vibrate. The lower energy photon beam that exits the sample exhibits a frequency shift. The frequency shifts from different molecular vibrations generate a measurable spectrum, which differs according to the compound. When energy is absorbed instead of bounced, fluorescence is generated.

Several years ago, visiting scholar Bing Qi observed that the Raman spectrum generated by exciting serum beta carotene was measurably different in healthy people and those with cancer. Extending this work, CPT researchers Wang and Ahmad Safaai-Jazi recently studied the blood serum of 61 persons: 29 with stomach cancer and 31 cancer-free. Both fluorescent and Raman results were identified.

“The very small sample of 61 humans showed strong potential for the technique,” Wang said. “Our work with the animal blood also revealed meaningful results, but is inconclusive until we develop baseline information for each of the animal species whose serum we studied.” For the animal sample study, CPT collaborated with John Robertson and William Huckle of the Virginia-Maryland Regional College of Veterinary Medicine.

The team plans to further investigate the process, with a larger study group of humans where food intake before the test can be



© Dennis Kunkel Microscopy, Inc.



Detecting cancer and bacteria with photonics – Left: The Center for Photonics Technology (CPT) has tested human and animal blood for cancer by passing laser photons through the sample and measuring the frequency shift of the beam on the other side. The fluorescence is generated by the energy that is absorbed. Right: Research Scientist Kristie Cooper demonstrates how optical fibers are coated in making sensors that can detect *F. Tularensis* (top), a Class A biothreat.

controlled. “We believe our results show promise that, with a more controlled study, the reliability of this method can be improved significantly,” Wang said.

“We hope this leads to the ability to detect cancer before it has grown so large that permanent cures are difficult,” he added.

Detecting one of the most infectious organisms

Another early warning system under development relates to detecting small amounts of bioterrorism agents before they create widespread problems. CPT researchers have demonstrated the feasibility of using a coated photonic sensor to detect traces of the Class A bioterrorism agent *F. tularensis*, the bacterium that produces tularemia, or “Rabbit Fever.”

F. tularensis is one of the most infectious disease-causing bacteria known to medicine. As few as 10 organisms can cause disease in a person, compared to the 10,000 anthrax spores required to trigger human disease.

When contracted naturally, the disease has a 2 percent fatality rate, and responds to antibiotics. It is not transmitted between humans, but is considered a potential biothreat, because it is readily available and can mimic other diseases such as Legionnaire’s Disease, typhoid fever, and plague. Inhaled tularemia can produce severe, life-threatening pneumonia.

CPT is developing rapid, inexpensive field tests to detect *F. tularensis*, both in blood samples and in the environment. Their ultra-sensitive sensor is based on an optical fiber with a thin precursor film, then a molecule-thick coating of antibodies. When the bacteria are present, their antigens are attracted to the antibodies and cause the organism to attach to the coated fiber. When the bacteria attach, the change in evanescence (light that escapes through the fiber wall) can be measured.

“As light travels down the core of the optical fiber, an evanescent wave penetrates through the outside wall,” explained Kristie Cooper (Ph.D. ’99), the lead research scientist on the biosensor effort. “The wave will interact with the film that we put on the fiber and either the wavelength or the intensity will change. Based on the refractive index, we will get a loss, showing an upside down peak. If the index changes, the peak will shift back and forth,” she said.

“The result is basically positive, or negative,” she said. “If there is no change, there are no bacteria present; if there is a change, there are. The size of the change will help us determine the concentration.”

In order to measure the change, the team is developing a refractometry transduction technique sensitive to index changes on the order of 0.0001 RIU. The sensors are expected to detect changes at the picometer (pM, 1×10^{-12} meter) to femtometer (fM, 1×10^{-15} meter) levels.

The keys to developing such sensitive sensors are the optical structure and the thin precursor film, according to Cooper. The precursor film is used to control the refraction index and to provide a cushion of organic layer over the silicon so that the receptor antibodies will attach. “We need to match carefully the film thickness and refractive index, cladding thickness, and grating period and length,” she said.

The precursor film is several layers deep, each layer only a molecule thick. The layers are attached to the fiber through a

layer-by-layer self assembly process. For each layer, the fiber is immersed in a solution of charged particles with the opposite charge. The molecules in the solution align in one direction and attach to the fiber through a chemical reaction. “The precursor film needs to be in a specific refractive index window,” Cooper said. “It needs to be a precise thickness. Our self-assembly technique is very handy for this, since it is self-limiting.”

CPT is collaborating with Thomas Inzana and Jane Duncan of the Virginia-Maryland Regional College of Veterinary Medicine on the project, which is sponsored by the U.S. Army and Virginia Tech’s Institute for Critical Technology and Applied Science (ICTAS).

The vet school team is working to develop a vaccine and is providing CPT with antibodies to the capsule, or the outermost layer of the bacterium. “They are developing high-affinity capsule-specific antibodies, which should provide better results than the conventional antibodies to whole organisms,” Cooper said. “The whole-organism antibodies are subject to cross-reactivity with similar antigens on other bacterial species,” she explained.

The sensors are designed for inexpensive assembly and can easily be developed for antibodies to many other organisms, Cooper said.



As an undergraduate researcher, Jeremy Michel calibrates the film layer for adding layers of self-assembled coatings on optical fibers. The molecule-thick layers are part of an optical fiber-based sensor for viruses and bacteria. The outer layer contains antibodies to the bioagent being tested. When viruses or bacteria attach to the sensor, the index of refraction of the fiber changes. The coatings are clear, but orange color is added for training and calibration.