

Optical microresonators: label-free detection down to single viral pathogens

Frank Vollmer

The constructive interference of coherent light inside microsphere cavities enables robust, ultrasensitive biosensors as observed from resonance-frequency shifts.

Rapid, sensitive, and specific detection of biological pathogens is of great importance for diagnosis of infectious disease, point-of-care (POC) testing, and biological security. All existing detection and identification schemes for relevant pathogens rely on amplification and labeling of molecules using conventional molecular biochemical methods, i.e. enzyme-linked or radio-immunosorbent assays, polymerase chain reaction (PCR), Western blots, and so forth, that call for skilled medical personnel and expensive laboratory equipment. These requirements prohibit their use in low-resource settings, such as for home healthcare applications or as fieldable instruments. To remedy this situation, we are developing a highly sensitive, miniaturized, optical-sensing component that allows detection of single pathogens or disease biomarkers without having to label them.¹

Our biosensing component derives its unprecedented sensitivity from the use of microsphere optical resonators. The optical resonance is created by launching and confining coherent light inside the microsphere, where it interferes constructively due to total-internal reflection: see Figure 1(A). Because optical resonators are almost immune to damping in a liquid, we are able to detect binding of single influenza A virus particles from discrete resonance frequency changes,² which obviates the need for chemical or fluorescent labeling.

Optical detection of single virus particles² was achieved by probing with a light field that is confined by the microsphere. Figure 1A exemplifies the device concept. The microsphere (~50 μ m radius and held on a stem) is brought in contact with a tapered optical fiber. On mechanical contact, light couples from the tapered fiber region to the microsphere where it then remains confined due to total internal reflection. The trapped light

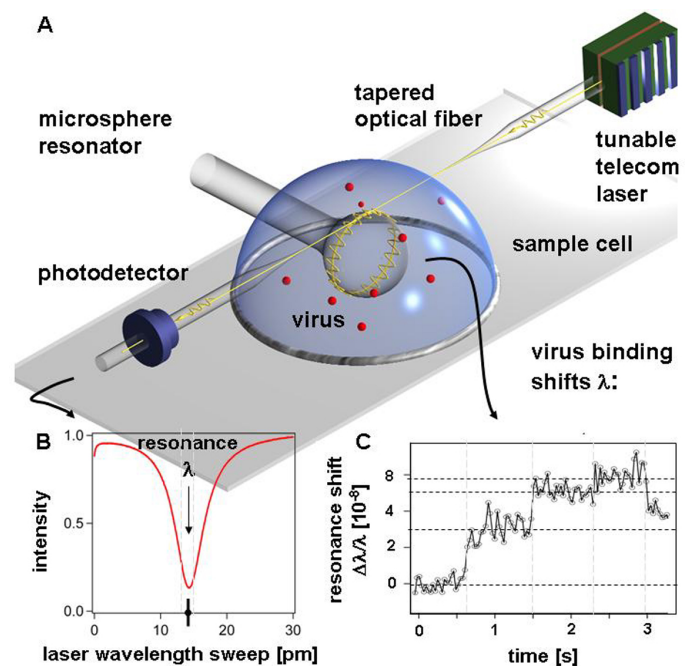


Figure 1. Optical resonance pathogen detector. (A) A wavelength-tunable telecom laser delivers near-IR light (shown in yellow) through the taper of an optical fiber to a glass microsphere. (B) At a specific resonance wavelength, the light couples to the microsphere and then no longer reaches the photodetector. A drop in the transmitted intensity is recorded, the minimum of which corresponds to the exact resonance wavelength, λ . (C) Ultrasensitive detection of single influenza A virus particles is demonstrated by monitoring changes ($\Delta\lambda$) of the resonance wavelength, and detecting discrete steps in the wavelength-shift signal as virus nanoparticles bind to the microsphere surface.

orbits on a circular trajectory close to the equator of the sphere (light wave shown in yellow), performing several thousand

Continued on next page

roundtrips at steady state. Such light recirculation provides the basis for extreme signal enhancement, down to single virus particles.

The measurement principle works as follows. A chip-scale telecom laser is tuned in wavelength so that the light returns in phase for each roundtrip inside the microsphere, forming an optical resonance. The resonance wavelength is precisely identified by monitoring transmission through the optical fiber as the wavelength of the laser is tuned. As the laser wavelength matches the resonance wavelength of the microsphere, the light couples from the tapered fiber to the microsphere. Because it no longer reaches the photodetector, a drop in intensity is recorded, the minimum of which corresponds to the exact resonance wavelength: see Figure 1(B). Once identified, the resonance wavelength changes further only if one or additional viral particles bind to the microsphere surface. Virus-binding events are monitored in real time by continuously tracking the resonance-wavelength shift: see Figure 1(C).

Detection of single particles relies on the ability to discriminate the wavelength-shift signal against background noise, a task that requires narrow linewidth, high-quality (Q) optical resonance and the use of nanoparticles that produce an observable resonance shift. Unfortunately, most important biological pathogens are virus nanoparticles in the 50–1000nm size range, and discerning the resulting wavelength-shift signal is extremely challenging (its magnitude scales inversely with the virus size to the third power). Fortunately, the magnitude of the wavelength-shift signal can be sufficiently enhanced by making the microsphere resonator smaller. We confirmed a reactive sensing mechanism with inverse dependence on mode volume in experiments with virus-sized polystyrene nanoparticles.² By comparing the electromagnetic theory for this reactive effect with experiments, we can determine the size ($\sim 100\text{nm}$) and mass ($\sim 5.2 \times 10^{-16}$ grams) of a bound influenza A virion directly from the optimal resonance wavelength shift.

As with any small-area detector, the speed of nanoparticle detection is limited by the time it takes to deliver the nanoparticle analyte (molecule of interest) to the sensing region. Even with the use of microfluidic flow for rapid sample delivery, close to the microresonator surface, detection timescales are limited by the diffusion time of the nanoparticle to the surface by random Brownian motion. Near the surface of the optical resonator, however, the nanoparticle is within the reach of the evanescent field. Consequently, at sufficient optical power, the nanoparticle is actively drawn toward the sensor surface by gradient forces,³ similar to those present in optical tweezers. Given sufficient binding sites, this trapping mechanism considerably increases the binding rate at extremely low particle concentrations (fM),

and we are able to speed up detection time by almost two orders of magnitude.³ In addition, the nanoparticle is drawn to the highest intensity region of the resonant light field where its presence produces the largest sensing signal (i.e., wavelength shift). In the absence of binding sites, the nanoparticles are still drawn toward the microsphere surface, but instead of binding, there are propelled around the microsphere resonator by radiation pressure.³ Within this orbital trap, radial stochastic motion is induced by thermal energy within the exponential-potential-well setup by the evanescent field, forcing a nanoparticle to visit the surface many times per micron during its circumnavigation. As a result, binding is essentially assured once the nanoparticle is pulled into this stochastic orbit. Surprisingly, the trapping threshold power for virus-sized particles is in the 10–100 μW range due to the buildup in intensity caused by the high Q of the optical resonator ($Q \sim 10^{5-6}$).

To summarize, we have developed an optical resonator biosensing platform for label-free single-virus detection. Resonant light fields in miniature microspheres are used for sensitive nanoparticle detection as well as for gradient-force-assisted delivery of the nanoparticle analyte to the region of highest sensitivity. Our biosensor prototype represents a standard technology component that may be integrated into other POC devices that similarly require high-sensitivity, label-free detection of viruses, other cellular pathogens (e.g., bacteria, fungi, protozoan cells), or pathogen-associated molecular components. Our future efforts will focus on further improving the sensitivity of our resonator platform in silicon-materials systems, and boosting the specificity for detection in complex fluids by simultaneously determining the size, shape, and affinity of a virus particle.

Financial support from the Wyss Institute and the Rowland Institute, both at Harvard University, is gratefully acknowledged.

Author Information

Frank Vollmer
Harvard University
Boston, MA

Frank Vollmer specializes in biofunctional photonics, which involves inventing, constructing, and using light fields to study biological systems. He is currently a scholar-in-residence at the newly formed Wyss Institute for Biologically Inspired Engineering at Harvard University. He has contributed several articles to

Continued on next page

SPIE Proceedings, and has been an invited speaker at a number of SPIE conferences

References

1. F. Vollmer and S. Arnold, *Whispering gallery mode biosensing: label-free detection down to single molecules*, **Nat. Methods** **5** (7), pp. 591–596, 2007. doi:10.138/nmeth.1221
2. F. Vollmer, S. Arnold, and D. Keng, *Single virus detection from the reactive shift of a whispering gallery mode*, **Proc. Natl Acad. Sci. U.S.A.** **105** (52), pp. 20701–20704, 2008. doi:10.1073/pnas.0808988106
3. S. Arnold, D. Keng, S. I. Shapova, S. Holler, W. Zurawsky, and F. Vollmer, *Whispering gallery mode carousel—a photonic mechanism for enhanced nanoparticle detection in biosensing*, **Opt. Express** **17** (8), pp. 6230–6238, 2009.