# Nanoparticle-enabled Biosensor Technologies for Rapid Diagnostics

Lateral flow assays are low-cost, easy-to-use, portable devices used for the rapid detection of a wide variety of analytes with applications in healthcare, agriculture, food, and environmental sciences.

These self-contained assays meet the essential requirements for use as point-of-care (POC) rapid diagnostics, offering high sensitivity, and selectivity for detection or quantification of biomolecules in complex samples including blood, urine, saliva, and other fluids. The popularity and success of lateral flow assays can be attributed to their general design which has remained almost unchanged since their first use decades ago as pregnancy tests. These assays became household fixtures as rapid diagnostic tests for SARS-CoV-2 infection.

Assays based on a lateral flow format rely on nanoscale reporter particles that generate a signal during use. The reporter particles are labeled with a molecule – often an antibody or nucleic acid – that will recognize an analyte in the sample and bind to a specific location on the test strip. Optical biosensors are the most common type of biosensor used in lateral flow assays and provide a visual readout of test results by measuring the interaction of an optical field with a biorecognition sensing element. Often these reporter particles are metallic nanoparticles, which due to their unique plasmonic properties, can be used in other emerging biosensor technologies, from photonic biosensors to biosensors utilizing Surface Enhanced Raman Spectroscopy (SERS) or Surface Enhanced Fluorescence (SEF).

This whitepaper describes three of these technologies – photonic biosensors, surface-enhanced Raman spectroscopy (SERS), and surface-enhanced fluorescence (SEF) spectroscopy.[1,2]

## Photonic Biosensors

Photonic biosensors use interactions between light, typically a spectroscopic laser, and a target analyte as a detection mechanism. The plasmonic properties of nanoparticles make them ideal for these types of diagnostics platforms, as they can enhance these interactions between light and analytes to improve the sensitivity of the measurement. When metallic nanoparticles are excited by a specific wavelength of light, the conduction electronics begin to coherently oscillate in an effect known as Surface Plasmon Resonance (SPR) (**Figure 1**). This process can be fine-tuned to match a particular target wavelength or optical effect with the use of nanoparticles of different shapes, size, or materials.

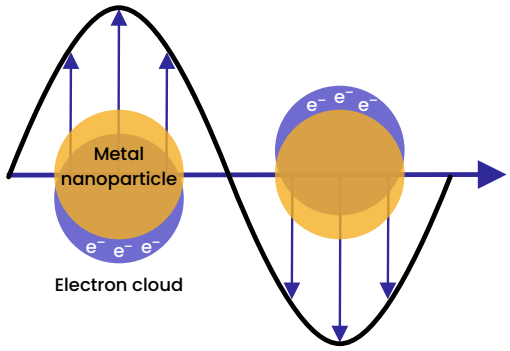


**Figure 1:** Conduction electrons on the nanoparticle surface undergo a collective oscillation when excited by light at specific wavelengths.

nanoComposix

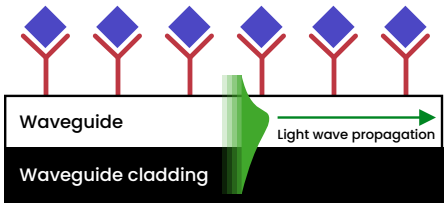


**Figure 2:** The evanescent field generated by light propagating through a waveguide.

Commonly used photonic biosensors are based on the evanescent-field principle (**Figure 2**), where energy from light traveling in a waveguide or thin-film extends beyond the surface of that waveguide or film in the form of an evanescent field. Analytes bound near the surface of the waveguide or film can then interact with this evanescent wave and create changes in the spectroscopic signal.[1] One variety of these photonic biosensors, known as nanoplasmonic biosensors, employ nanometer-sized metallic films or nanostructures (mainly gold), which absorb light and excite coherent electron resonances (surface plasmon polaritons) at the surface. This resonance can either propagate along with the thin film (propagating SPR) or be confined to the vicinity of the nanoparticle (localized SPR, or LSPR). In both cases, it generates a strong near-field enhancement that penetrates a few nanometers (10–500 nm) into the surroundings.

While nanoplasmonic biosensors have a demonstrated functionality, they can have a limited ability to detect very low amounts of analyte and their designs can be complicated to integrate and miniaturize for rapid diagnostic systems. Silicon waveguide and resonator photonic biosensor designs offer a powerful alternative. They can be incorporated as detectors into microfluidic lab-on-a-chip diagnostics, and in some cases, their sensitivity can be over an order of magnitude better than nanoplasmonic equivalents.

One compelling silicon-nanophotonic technology is the photonic ring resonator (**Figure 3**). In this technology, light is coupled from a straight waveguide into a ring-shaped waveguide with a diameter tuned such that the peaks and troughs of a specific wavelength of light will be an equal number inside the ring and resonate on themselves. The resonant light circulates along the ring resonator and has an evanescent field that reaches into the surrounding medium (e.g., liquid, gas, and polymer coatings). Capture antibodies bind analyte near the surface of the waveguide and change the resonance through interactions with the evanescent field. This leads to a detectable shift in the resonant wavelength of the ring that is proportional to the amount of analyte.

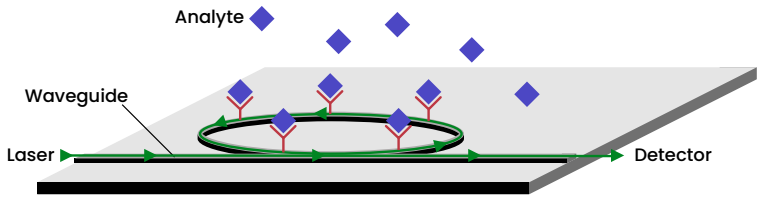


**Figure 3:** Diagram of a photonic ring resonator.

While the presence of analyte alone can create a detectable shift, the inclusion of a gold or silver nanoparticle, which has a powerful surface plasmon resonance, can increase interactions with the evanescent field through changes in the local dielectric environment. In turn, each binding event from an analyte yields a greater shift in the resonant wavelength of the ring, providing signal enhancement. **Figure 4** demonstrates how a biofunctionalized nanoparticle can be bound to the analyte in a sandwich pair, similar to antibody pair schemes used in some lateral flow assays.

Photonic ring resonators are also well-suited for capturing multiple analytes in a single test which is economically advantageous and more convenient for users. In this case, multiple rings at different wavelengths can be designed to capture different analytes.

nanoComposix

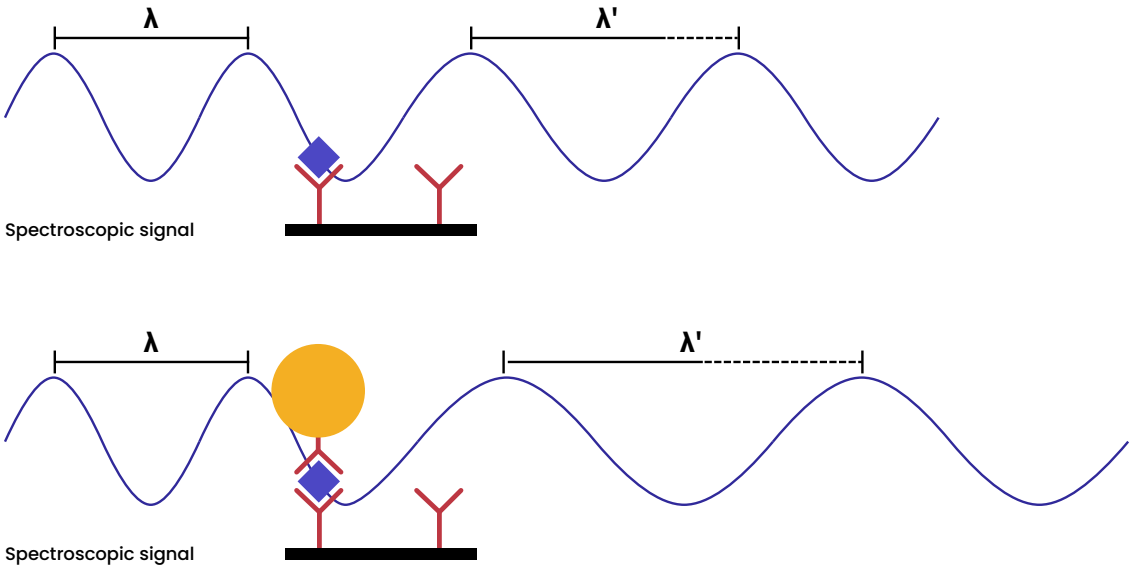


**Figure 4:** The addition of nanoparticles to the analyte binding scheme creates a greater shift in the resonant wavelength of a ring resonator.

## Surface-Enhanced Raman Spectroscopy Biosensors

Raman spectroscopy can be used to identify molecules by their unique vibrational modes. While intrinsic Raman scattering of photons from molecules is weak and requires long measurement times to obtain a Raman spectrum, surface-enhanced Raman scattering (SERS) from molecules near the surface of plasmonic metal nanoparticles offers the potential for intensities comparable to that of fluorescent tags.[3] The SERS effect can enhance the Raman scattering of bound molecules by as much as 14 orders of magnitude, allowing for the detection of even single molecules. The enhancement is driven by the high electric field intensities created at locations on the nanoparticle surface and is therefore highly dependent on the nanoparticle geometry, surface features, and the specific position of the molecule.

Researchers at University of Korea, Seoul have used gold nanoparticles to collect SERS signals from exosomes derived from both healthy and lung cancer cell lines. These signals were then used to train deep learning models to classify circulating exosomes accordingly with high accuracy. They noted that results from these experiments demonstrated that nanoplasmonic sensing techniques such as this might be used to detect early-stage cancer without specific biomarkers.[4] Advances like this show how SERS might be used to enable more rapid or versatile diagnostic platforms.

## Surface-Enhanced Fluorescence Biosensors

While fluorescent molecules are among the most popular biosensing reagents, they have significant drawbacks, including low optical cross-sections which make individual fluorophores difficult to detect, and poor photostability which can degrade emission, complicating detection and quantification. Surface-enhanced fluorescence (SEF) is a phenomenon first observed in the 1970s that occurs when a fluorophore is placed near the high electromagnetic fields at the surface of a plasmonic metal nanoparticle, enhancing the fluorophore emission intensity by orders of magnitude. The enhancement can be attributed to two effects:

- Focusing on the incoming light due to the large absorption and scattering cross-sections of the plasmonic particle
- A decrease in the fluorescence lifetime of the fluorophore that allows the excited state to return to the ground state at a higher frequency

By leveraging the plasmonic properties of a nanoparticle, an SEF assay can deliver greater sensitivity.

nanoComposix

Researchers at Duke University have used silver nanocubes to enhance fluorescence by integrating a sandwich immunoassay microarray within a plasmonic nanogap cavity (**Figure 5**).[5] This is an important improvement that helps increase performance, simplify detection, and reduce cost for POC diagnostic tests based on fluorescent protein microarray technologies. The immunoassay consists of inkjet-printed antibodies on a polymer brush which is grown on a gold film. Colloidally synthesized silver nanocubes are placed on top and interact with the underlying gold film creating high local electromagnetic field enhancements. By varying the thickness of the brush from 5 to 20 nm, up to a 151-fold increase in fluorescence and 14-fold improvement in the limit-of-detection is observed for the cardiac biomarker B-type natriuretic peptide (BNP) compared to the unenhanced assay, paving the way for a new generation of POC clinical diagnostics.

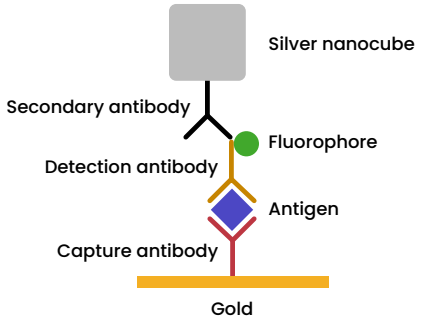


**Figure 5:** Integration of a sandwich immunoassay microarray within a plasmonic nanogap cavity.

## Conclusion

As described in this whitepaper, emerging biosensor platforms like photonic, SEF, and SERS can be considered to address challenges in rapid diagnostics. Integrating nanoparticles into the assay design can enhance sensitivity, which is critical to enabling earlier detection of disease, developing assays to detect new biomarkers, or even modifying an existing platform to a different type of sample matrix, such as saliva, which is easier to collect at POC.

At nanoComposix, LLC (a Fortis Life Sciences company) our core competency is developing and manufacturing metallic and metal oxide nanoparticles for rapid diagnostics at scales that support our partners from early development through commercial manufacturing. We have experience creating a wide variety of plasmonic nanoparticles of different metals, shapes, and composite structures with properties tuned for these novel detection platforms. Review our large library of off-the-shelf nanoparticles for research use or contact us today to speak with an expert regarding bulk scale requirements and custom developments.

**For more information, visit fortislife.com**

nanoComposix

## References

1. Maria Soler, Olalla Calvo-Lozano, M.-Carmen Estevez and Laura M. Lechuga. Nanophotonic Biosensors: Driving Personalized Medicine. Optica.2020:Volume 31. https://doi.org/10.1364/OPN.31.4.000024

2. Zeng, Shuwen & Baillargeat, Dominique & Ho, Ho-Pui & Yong, Ken-Tye. Nanomaterials enhanced surface plasmon resonance for biological and chemical sensing applications. Chem. Soc. Rev., 2014,43, 3426-3452. https://doi.org/10.1039/C3CS60479A

3. X.-M. Qian and S. M. Nie. Single-molecule and single-nanoparticle SERS: from fundamental mechanisms to biomedical applications Chem. Soc. Rev., 2008,37, 912-920.

4. Hyunku Shin, Seunghyun Oh, Soonwoo Hong, Minsung Kang et al. Early-Stage Lung Cancer Diagnosis by Deep Learning-Based Spectroscopic Analysis of Circulating Exosomes. ACS Nano 2020, 14, 5, 5435–544. https://doi.org/10.1021/acsnano.9b09119

5. Daniela F. Cruz , Cassio M. Fontes, Daria Semeniak et al. Ultrabright Fluorescence Readout of an Inkjet-Printed Immunoassay Using Plasmonic Nanogap Cavities. Nano Lett. 2020, 20, 6, 4330–4336. https://doi.org/10.1021/acs.nanolett.0c01051

nanoComposix