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Review

Advances in Immunoassay Technologies: Comprehensive Insights into Principles, Applications, and Emerging Trends

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Abstract

Immunoassays are sensitive and specific analytical techniques that rely on antigen–antibody interactions to detect and quantify biomolecules in complex biological samples. It outlines the historical development of immunoassays, beginning with the pioneering radio immunoassay developed by Rosalyn S. Yalow and Solomon Person, based on the antigen–antibody specificity concept proposed by Paul Ehrlich, and later advancements such as ELISA introduced by Eva England and Peter Perlman, along with monoclonal antibody technology developed by Georges Kohler and César Mil stein. It summarizes major immunoassay formats, including competitive and sandwich assays, and key techniques such as ELISA, Possible spelling mistake found. immunoassay (CLIA), fluorescent immunoassay (FIA), radio immunoassay (RIA), Western blotting, flow cytometry, lateral flow immunoassay (LFIA), and surface-enhanced Raman spectroscopy (SERS).It further highlights recent innovations such as multiplex platforms, nanotechnology-enhanced detection, microfluidic integration, digital immunoassays, and artificial intelligence–assisted analysis, emphasizing the expanding role of immunoassays in clinical diagnostics, biomedical research, pharmaceutical analysis, and point-of-care testing.

Keywords: ELISA, Radioimmunoassay, Western blotting, Chemiluminescent immunoassay, Fluorescent immunoassay, Flow cytometry, Lateral flow immunoassay, Surface Enhanced Ramen Spectroscopy immunoassay.

1. INTRODUCTION

Accurate detection and quantification of biomolecules are essential in clinical diagnostics, pharmaceutical development, and biomedical research. Immunoassays are analytical methods that utilize the high specificity and affinity of antigen–antibody interactions to measure analytes in complex biological matrices such as blood, urine, and plasma.

Immunoassays offer several advantages including high sensitivity, minimal sample preparation, cost-effectiveness, and adaptability to automation. Depending on assay design, detection signals may be directly or

inversely proportional to analyte concentration. These characteristics have established immunoassays as indispensable tools in modern laboratory medicine. [1]

2. HISTORY OF IMMUNOASSAYS

Rosalyn S. Yalow and Solomon Berson lay the groundwork for immunoassay technology in 1959, when they created the first radioimmunoassay (RIA) for insulin measurement. This innovation proved that ultra-low hormone concentrations may be detected with remarkable specificity. Yalow was later awarded the Nobel Prize in Physiology or Medicine in 1977. [2]

The conceptual basis of antigen–antibody specificity traces back to the "lock-and-key" theory proposed by Paul Ehrlich in the early 20th century.

In 1971, Peter Perlmann and Eva Engvall introduced the enzyme-linked immunosorbent assay (ELISA), eliminating the need for radioactive labels. The development of monoclonal antibody technology by Georges Köhler and César Milstein in 1975 further enhanced assay specificity and reproducibility.

Subsequent decades saw the emergence of chemiluminescent, fluorescent, electrochemical, and nanoparticle-based immunoassays, greatly improving analytical sensitivity and automation. [3]

3. PRINCIPLE OF IMMUNOASSAYS

Immunoassays are based on the following key principles:

- **Antigen-Antibody Interaction**: Antigens attach to complementary antibodies. This extremely specific interaction serves as the foundation for analyte detection.
- **Detectable Labels:** Labels coupled to antigens or antibodies provide measurable signals. Radioisotopes, enzymes, fluorophores, chemiluminescent chemicals, and electrochemiluminescent tags are examples of commonly used labels.
- **Assay Formats:** Competitive Immunoassay - Analytes with and without labels fight for limited antibody binding sites. Signal intensity is inversely related to analyte concentration. Non-competitive (Sandwich) Immunoassay – The analyte is trapped between two antibodies. The intensity of the signal is directly proportional to the concentration of analytes.
- Detection and Quantification: Analyte concentration is determined by comparing signal intensity to a calibration curve.[4]

3.1. Elisa (Enzyme-Linked Immunosorbent Assay)

Enzyme-Linked Immunosorbent Assay (ELISA) is a highly sensitive and specific immunoassay technique used for the qualitative and quantitative detection of antigens or antibodies in biological samples. Since its development in 1971, ELISA has become a cornerstone of clinical diagnostics, research laboratories, and pharmaceutical analysis. The assay relies on antigen–antibody interactions and enzyme-mediated signal amplification to generate measurable signals. [5]

- **DIRECT ELISA:** Direct ELISA is a solid-phase immunoassay used to detect specific antigens by directly coating them onto a microtiter plate. An enzyme-labeled primary antibody binds to the antigen, and upon addition of a substrate, a measurable color change occurs. The intensity of the color is proportional to the antigen concentration.
- **INDIRECT ELISA:** Indirect ELISA is used to detect specific antibodies in a sample. The antigen is first coated onto a microtiter plate, followed by addition of the primary antibody (from the sample) and an enzyme-labeled secondary antibody that binds to the primary antibody. After adding a substrate, a color change develops, and the intensity is proportional to the amount of antibody present.
- **SANDWICH ELISA:** Sandwich ELISA is used to detect and quantify specific antigens in a sample. A capture antibody is first coated onto the microtiter plate to bind the target antigen, followed by the addition of a second enzyme-labeled detection antibody forming an antibody–antigen–antibody "sandwich." Upon substrate addition, a color change occurs, and the intensity is proportional to the antigen concentration.
- **COMPETITIVE ELISA:** Small antigens or antibodies based on competitive binding can be found using competitive ELISA. Using this technique, a tagged antigen and a sample antigen compete to bind to a particular antibody. The concentration of the target analyte in the sample is inversely correlated with the color intensity that develops following substrate addition.[6]

3.2. Recent Advancements in Elisa

❖ **MULTIPLEX ELISA:** Multiplex enzyme-linked immunosorbent assay is an advanced immunoassay platform that enables the simultaneous detection and quantification of multiple analytes within a single biological sample. Unlike conventional ELISA, which measures one analyte per well, multiplex ELISA utilizes spatial separation or bead-based encoding strategies to detect multiple biomarkers in parallel. Multiplex ELISA has become indispensable in cytokine profiling, oncology biomarker panels, autoimmune disease monitoring, vaccine studies, and systems biology research.

In the **array-based multiplex ELISA**, different capture antibodies are spotted at defined locations within the same well. The sample is added, and multiple analytes bind to their respective capture antibodies. Detection antibodies, usually biotinylated and followed by enzyme conjugates, are added to form specific antibody–antigen complexes. Upon addition of substrate, signals are

generated and measured using specialized imaging systems.

In the **bead-based multiplex ELISA**, microspheres coated with distinct capture antibodies are mixed with the sample. Each bead population is internally labeled with a unique fluorescent signature, allowing identification of specific analytes. After binding and detection antibody addition, fluorescence intensity is measured using flow cytometry–based or dedicated multiplex analyzers.[7]

3.3. Radioimmunoassay

Radioimmunoassay (RIA) is a highly sensitive immunoassay technique used to measure minute concentrations of antigens, such as hormones or drugs, in biological samples. It is based on competitive binding between a radiolabeled antigen and the sample antigen for a specific antibody. The bound and free antigens are separated, and radioactivity is measured using a gamma counter. The detected radioactivity is inversely proportional to the concentration of the analyte in the sample. [8]

3.4. Limitations of radioimmunoassay

Radioimmunoassay involves the use of radioactive isotopes, which pose potential health and environmental hazards and require stringent regulatory compliance, specialized laboratory infrastructure, and trained personnel. The short half-life of radioisotopes limits reagent stability and increases operational costs related to storage and waste disposal. Owing to these safety concerns and the widespread availability of safer, non-radioactive alternatives such as ELISA and chemiluminescent immunoassays, RIA is now largely replaced and is rarely employed in routine human diagnostic practice. [8]

3.5. Western Blotting

Western blotting is an analytical technique used to detect and quantify specific proteins in a complex biological sample. Proteins are first separated based on molecular weight using SDS-PAGE and then transferred onto a membrane (nitrocellulose or PVDF). The membrane is incubated with a specific primary antibody followed by a labeled secondary antibody for detection. The target protein is visualized as a band using chemiluminescent, colorimetric, or fluorescent methods. [9]

3.6. Recent Advancements in Western Blotting

- **Automated capillary immunoassay:** This system automates protein separation and immunodetection, significantly improving reproducibility, sensitivity, and quantitation with minimal sample input. Fluorescent multiplex detection, which allows simultaneous detection of multiple proteins on the same blot using distinct fluorescent labels, enhancing throughput and comparative analysis in a single assay.[10]
- **Microfluidic Based Western Blotting:** Integration of microfluidic platforms has miniaturized Western blot workflows, reducing sample volume and assay time while increasing throughput. These systems enable rapid, parallel protein analysis with improved sensitivity and potential for single-cell applications.[10]

4. CHEMILUMINESCENT IMMUNOASSAY

Chemiluminescent immunoassay (CLIA) refers to immunoassay techniques in which the detection signal is generated by a chemiluminescent reaction - a chemical reaction that produces light without requiring external light source excitation. The intensity of emitted light correlates directly with the quantity of target analyte (e.g., antigen or antibody) in a sample, enabling highly sensitive quantification. CLIA has emerged as a leading non-isotopic immunoassay method in clinical diagnostics due to its rapid turnaround, wide dynamic range, and automated compatibility. CLIA can be configured in several assay designs, similar to other immunoassays:

- **Direct CLIA:** Labeled antibody directly binds the analyte. Simple but less sensitive.
- **Sandwich CLIA:** Capture antibody immobilizes the analyte, then a second labeled antibody binds, boosting signal and specificity, ideal for larger proteins or biomarkers.
- **Competitive CLIA:** High analyte concentrations compete with labeled analyte analogs for antibody binding, useful for small molecules.[11]

4.1. Recent Advancements of Clia

- **Automated Magnetic Digital Microfluidic CLIA Systems:** Compact CLIA platforms that combine magnetic digital microfluidics with automated workflows significantly improve detection speed and sensitivity for protein biomarkers. These systems manipulate magnetic beads in microdroplets to automate separation, washing, and signal generation, enabling rapid assays (e.g., cardiac troponin I) with low limits of detection and high linear range, enhancing clinical diagnostic applicability.[12]
- **Deep Learning-Enhanced Chemiluminescent Vertical Flow Assays for Point-of-Care Testing:** Innovations integrating chemiluminescent vertical flow assays with deep learning algorithms enable highly sensitive, rapid quantification of biomarkers such as cardiac troponin I using portable readers. These platforms achieve laboratory-grade performance with broad dynamic range, low sample volume, and rapid turnaround, making them promising for decentralized and emergency diagnostics. [12]

4.2. Fluorescent Immunoassay

Fluorescent immunoassays (FIAs) are analytical techniques in which antigen–antibody binding events are quantified through fluorescence emission from labeled antibodies or detection probes. Unlike colorimetric systems, FIAs use fluorescent dyes or nanoparticles that emit light upon excitation, providing high sensitivity, wide dynamic range, and multiplexing capability, making them valuable in clinical diagnostics, environmental monitoring, and research applications.

- **Direct FIA:** A fluorescently labeled primary antibody binds directly to the target antigen. This format is simple and rapid but offers relatively lower signal amplification.
- **Indirect FIA:** An unlabeled primary antibody binds the antigen, followed by a fluorescently labeled secondary antibody. This approach enhances sensitivity through signal amplification.
- **Sandwich FIA:** A capture antibody immobilizes the antigen, and a fluorescently labeled detection antibody binds to another epitope, forming a "sandwich." This format provides high specificity and is widely used for protein biomarker detection.
- **Competitive FIA:** Labeled and unlabeled antigens compete for antibody binding sites. The fluorescence signal is inversely proportional to analyte concentration and is suitable for small molecules.
- **Time-Resolved FIA (TRFIA):** A sensitive immunoassay technique that uses lanthanide-labeled antibodies (e.g., europium) with long fluorescence lifetimes. Fluorescence is measured after a time delay following excitation, which eliminates short-lived background signals and improves sensitivity and specificity.[13]

4.3. Recent Advancements of Fluorescent Immunoassay

- **Homogeneous Fluorescent Immunosensors for Rapid, Wash-Free Detection:** Recent research has emphasized homogeneous fluorescent immunoassay formats that eliminate traditional wash and separation steps, significantly simplifying workflows and reducing assay time. By using engineered fluorescent probes and antibody constructs that produce a detectable signal change upon target binding, these assays enable rapid, sensitive detection in complex biological samples without extensive sample processing — a key step toward point-of-care and high-throughput applications.[14]
- **Nanomaterial-Enhanced Fluorescence Labels for Ultra-Sensitive Multiplexing:** Advances in fluorescent nanomaterials, including quantum dots (QDs) and up-conversion nanoparticles (UCNPs), have substantially improved FIA performance. These labels offer higher brightness, reduced photobleaching, and narrow emission spectra, enabling highly sensitive detection and multiplexed assays with minimal spectral overlap. Nanomaterial-based FIAs demonstrate significantly lower detection limits and enhanced multiplex capability for simultaneous measurement of several analytes in a single assay.[14]

4.4. Flow Cytometry

Flow Cytometry Immunoassay is a powerful analytical technique used to detect and quantify specific cells, proteins, or biomarkers in a heterogeneous cell population. It employs fluorescent-labeled antibodies that bind to target antigens on or within cells. As cells pass individually through a laser beam, scattered light and emitted fluorescence are measured, allowing multiparametric analysis. This technique is widely used in immunology, oncology, hematology, and clinical diagnostics for cell characterization and biomarker detection. [15]

4.5. Recent Advancements of Flow Cytometry

- **High-Dimensional Spectral Flow Cytometry for Deep Immune Profiling:** Recent technological developments in spectral flow cytometry extend conventional multi-color analysis by capturing full emission spectra from fluorophores rather than discrete channels. Advanced unmixing algorithms now allow simultaneous measurement of 40+ parameters per cell with reduced spectral overlap, enabling deep immune profiling and high-resolution characterization of cellular phenotypes in complex tissues such as tumor microenvironments and peripheral blood. This advancement significantly expands the analytical capacity of flow cytometry in systems immunology and clinical research.[16]
- **Integration of Machine Learning for Automated Population Identification:** The application of machine learning and artificial intelligence to flow cytometry data analysis has enhanced accuracy, reproducibility, and throughput. Algorithms such as t-SNE, UMAP, and FlowSOM are now routinely used to identify rare cell populations, cluster multidimensional data, and visualize high-parameter datasets. These computational tools reduce user bias in gating strategies, uncover novel cellular subsets, and improve interpretation of complex phenotypic landscapes in both research and diagnostic settings.[16]

5. LATERAL FLOW IMMUNOASSAY

Lateral Flow Immunoassay (LFIA) is a rapid immunodiagnostic technique used for the qualitative or semi-quantitative detection of antigens or antibodies in biological samples. It works on the principle of capillary action, where the sample moves along a membrane containing labeled antibodies. If the target analyte is present, a visible colored line appears in the test region.

LFIA is simple, cost-effective, rapid, and widely used for point-of-care testing. [17]

5.1. Recent Advancements of Lateral Flow Immunoassay

- **Fluorescent and Nanoparticle-Enhanced LFIA:** Integration of advanced labels such as quantum dots, up-conversion nanoparticles, and fluorescent nanobeads has significantly improved LFIA sensitivity and enabled quantitative and multiplex detection, overcoming limitations of traditional colloidal gold visual readouts.[18]
- **Digital and Smartphone-Assisted LFIA Platforms:** Development of portable digital readers and smartphone-based imaging systems with AI-assisted signal analysis allows objective quantification, reduced user bias, enhanced limit of detection, and real-time data transmission, expanding LFIA utility in point-of-care diagnostics.[18]

5.2. Surface Enhanced Ramen Spectroscopy (Sers) Immunoassay

Surface-Enhanced Raman Spectroscopy (SERS) immunoassay is a highly sensitive analytical technique that combines antigen–antibody specificity with Raman signal enhancement for biomarker detection. In this method, antibodies are conjugated to metallic nanostructures (such as gold or silver nanoparticles), which amplify the Raman scattering signal of reporter molecules. When the target analyte binds to the antibody, a characteristic Raman spectral signal is generated and measured using a Raman spectrometer. SERS immunoassays offer ultra-high sensitivity, molecular specificity, and potential for multiplex detection in clinical diagnostics and biomedical research. [19]

5.3. Recent Advancements of Sers Immunoassay

- **Core–Shell Nanotags with Ultra-High Enhancement for Attomolar Sensitivity:** Recent development of engineered core–shell plasmonic nanoparticles (e.g., Au@Ag and alloy nanostructures) with optimized "hot spots" has significantly increased SERS enhancement factors. These nanotags enable attomolar level detection of protein biomarkers in complex biological samples, enhancing assay sensitivity and lowering limits of detection compared to conventional SERS probes.[20]
- **Microfluidic-Integrated and Multiplex SERS Immunoassays:** Integration of SERS immunoassays with microfluidic lab-on-chip platforms has improved fluid control, reaction kinetics, and reproducibility. Combined with spectrally distinct Raman reporter labels, these systems achieve simultaneous, high-throughput detection of multiple analytes on a single chip, supporting advanced diagnostic panels in clinical and point-of-care settings.[20]

5.4. Future Perspectives of Immunoassay

- **Next-Generation Sensitivity via Nanotechnology and Signal Amplification:** Future immunoassays will increasingly harness engineered nanomaterials — such as quantum dots, plasmonic nanoparticles, and upconversion nanoparticles — to amplify detection signals far beyond current limits. Structural and surface modifications of nanoparticles can create "hot spots" for enhanced optical or Raman signals, enabling ultra-low limit of detection (LOD) in the femtomolar to attomolar range. Additionally, digital immunoassay formats that count single binding events — such as digital ELISA — will further enhance sensitivity and enable early disease detection.[21]
- **Integrated and Portable Point-of-Care Platforms:** Traditional immunoassays often require centralized laboratory infrastructure. Future prospects include fully integrated point-of-care (POC) devices that combine immunochemical reactions with on-board fluidics, detection, and data processing. Portable formats such as smartphone-based readers, microfluidic cartridges, and lab-on-chip immunoassays will support rapid, decentralized diagnostics, particularly in resource-limited settings or emergency response scenarios. The integration of low-cost optics and miniaturized biosensors will make high-performance immunoassays accessible outside conventional laboratories.[22]
- **Multiplexed and High-Throughput Assays:** Advances in fluorescent labels, spectral detection, and spatial encoding will enable immunoassays to simultaneously quantify dozens to hundreds of analytes in a single sample. High-plex immunoassays will be critical in fields such as systems biology, biomarker discovery, and personalized medicine, where comprehensive molecular profiling is essential. Techniques such as spectral cytometry, DNA barcoding, and mass cytometry will further expand multiplexing capacities.[23]
- **Digital and AI-Enhanced Data Interpretation:** As immunoassay data complexity increases — particularly with high-parameter assays — computational methods will play a central role in interpretation. Machine learning and artificial intelligence (AI) will automate pattern recognition, reduce operator bias, and detect subtle trends in large datasets. Predictive modeling can enhance assay interpretation in complex matrices and support clinical decision-making by correlating biomarker patterns with disease states and outcomes.[24]
- **Convergence with Other Diagnostic Modalities:** Immunoassays will increasingly integrate with other analytical platforms such as nucleic acid amplification tests, biosensors (electrochemical, optical), and sequencing technologies to provide multi-modal diagnostic insights. Hybrid assays that

combine protein detection with genomic or transcriptomic information will support comprehensive phenotyping and precision medicine.[25]

6. REFERENCES

1. Yalow RS, Berson SA. Immunoassay of endogenous plasma insulin in man. J Clin Invest. 1960;39(7):1157–1175. doi: https://doi. org/10.1172/JCI104130
2. Ehrlich P. On immunity with special reference to cell life. Proc R Soc Lond. 1900; 66:424–448. doi: https://doi.org/10.1098/rspl.1899.0146
3. Engvall E, Perlmann P. Enzyme-linked immunosorbent assay (ELISA): quantitative assay of immunoglobulin G. Immunochemistry. 1971; 8(9):871–874.doi: https://doi.org/10. 1016/0019-2791(71) 90454-X
4. Köhler G, Milstein C. Continuous cultures of fused cells secreting antibody of predefined specificity. Nature. 1975;256:495–497.doi: https://doi.org/10.1038/256495a0
5. Crowther JR. The ELISA Guidebook. Totowa (NJ): Humana Press; 2000.doi: https://doi.org/10.1385/ 1592590497
6. Wild D, editor. The Immunoassay Handbook: Theory and Applications of Ligand Binding, ELISA and Related Techniques. 4th ed. Oxford: Elsevier; 2013.doi: https://doi.org/10.1016/C2010-0-66258-4
7. Rissin DM, Kan CW, et al. Single-molecule enzyme-linked immunosorbent assay detects serum proteins at subfemtomolar concentrations. Nat Biotechnol. 2010;28:595–599.doi: https://doi.org/10.1038/nbt.1641
8. Hales CN, Randle PJ. Immunoassay of insulin with insulin-antibody precipitate. Biochem J. 1963;88:137–146.doi: https://doi.org/10.1042/bj0880137
9. Towbin H, Staehelin T, Gordon J. Electrophoretic transfer of proteins from polyacrylamide gels to nitrocellulose sheets. Proc Natl Acad Sci U S A. 1979; 76:4350–4354.doi: https://doi.org/10.1073/pnas.76.9.43 50
10. Burnette WN. "Western blotting": electrophoretic transfer of proteins from sodium dodecyl sulfate–polyacrylamide gels to unmodified nitrocellulose and radiographic detection with antibody and radioiodinated protein A. Anal Biochem. 1981;112:195–203.doi: https://doi.org/10.1016/0003-2697(81) 90281-5
11. Weeks I, Beheshti I, McCapra F, et al., Acridinium ester labels for immunoassay. Clin Chem. 1983; 29(8):1474–1479.doi: https://doi.org/10.1093/clinchem/29. 8.1474
12. Kricka LJ. Clinical applications of chemiluminescence. Clin Chem. 1991; 37(9):1472–1481.doi: https://doi.org/10.1093/ clinchem/37.9.1472
13. Soini E, Kojola H. Time-resolved fluorometer for lanthanide chelates—A new generation of nonisotopic immunoassays. Clin Chem. 1983;29(1):65–68.doi: https://doi.org/10.1093/ clinchem/29.1.65
14. Medintz IL, Uyeda HT, Goldman ER, Mattoussi H. Quantum dot bioconjugates for imaging, labeling and sensing. Nat Mater. 2005; 4:435–446.doi: https://doi.org/10.1038/ nmat1390
15. Herzenberg LA, Tung J, Moore WA, Herzenberg LA, Parks DR. Interpreting flow cytometry data: a guide for the perplexed. Nat Immunol. 2006;7:681–685. doi: https://doi.org/ .1038/ni0706-681
16. Maecker HT, Trotter J. Flow cytometry controls, instrument setup, and the determination of positivity. Cytometry A. 2006; 69A:1037–1042.doi: https://doi.org/10.1002/ cyto.a.20333
17. Posthuma-Trumpie GA, Korf J, van Amerongen A. Lateral flow (immuno)assay: its strengths, weaknesses, opportunities and threats. Anal Bioanal Chem. 2009; 393:569–582.doi: https://doi.org/10. 1007/s00216-008-2287-2
18. Parolo C, Merkoci A. Paper-based nanobiosensors for diagnostics. Chem Soc Rev. 2013;42:450–457.doi: https://doi.org/10.1039/ C2CS35255A
19. Kneipp K, Wang Y, Kneipp H, Perelman LT, Itzkan I, Dasari RR, et al. Single molecule detection using surface-enhanced Raman scattering. Phys Rev Lett. 1997; 78:1667–1670.doi: https://doi.org/10.1103/PhysRevLett. 78.1667
20. Pilot R, Signorini R, Durante C, Orian L, Bhamidipati M, Fabris L. A review on surface-enhanced Raman scattering. Biosens Bioelectron. 2019; 143:111530.doi: https://doi. org/10.1016/j.bios.2019.111530
21. Zhou L, Liu Y, Lu Y, Zhou P, Lu L, Lv H, et al. Recent advances in immunoassays based on nanozymes. Biosensors. 2022;12(12):1119.doi: https://doi.org/10.3390/bios12121119
22. Yetisen AK, Akram MS, Lowe CR. Paper-based microfluidic point-of-care diagnostic devices. Lab Chip. 2013;13:2210-2251.doi: https://doi.org/10.1039/C3LC50169H
23. Kingsmore SF. Multiplexed protein measurement: technologies and applications of protein and antibody arrays. Nat Rev Drug Discov. 2006; 5:310-320.doi: https://doi.org/10. 1038/nrd2006
24. Topol EJ. High-performance medicine: the convergence of human and artificial intelligence. Nat Med. 2019;25:44-56.doi: https://doi.org/10.1038/s41591-018-0300-7
25. Mishra A, Singh PK, Chauhan N, Roy S, Tiwari A, Gupta S, et al. Emergence of integrated biosensing-enabled digital healthcare devices. Sensors & Diagnostics. 2024; 3:718-744.doi: https://doi.org/10. 1039/D4SD00017