Biomarkers in Translational Viral Diagnostics: Biological Foundations, Integrating Pathogen Detection, Host Response, and Multi-Omics for Clinical Decision-Making

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Abstract. Viral diseases remain a major global public health problem and the need for rapid, sensitive and accurate diagnostic testing. Translational viral diagnostics is based on biomarkers (molecular markers of viral infection, host response, severity and treatment outcome). This review combines the principal types of biomarkers, including pathogen biomarkers (viral nucleic acids, proteins, mutations), host response biomarkers (cytokines and chemokines, immune cell populations, serological markers), and multi-omics biomarkers from transcriptomics, proteomics, metabolomics, epigenetics, and microbiome analysis. The technologies for their detection will be discussed, including PCR, next-generation sequencing, CRISPR-based technologies, immunoassays, biosensors, and Artificial Intelligence (AI), as well as the clinical applications of biomarkers in early detection, severity assessment, therapeutic response, and vaccine response. Progress is still minimal, with biological variability, assay harmonisation, and cost and global access to diagnostics still to be addressed. Researchers and developers will direct the next generation of technologies toward multi-omics integration, point-of-care devices, next-generation CRISPR, amplification-free detection, and AI-driven predictive diagnostics and disease surveillance. The entire shift in biomarker-based diagnostics in clinical management, public health strategy, and preparedness for the emergence of viral diseases is a paradigm shift.

Keywords: Biomarkers; Viral Diagnostics; Translational Medicine; Host Immune Response; Pathogen-Derived Biomarkers; Multi-Omics; Transcriptomics; Proteomics.

INTRODUCTION

While many viral diseases have been a health problem locally, seasonally, and globally, some are more serious than others, and, in our current COVID-19 pandemic, viral diseases will not cease to be a significant health issue. Traditional laboratory methods such as serology, viral culture, and nucleic acid amplification testing (NAATs) have been used as the basis for clinical virology but have limitations, including limited ability to detect early infection, low sensitivity, technical requirements, and the need to account for genetic mutations [1, 2]. As a result, biomolecules (molecular markers of infection, immune status, disease severity and treatment response) have emerged as an important tool for developing next-generation diagnostics for viral infections. Researchers closely couple these approaches with basic and clinical virology [3]. There are many different types of biomarkers. Pathogen biomarkers (viral nucleic acids, proteins, and mutations) play a fundamental role in lab diagnosis and variant surveillance, especially during COVID-19, for confirming cases, assessing viral load, and monitoring new variants such as Delta and Omicron [4].

However, pathogen detection cannot convey the complexities of the immune response and host response biomarkers are also important, such as cytokines (IL-6, TNF-α, IP-10), lymphopenia, and serological markers (IgM, IgG, neutralising anti-

bodies), which can help to assess risk and guide clinical care, for instance in COVID-19 [5].

Researchers extend this approach with multi-omics biomarkers, which they use to identify host–viral interactions through transcriptomic profiles that distinguish viral from bacterial infection, proteomic and metabolomic changes that reveal pathway alterations, and epigenetic changes that reflect latent and long-term effects of viral infection, as well as microbiome changes that indicate susceptibility and severity [6]. This includes novel detection technologies such as PCR, next-generation sequencing (NGS), CRISPR-based methods [7], immunoassays, lateral flow biosensors, microfluidics, and artificial intelligence (AI), which have revolutionised the sensitivity, speed, and cost of biomarker detection [8]. Biomarkers enable early detection and diagnosis (before symptoms and for prognosis), detection of chronic infections (e.g., HIV, HBV), and assessment of vaccines (e.g., neutralising antibodies as a surrogate marker of protection against COVID-19 [9]). But biological variability, lack of standardisation, assay performance variability, lack of access to new technologies, regulatory hurdles, and global diagnostic inequality still pose challenges [10]. Integration of multi-omics, predictive diagnostics using AI, new CRISPR technologies, point-of-care and home testing, and global monitoring of biomarkers will be necessary for future advances [11].

Literature Review

In recent decades, viral diagnostic technologies have evolved to rapidly meet the demand for sensitive, specific testing to guide outbreak management. Traditional methods (viral culture, microscopy, serology) are informative but not sensitive or fast, and depend on the presence of viable virus. Nucleic acid amplification tests (NAATs), such as PCR and reverse transcription PCR (RT-PCR), are now the preferred methods for detecting HIV, HBV, influenza, and SARS-CoV-2 [4]. Researchers most commonly report pathogen biomarkers such as viral nucleic acids, proteins, and genetic mutations. Clinicians currently use RT-PCR as the best available method for early detection of viral RNA. However, researchers have observed false-negative results during the emergence of variants because primer mismatches can reduce test accuracy [12, 13]. Protein biomarkers such as dengue virus NS1 and HIV p24 remain important for early detection despite their lower analytical sensitivity compared with molecular tests [14, 15]. In the presence of SARS-CoV-2, next-generation sequencing has become a key surveillance tool for identifying variants, detecting antiviral drug resistance, and studying transmission [16, 17]. Biomarkers of host response are now recognised as important for disease severity and prognosis. The association of the most common cytokines (IL-6, TNF-α, IP-10) with inflammation in influenza, Ebola, viral hepatitis and COVID-19 [5, 18]. Lymphopenia, decreased CD4+/CD8+ T cells, dysfunctional NK cells, and exhaustion markers (PD-1, TIM-3) are cellular parameters that reflect immunosuppression in HIV, HCV, and severe COVID-19 [19–21]. The serological markers, especially IgM/IgG levels and neutralising antibodies, have played an important role in vaccine evaluation [9, 22]. Multi-omics technologies provide a more detailed understanding of virus-host interactions. Researchers use transcriptomic interferon-stimulated gene (ISG) signatures, such as OAS1, MX1, and IFIT1, to distinguish between viral and bacterial infections, and multiple studies consistently link dysregulated interferon responses to severe COVID-19 [23, 24]. Proteomics identifies dysregulated complement, coagulation, and acute-phase responses, while metabolomics reveals changes in glycolysis, lipids, and amino acids [25, 26]. Epigenomic studies (DNA methylation, chromatin accessibility) are increasingly important for understanding the outcomes of chronic diseases, such as post-acute COVID-19 and chronic viral infections [27, 28]. Microbiome scientists have linked the depletion of Faecalibacterium prausnitzii with the severity of COVID-19 [29, 30]. Researchers and clinicians can use several technologies to measure biomarkers, including PCR, digital PCR, next-generation sequencing (NGS), CRISPR-based technologies (SHERLOCK and DETECTR), immunoassays, biosensors, and artificial intelligence (AI). Biological variability, standardisation, external validity, cost, regulatory issues and access are among the common problems in this area [3, 31]. These challenges drive multi-site validation, decentralised and cost-effective systems, and global health equity. To conclude, biomarker science has transformed the way viral diagnostics are conducted, but challenges in translation and implementation remain, and continued development of biomarker discovery, integration, and equity is needed.

RESULTS AND DISCUSSION

Comparative Analysis of Biomarker Categories. Translational viral diagnostics rely on biomarkers for early detection, prognosis, therapy monitoring, and surveillance. While each biomarker class is discussed separately in later sections, this section provides a comparative assessment, highlighting similarities and differences, strengths and limitations, diagnostic performance, implications for clinical integration, and reliability and scalability issues.

Table 1 – Comparative Analysis of Viral Biomarker Categories

Biomarker Category	Representative Examples	Detection Methods	Diagnostic Applications	Strengths	Limitations
Pathogen-Derived Nucleic Acids	SARS-CoV-2 RNA, HIV RNA, HBV DNA	RT-PCR, qPCR, Digital PCR, LAMP, NGS	Early detection, confirmatory diagnosis, viral load quantification, occult infections	Very high sensitivity and specificity; detects infection before immune response; quantifies viral burden	Requires lab infrastructure; expensive reagents; contamination-sensitive; false negatives with variant mutations
Pathogen-Derived Proteins/ Antigens	SARS-CoV-2 N protein, HIV p24, Dengue NS1	ELISA, Rapid Antigen Tests, Biosensors	Rapid screening, point-of-care diagnosis, and early detection at high viral load	Fast, cheap, field-deployable; minimal equipment; immediate clinical decisions	Lower sensitivity; reduced accuracy at low viral loads; antigenic drift reduces reliability
Genomic Mutations (Variants, Resistance)	Spike mutations (N501Y, E484K); HIV drug-resistance mutations	Whole-Genome Sequencing, Targeted NGS	Variant surveillance, evolutionary tracking, drug resistance monitoring	Enables public health action; essential for vaccine and drug updates; identifies emerging strains	High cost; requires bioinformatics; uneven global surveillance capacity
Cytokine and Chemokine Biomarkers	IL-6, TNF-α, IP-10	Multiplex cytokine panels, ELISA	Prognosis, severity prediction, therapeutic decisions	Strong correlation with severity; early indication of immune dysregulation	High inter-individual variability; affected by comorbidities; timing-dependent
Cellular Biomarkers	CD4/CD8 counts, NK cell activity, T-cell exhaustion markers	Flow cytometry, functional assays	Disease monitoring, prognosis, and immune competency assessment	Mechanistic insights guide immunotherapies; valuable in chronic infections	Requires specialised labs, expensive instrumentation, and time-consuming
Serological Biomarkers	IgM, IgG, Neutralising antibodies	ELISA, Neutralisation assays, LFAs	Past infection confirmation, vaccine response, and serosurveillance	Essential for population immunity studies; robust for vaccine evaluation	Window periods, variable antibody durability, and lower usefulness in early infection
Transcriptomic Biomarkers	ISGs (OAS1, IFIT1, MX1)	RNA-seq, Host gene expression panels	Discriminating viral vs bacterial infections; early host response profiling	Very high discriminatory power; early detection via host response	Requires sequencing facilities; high computational complexity
Proteomic Biomarkers	Complement proteins, acute-phase proteins	Mass spectrometry, high-throughput proteomics	Severity prediction, mechanistic insights	Reveals deep biological pathways; multi-marker power	High cost; lack of standardised protocols

Biomarker Category	Representative Examples	Detection Methods	Diagnostic Applications	Strengths	Limitations
Metabolomic Biomarkers	Kynurenine, lactate, lipid profiles	LC-MS, NMR spectroscopy	Prediction of severe disease, monitoring metabolic dysregulation	Sensitive to physiologic shifts; integrative with other omics	Highly variable; influenced by diet, microbiome, and comorbidities
Epigenetic Biomarkers	DNA methylation, chromatin accessibility	Bisulfite sequencing, ATAC-seq	Long-term effects, viral latency, prognosis	Stable signals; potential for long-term monitoring	Emerging field; expensive assays; limited clinical validation
Microbiome Biomarkers	Gut microbial taxa, SCFAs	16S rRNA sequencing, shotgun metagenomics	Disease susceptibility prediction, severity correlation	Adds holistic host–microbe insights; emerging therapeutic potential	High interpersonal variability; complex interpretation
AI/ML-Integrated Biomarkers	Predictive panels combining multi-omics	Machine learning platforms	Early diagnosis, risk prediction, decision support	Enables integration of massive datasets; real-time analytics	Requires large training datasets; risk of algorithmic bias

Comparative Discussion of Biomarker Categories. Typically, the difference between biomarker classes lies in the timing of their diagnostic value and their sensitivity to change. Pathogen-derived nucleic acids provide the earliest and most sensitive detection (days before disease symptoms) and are the gold standard for confirmation and quantification. Antigens have the advantage of detecting infection at high viral load, which makes them ideal for screening but not for early diagnosis. Host biomarkers go beyond simple detection to provide signals of immune activation, inflammation, and risk of severity, although host variability, comorbidities, and timing of assessment affect the accuracy of these markers.

Table 2 – Utility Across the Disease Timeline

Infection Stage	Most Useful Biomarkers	Rationale
Pre-symptomatic	Viral RNA (PCR), transcriptomic ISGs	Viral replication is detectable early; host ISGs rise quickly
Acute symptomatic phase	Antigens, cytokines (IL-6, IP-10), viral load	High viral load; immune activation peaks
Severe disease	Cytokines, proteomics, metabolomics	Reveal immune dysregulation and organ involvement
Recovery or chronic stage	Antibodies, epigenetics, cellular markers	Long-term immunity, latency, or immune memory

1) Strengths and Weaknesses by Biomarker Group. Pathogen-Derived Biomarkers are extremely specific for the pathogen and facilitate the genomic monitoring of evolution, emergence and antiviral resistance (very important for HIV and HBV). The major drawbacks of their use are that they can yield false-negative results due to poor sampling or an incorrect primer–variant match, and that they require laboratory infrastructure. Host response biomarkers also reflect disease severity and immune dysfunction, even when the virus is not detectable. When there is inter-individual variability (age, genetics, comorbidities, concurrent inflammatory diseases), it is unlikely that there will be a universal threshold for either prognosis or monitoring. Non-viral conditions may also generate signatures similar to those of viruses, reducing the specificity of these approaches. Multi-omics biomarkers, including transcriptomic, proteomic, metabolomic, epigenetic, and microbiome signatures, provide deep mechanistic insights and enable molecular patient stratification but are costly, resource-intensive, and lack standardised validation pipelines, which limit clinical implementation in the near term.

2) Comparative Clinical Impact. As practical diagnostic tools, pathogen-derived biomarkers remain the most prevalent due to their direct confirmation of infection. PCR and rapid antigen tests are leading point-of-care workflows. A key example is the use of host biomarkers, especially cytokines (e.g., IL-6 and IP-10), as essential prognostic markers in COVID-19 to identify patients

at risk of deterioration. Transcriptomic biomarkers are highly useful for discriminating viral from bacterial infections, thus contributing to antimicrobial stewardship. Proteomic and metabolomic markers have been used to unravel the pathophysiology of COVID-19 and dengue, revealing immune, coagulation, and metabolic changes. Epigenetic and microbiome biomarkers are promising but are only partially established and not yet commonly used in clinical diagnostics.

Table 3 – Translational Readiness of Biomarker Categories

Biomarker Category	Readiness Level
Viral nucleic acids (PCR)	Very High – fully integrated globally
Viral antigens	High – widely used in POC settings
Serology	High – essential for immunity monitoring
Cytokines (e.g., IL-6)	Moderate – used clinically but not universally standardised
Transcriptomics	Moderate – diagnostic potential but costly
Proteomics/ Metabolomics	Low–Moderate – high research value, limited clinical deployment
Epigenetics/ Microbiome	Low – emerging, experimental
AI/ML integrative biomarkers	Growing – dependent on big data and validation

This assessment indicates that pathogen-derived biomarkers remain the most clinically mature, while multi-omics and AI-supported signatures represent the direction of future advances.

Synthesis: Why Multiple Biomarker Categories Are Necessary. There is no single class of biomarkers that provides a comprehensive picture of viral infection, its progression, and outcome. Pathogen-derived markers identify infection; host response markers identify severity, immune system activation, and physiological impact; and multi-omics markers identify mechanistic pathways and predict long-term risk. AI and new platforms are woven into the layers. Comprehensive diagnostic platforms are then the most effective diagnostic frameworks, especially in rapidly evolving pandemics, that incorporate multiple biomarkers to detect patients earlier.

Pathogen-Derived Biomarkers. Pathogen-derived biomarkers, including pathogen presence, replication, and viral genome, are fundamental to viral diagnostics. Viral nucleic acids and proteins facilitate sensitive, specific detection, guide clinical management and enable surveillance [2, 4]. These are the main methods used to sensitively detect RNA viruses (such as SARS-CoV-2 and influenza) and DNA viruses (such as hepatitis B virus [HBV] and cytomegalovirus [CMV]) at low concentrations [32, 33]. RT-PCR is a gold-standard test that can provide a diagnosis earlier than symptoms, before the onset of symptoms. Estimation of viral load helps manage the clinical course, predict outcome, and guide treatment. The viral load of SARS-CoV-2 is associated with symptom severity, whereas viral loads of HIV RNA and HBV DNA are used to guide treatment [34–36]. Digital PCR is superior to conventional PCR in terms of sensitivity for low- and challenging-to-detect samples [37]. Viral proteins are also used as diagnostic markers, such as the SARS-CoV-2 nucleocapsid and spike proteins, HIV p24, and dengue NS1, which help identify active infection at early stages [14, 38]. Antigen rapid diagnostic tests (Ag-RDTs) are point-of-care tests that are less sensitive than other tests [15, 39]. Genomic mutations provide clues to viral evolution, spread and immune evasion. Next-generation sequencing can measure these variants in real time, as seen with COVID [16, 17]. Although genetic mutations do not change a strain's infectivity, they can affect diagnostic efficiency and influence therapeutic strategies, including antiviral resistance testing (avr) [13, 40].

Host-response markers are used in addition to pathogen testing to evaluate immune responses. Inflammation and severity markers include cytokines such as IP-10, IL-6, and TNF-α [41–43]. Type I interferons and interferon-stimulated gene products (OAS1, IFIT1) can distinguish between viral and bacterial infections and their associated severe disease [23, 24]. Changes in immune cells are reflected in cellular biomarkers. T-cell changes occur in response to viral infections, including depletion and exhaustion in chronic infections (HIV, HCV) and the expansion of activated $CD8^+$ T cells in acute infections [20, 44]. Exhaustion markers (PD-1, TIM-3, LAG-3) reflect dysfunctional states. There are also impaired NK cells and lymphopenia, which are linked to severe disease, are easily quantifiable, and therefore a reliable predictor of poor prognosis [19, 21]. Serological markers are indicators of humoral responses. IgM is good for early diagnosis, and IgG for exposure. Neutralising antibodies

correlate with immunity and are essential in immune, therapeutic, and vaccine evaluations [22, 45]. Vaccine effectiveness and booster strategies, as well as the identification of exposure-risk groups, are based on antibody levels and persistence [9].

Multi-Omics and Emerging Biomarkers. Researchers now use multi-omics approaches, including transcriptomics, proteomics, metabolomics, epigenomics, and microbiome analysis, to study host-pathogen interactions. High-throughput technologies enable these approaches by measuring thousands of molecules simultaneously [6]. These tools provide novel markers not found in standard tests. They are increasingly crucial for understanding complex diseases such as long COVID, for precision diagnostics and prognostics, and for the discovery of new therapies. Transcriptomics (RNA-seq) can be used to identify differentially expressed genes during infection, including interferon-stimulated genes (ISGs; e.g., MX1, OAS1, IFIT1), which can distinguish between viral and bacterial infections [23]. Gene signatures derived from machine learning have high diagnostic accuracy, and interferon response dysfunction is linked to disease severity [24, 46]. Single-cell RNA-seq also identifies cell-specific immune defects, and host mRNA panels are being used clinically.

Proteomics uncovers protein signature of inflammation, coagulation and cell damage. In COVID-19, proteins such as CRP, complement, and fibrinogen correlate with disease severity [6, 25]. Influenza, hepatitis C virus, and dengue also exhibit similar protein profiles despite their distinct pathogenic mechanisms. Metabolomics reflects metabolic changes in infections. Viral infections alter processes such as glycolysis and lipid and amino acid metabolism, with metabolites such as lactate and kynurenine associated with disease severity and organ failure [26, 47].

Epigenomics: Infection alters gene expression. Alterations in DNA methylation and chromatin accessibility have been observed in SARS-CoV-2, HIV, and HBV and are associated with immune responses, disease processes, and viral persistence [27, 28]. These may be implicated in long-term effects such as long COVID. The microbiome also impacts viral infections. Loss of microbiota diversity and the absence of protective species are linked to increased disease severity in SARS-CoV-2 and influenza [29, 30].

All these advances rely heavily on technological advances. The current primary approaches to diagnosis are still based on nucleic-acid amplification tests such as PCR and LAMP, and variant monitoring and resistance profiling (variant Surveillance) still rely on next-generation sequencing [32, 48]. Newer technologies, such as CRISPR diagnostics and biosensors, enable rapid, at-the-point-of-care testing [49]. The integration of multi-omics and clinical information is possible through machine learning, which can uncover complex patterns in biomarker data and enhance diagnosis and treatment [11, 46].

Biomarker Discovery to Clinical Implementation. The discovery of meaningful biomarkers using techniques such as transcriptomics, proteomics, metabolomics and serology is the first step toward the development of clinical tools. Identification is based on biological plausibility, reproducibility, clinical significance, sample availability, and sample stability. To minimise attrition, patient variability, and the collection and storage processes, these should be considered early in the pre-analysis stage. Analytical validation confirms assay performance (sensitivity, specificity, limit of detection, precision and robustness), which is important in viral diagnostics as targets can be low and related viruses need to be separated. Clinical validation is the evaluation of the biomarker's ability to distinguish between patient status and outcome (sensitivity and specificity; Receiver Operating Characteristic (ROC) analysis). It is important to have clinical utility (benefit to patients or medical decision-making), and there is evidence from multi-site and external validation studies.

Regulatory endorsement corresponds to contexts of use. Researchers and developers generate evidence for programs such as the FDA Biomarker Qualification Program and diagnostic approval by demonstrating analytical and clinical performance. In an effective translation process, early regulatory engagement supports implementation. Healthcare systems integrate process standardisation, quality assurance, training, and EMR integration into clinical workflows. Developers of point-of-care tests must consider user interface, speed, and cost, particularly in low-resource areas. Post-market monitoring teams ensure performance and adjust diagnostics to account for changes in viral characteristics [50].

Integration of multi-omics and AI-based approaches improves biomarker discovery by

blending data to develop predictive diagnostic and prognostic signatures. However, there are some disadvantages, including data integration, data validation, model interpretability, and assay development. Equality is also on the agenda, and there is a need to make new technologies cost-effective.

Lack of reproducibility, validation and clinical relevance are issues. The task of translation must be carried out in collaboration with other disciplines, with validation and clinical relevance. Adopted, biomarkers play a key role in the management of viral diseases. Nucleic-acid testing helps in early diagnosis and screening, such as SARS-CoV-2 RT-PCR, HIV RNA, HBV DNA, etc., and protein (e.g., dengue NS1, HIV p24) and host-response-based detection during the disease course [46]. Predictive markers of disease severity and outcomes include cellular markers such as lymphopenia, T cell exhaustion markers, markers for NK cells (dysfunction), prognostic markers (e.g., IL-6, CRP, ferritin, IP-10, TNF-α) and metabolomic/proteomic markers of inflammation or tissue damage [5, 18, 25, 26].

Therapy monitoring is based on viral load (gold standard) for HIV, HBV and HCV, and on host markers for immune recovery and response to therapy—the use of IL-6 and D-dimer to guide treatment for COVID-19 [51]. Neutralising antibodies are correlates of protection in vaccine development, while IgG antibodies and T-cell markers indicate immune persistence and T-cell responses [9, 45]. Biomarkers also aid post-vaccination monitoring, such as waning immunity and variant detection, and are under investigation for vaccine safety.

Challenges and Future Prospects. Several obstacles persist. Biomarker expression varies with biological factors (age, genetic makeup, disease, and immune status), and the lack of standardised protocols and reference ranges across laboratories worldwide contributes to variability in reproducibility. The limitations are substantial: PCR, sequencing, multi-omics, and flow cytometry technologies are costly and require more expertise and equipment, which are not available in LMIC countries. Progress is hampered by ethical and regulatory issues - approval times, privacy, and access. High-throughput multi-omics data can be complex to aggregate, subject to batch effects, less well validated, and prone to algorithmic bias. It is necessary to improve standards, technology, and its costs, access, and regulations to solve these problems.

The forecast is still favourable. In the future, clinicians will use integrated signals from pathogens and host immune response readouts for multidimensional diagnostics to enable early detection, severity stratification, and therapeutic intervention. Novel technologies (such as portable sequencing, lab-on-a-chip and wearable biosensors, and CRISPR-based diagnostics) will move testing to the bedside. Advances in artificial intelligence and machine learning will further enhance the identification of biomarkers, predictive modelling, and real-time monitoring, and multiplexed omics will help elucidate mechanisms of acute infection and post-infection complications. Epigenetics, microbiome science, and digital health technologies will expand the range of biomarkers and help build an integrated, cost-effective, and scalable diagnostics platform to improve global readiness.

CONCLUSIONS

Viral diagnostics increasingly rely on biomarkers that provide rapid, reliable information on infection, severity, and therapeutic response. Nucleic acids, proteins, and genetic mutations are viral biomarkers that have enhanced diagnostic specificity and enabled global surveillance of viral evolution. Biomarkers of the host response, such as cytokines, cell signatures and antibodies, give crucial insights into disease course and management. Integrative omics technologies have also uncovered new aspects of viral pathogenesis.

There are limitations: variability, lack of standardisation, costs, and access. Researchers and developers must resolve several issues before they can widely use this technology. Recent developments in multi-omics integration, point-of-care technologies, and AI-based predictive analytics will improve diagnostic speed, scalability, and accuracy. In the future, viral diagnostics will continue to rely heavily on biomarkers, which are essential for personalised medicine and pandemic preparedness.

REFERENCES

1. Zuo, T., Zhang, F., Lui, G. C., Yeoh, Y. K., Li, A. Y., Zhan, H., Wan, Y., Chung, A. C., Cheung, C. P., Chen, N., Lai, C. K., Chen, Z., Tso, E. Y., Fung, K. S., Chan, V., Ling, L., Joynt, G., Hui, D. S., Chan, F. K., & Ng, S. C. (2020). Alterations in gut microbiota of patients with COVID-19 during time of hospitalisation. *Gastroenterology, 159*(3), 944-955. doi: 10.1053/j.gastro.2020.05.048

2. Binnicker, M. J. (2020). Challenges and Controversies to Testing for COVID-19. *Journal of Clinical Microbiology, 58*(11). doi: 10.1128/jcm.01695-20

3. Al-Hasan, M., Mehta, N., Yang, J. D., & Singal, A. G. (2024). Role of biomarkers in the diagnosis and management of HCC. *Liver Transplantation, 31*(3), 384–394. doi: 10.1097/lvt.0000000000000398

4. Teng, C., Thampy, U., Bae, J. Y., Cai, P., Dixon, R. A., Liu, Q., & Li, P. (2021). Identification of phenotypes among COVID-19 patients in the United States using latent class analysis. *Infection and Drug Resistance, 14*, 3865–3871. doi: 10.2147/idr.s331907

5. Del Valle, D. M., Kim-Schulze, S., Huang, H., Beckmann, N. D., Nirenberg, S., Wang, B., Lavin, Y., Swartz, T. H., Madduri, D., Stock, A., Marron, T. U., Xie, H., Patel, M., Tuballes, K., Van Oekelen, O., Rahman, A., Kovatch, P., Aberg, J. A., Schadt, E., & Gnjatic, S. (2020). An inflammatory cytokine signature predicts COVID-19 severity and survival. *Nature Medicine, 26*(10), 1636–1643. doi: 10.1038/s41591-020-1051-9

6. Overmyer, K. A., Shishkova, E., Miller, I. J., Balnis, J., Bernstein, M. N., Peters-Clarke, T. M., Meyer, J. G., Quan, Q., Muehlbauer, L. K., Trujillo, E. A., He, Y., Chopra, A., Chieng, H. C., Tiwari, A., Judson, M. A., Paulson, B., Brademan, D. R., Zhu, Y., Serrano, L. R., & Jaitovich, A. (2020). Large-Scale multi-omic analysis of COVID-19 severity. *Cell Systems, 12*(1), 23-40. doi: 10.1016/j.cels.2020.10.003

7. Patchsung, M., Jantarug, K., Pattama, A., Aphicho, K., Suraritdechachai, S., Meesawat, P., Sappakhaw, K., Leelahakorn, N., Ruenkam, T., Wongsatit, T., Athipanyasilp, N., Eiamthong, B., Lakkanasirorat, B., Phoodokmai, T., Niljianskul, N., Pakotiprapha, D., Chanarat, S., Homchan, A., Tinikul, R., & Uttamapinant, C. (2020). Clinical validation of a Cas13-based assay for the detection of SARS-CoV-2 RNA. *Nature Biomedical Engineering, 4*(12), 1140–1149. doi: 10.1038/s41551-020-00603-x

8. Mao, T., Nan, L., & Shum, H. C. (2024). Digital quantification and ultrasensitive detection of a single influenza virus using Microgel-in-Droplet Enzyme-Linked immunosorbent assay. *Analytical Chemistry, 96*(41), 16134–16144. doi: 10.1021/acs.analchem.4c02429

9. Khoury, D. S., Cromer, D., Reynaldi, A., Schlub, T. E., Wheatley, A. K., Juno, J. A., Subbarao, K., Kent, S. J., Triccas, J. A., & Davenport, M. P. (2021). Neutralising antibody levels are highly predictive of immune protection from symptomatic SARS-CoV-2 infection. *Nature Medicine, 27*(7), 1205–1211. doi: 10.1038/s41591-021-01377-8

10. Cheng, M. P., Papenburg, J., Desjardins, M., Kanjilal, S., Quach, C., Libman, M., Dittrich, S., & Yansouni, C. P. (2020). Diagnostic Testing for Severe Acute Respiratory Syndrome–Related Coronavirus 2. *Annals of Internal Medicine, 172*(11), 726–734. doi: 10.7326/m20-1301

11. Jin, J., & Kim, J. (2024). Multi-omics and Artificial Intelligence technology for elucidating disease pathophysiology. *Preprints.org*. doi: 10.20944/preprints202408.0350.v1

12. Vandenberg, O., Martiny, D., Rochas, O., Van Belkum, A., & Kozlakidis, Z. (2020). Considerations for diagnostic COVID-19 tests. *Nature Reviews Microbiology, 19*(3), 171–183. doi: 10.1038/s41579-020-00461-z

13. Ravi, P., Mantia, C., Su, C., Sorenson, K., Elhag, D., Rathi, N., Bakouny, Z., Agarwal, N., Zakharia, Y., Costello, B. A., McKay, R. R., Narayan, V., Alva, A., McGregor, B. A., Gao, X., McDermott, D. F., & Choueiri, T. K. (2020). Evaluation of the safety and efficacy of immunotherapy rechallenge in patients with renal cell carcinoma. *JAMA Oncology, 6*(10), 1606. doi: 10.1001/jamaoncol.2020.2169

14. Waggoner, J. J., Gresh, L., Vargas, M. J., Ballesteros, G., Tellez, Y., Soda, K. J., Sahoo, M. K., Nuñez, A., Balmaseda, A., Harris, E., & Pinsky, B. A. (2016). Viremia and clinical presentation in Nicaraguan patients infected with the Zika virus, Chikungunya virus, and Dengue virus. *Clinical Infectious Diseases, 63*(12), 1584–1590. doi: 10.1093/cid/ciw589

15. Brümmer, L. E., Katzenschlager, S., Gaeddert, M., Erdmann, C., Schmitz, S., Bota, M., Grilli, M., Larmann, J., Weigand, M. A., Pollock, N. R., Macé, A., Carmona, S., Ongarello, S., Sacks, J. A., & Denkinger, C. M. (2021). Accuracy of novel antigen rapid diagnostics for SARS-CoV-2: A living systematic review and meta-analysis. *PLoS Medicine, 18*(8), e1003735. doi: 10.1371/journal.pmed.1003735

16. Harvey, W. T., Carabelli, A. M., Jackson, B., Gupta, R. K., Thomson, E. C., Harrison, E. M., Ludden, C., Reeve, R., Rambaut, A., Peacock, S. J., & Robertson, D. L. (2021). SARS-CoV-2 variants, spike mutations and immune escape. *Nature Reviews Microbiology, 19*(7), 409–424. doi: 10.1038/s41579-021-00573-0

17. Khan, S., Yahiro, T., Kimitsuki, K., Hashimoto, T., Matsuura, K., Yano, S., Noguchi, K., Sonezaki, A., Yoshizawa, K., Kumasako, Y., Akbar, S. M. F., & Nishizono, A. (2024). Exploring the replication and pathogenic characteristics of alpha, delta, and Omicron variants of SARS-COV-2. *International Journal of Molecular Sciences, 25*(23), 12641. doi: 10.3390/ijms252312641

18. Gavkare, A. M., Nanaware, N. L., Sonar, M. N., Dhotre, S. V., Mumbre, S. S., & Nagoba, B. S. (2025). Gut microbiome and viral infections: A hidden nexus for immune protection. *World Journal of Virology, 14*(3), 111912. doi: 10.5501/wjv.v14.i3.111912

19. Maucourant, C., Filipovic, I., Ponzetta, A., Aleman, S., Cornillet, M., Hertwig, L., Strunz, B., Lentini, A., Reinius, B., Brownlie, D., Cuapio, A., Ask, E. H., Hull, R. M., Haroun-Izquierdo, A., Schaffer, M., Klingström, J., Folkesson, E., Buggert, M., Sandberg, J. K., & Group, K. C. S. (2020). Natural killer cell immunotypes related to COVID-19 disease severity. *Science Immunology, 5*(50). doi: 10.1126/sciimmunol.abd6832

20. Gaebler, C., Wang, Z., Lorenzi, J. C. C., Muecksch, F., Finkin, S., Tokuyama, M., Cho, A., Jankovic, M., Schaefer-Babajew, D., Oliveira, T. Y., Cipolla, M., Viant, C., Barnes, C. O., Bram, Y., Breton, G., HäGglöF, T., Mendoza, P., Hurley, A., Turroja, M., & Nussenzweig, M. C. (2021). Evolution of antibody immunity to SARS-CoV-2. *Nature, 591*(7851), 639–644. doi: 10.1038/s41586-021-03207-w

21. Tan, L., Wang, Q., Zhang, D., Ding, J., Huang, Q., Tang, Y., Wang, Q., & Miao, H. (2020). Lymphopenia predicts COVID-19 disease severity: a descriptive and predictive study. *Signal Transduction and Targeted Therapy, 5*(1), 33. doi: 10.1038/s41392-020-0148-4

22. Post, M. J., Levenberg, S., Kaplan, D. L., Genovese, N., Fu, J., Bryant, C. J., Negowetti, N., Verzijden, K., & Moutsatsou, P. (2020). Scientific, sustainability and regulatory challenges of cultured meat. *Nature Food, 1*(7), 403–415. doi: 10.1038/s43016-020-0112-z

23. Arunachalam, P. S., Wimmers, F., Mok, C. K. P., Perera, R. a. P. M., Scott, M., Hagan, T., Sigal, N., Feng, Y., Bristow, L., Tsang, O. T., Wagh, D., Coller, J., Pellegrini, K. L., Kazmin, D., Alaaeddine, G., Leung, W. S., Chan, J. M. C., Chik, T. S. H., Choi, C. Y. C., & Pulendran, B. (2020). Systems biological assessment of immunity to mild versus severe COVID-19 infection in humans. *Science, 369*(6508), 1210–1220. doi: 10.1126/science.abc6261

24. Hadjadj, J., Yatim, N., Barnabei, L., Corneau, A., Boussier, J., Smith, N., Péré, H., Charbit, B., Bondet, V., Chenevier-Gobeaux, C., Breillat, P., Carlier, N., Gauzit, R., Morbieu, C., Pène, F., Marin, N., Roche, N., Szwebel, T., Merkling, S. H., & Terrier, B. (2020). Impaired type I interferon activity and inflammatory responses in severe COVID-19 patients. *Science, 369*(6504), 718–724. doi: 10.1126/science.abc6027

25. Messner, A. H., Walsh, J., Rosenfeld, R. M., Schwartz, S. R., Ishman, S. L., Baldassari, C., Brietzke, S. E., Darrow, D. H., Goldstein, N., Levi, J., Meyer, A. K., Parikh, S., Simons, J. P., Wohl, D. L., Lambie, E., & Satterfield, L. (2020). Clinical Consensus Statement: Ankyloglossia in children. *Otolaryngology, 162*(5), 597–611. doi: 10.1177/0194599820915457

26. Wu, T., Jia, X., Shi, H., Niu, J., Yin, X., Xie, J., & Wang, X. (2020). Prevalence of mental health problems during the COVID-19 pandemic: A systematic review and meta-analysis. *Journal of Affective Disorders, 281*, 91–98. doi: 10.1016/j.jad.2020.11.117

27. Balnis, J., Madrid, A., Drake, L. A., Vancavage, R., Tiwari, A., Patel, V. J., Ramos, R. B., Schwarz, J. J., Yucel, R., Singer, H. A., Alisch, R. S., & Jaitovich, A. (2024). Blood DNA methylation in post-acute sequelae of COVID-19 (PASC): a prospective cohort study. *EBioMedicine, 106*, 105251. doi: 10.1016/j.ebiom.2024.105251

28. Süt, B. B. (2021). Epigenetic regulation mechanisms in viral infections: A Special focus on COVID-19. In *IntechOpen eBooks*. doi: 10.5772/intechopen.98866

29. Zuo, F., Gao, J., Kurkcu, A., Yang, H., Ozbay, K., & Ma, Q. (2021). Reference-free video-to-real distance-approximation-based urban social distancing analytics amid the COVID-19 pandemic. *Journal of Transport & Health, 21*, 101032. doi: 10.1016/j.jth.2021.101032

30. De Steenhuijsen Piters, W. A. A., Heinonen, S., Hasrat, R., Bunsow, E., Smith, B., Suarez-Arrabal, M., Chaussabel, D., Cohen, D. M., Sanders, E. A. M., Ramilo, O., Bogaert, D., & Mejias, A. (2016). Nasopharyngeal Microbiota, Host Transcriptome, and Disease Severity in Children with Respiratory Syncytial Virus Infection. *American Journal of Respiratory and Critical Care Medicine, 194*(9), 1104–1115. doi: 10.1164/rccm.201602-0220oc

31. Narayanasamy, S., Okware, B., Muttamba, W., Patel, K., Duedu, K. O., Ravi, N., Ellermeier, N., Shey, M., Woods, C. W., & Sabiiti, W. (2022). Global inequity of COVID-19 diagnostics: challenges and opportunities. *Journal of Epidemiology & Community Health, 76*(12), 972–975. doi: 10.1136/jech-2022-219333

32. Mautner, L., Baillie, C., Herold, H. M., Volkwein, W., Guertler, P., Eberle, U., Ackermann, N., Sing, A., Pavlovic, M., Goerlich, O., Busch, U., Wassill, L., Huber, I., & Baiker, A. (2020). Rapid point-of-care detection of SARS-CoV-2 using reverse transcription loop-mediated isothermal amplification (RT-LAMP). *Virology Journal, 17*(1), 160. doi: 10.1186/s12985-020-01435-6

33. Wang, C., Horby, P. W., Hayden, F. G., & Gao, G. F. (2020). A novel coronavirus outbreak is a global health concern. *The Lancet, 395*(10223), 470–473. doi: 10.1016/s0140-6736(20)30185-9

34. UNAIDS. (2021). 2021 UNAIDS Global AIDS Update — Confronting inequalities — Lessons for pandemic responses from 40 years of AIDS. Retrieved from https://www.unaids.org/en/resources/documents/2021/2021-global-aids-update

35. Terrault, Z. (2023). La maltraitance psychologique envers les enfants pendant la pandémie de COVID-19. *Psycause Revue Scientifique Étudiante De L École De Psychologie De L Université Laval, 13*(1), 23–31. doi: 10.51656/psycause.v13i1.51845

36. Pujadas, E., Ibeh, N., Hernandez, M. M., Waluszko, A., Sidorenko, T., Flores, V., Shiffrin, B., Chiu, N., Young-Francois, A., Nowak, M. D., Paniz-Mondolfi, A. E., Sordillo, E. M., Cordon-Cardo, C., Houldsworth, J., & Gitman, M. R. (2020). Comparison of SARS-CoV-2 detection from nasopharyngeal swab samples by the Roche cobas 6800 SARS-CoV-2 test and a laboratory-developed real-time RT-PCR test. *Journal of Medical Virology, 92*(9), 1695–1698. doi: 10.1002/jmv.25988

37. Alteri, C., Cento, V., Antonello, M., Colagrossi, L., Merli, M., Ughi, N., Renica, S., Matarazzo, E., Di Ruscio, F., Tartaglione, L., Colombo, J., Grimaldi, C., Carta, S., Nava, A., Costabile, V., Baiguera, C., Campisi, D., Fanti, D., Vismara, C., & Perno, C. F. (2020). Detection and quantification of SARS-CoV-2 by droplet digital PCR in real-time PCR-negative nasopharyngeal swabs from suspected COVID-19 patients. *PLoS ONE, 15*(9), e0236311. doi: 10.1371/journal.pone.0236311

38. Guglielmi, G. (2020). Fast coronavirus tests: what they can and can't do. *Nature, 585*(7826), 496–498. doi: 10.1038/d41586-020-02661-2

39. Peeling, R. W., Olliaro, P. L., Boeras, D. I., & Fongwen, N. (2021). Scaling up COVID-19 rapid antigen tests: promises and challenges. *The Lancet Infectious Diseases, 21*(9), 290–295. doi: 10.1016/s1473-3099(21)00048-7

40. Liu, Q., Chi, S., Dmytruk, K., Dmytruk, O., & Tan, S. (2022). Coronaviral infection and interferon response: The Virus-Host Arms Race and COVID-19. *Viruses, 14*(7), 1349. doi: 10.3390/v14071349

41. Herold, T., Jurinovic, V., Arnreich, C., Lipworth, B. J., Hellmuth, J. C., Von Bergwelt-Baildon, M., Klein, M., & Weinberger, T. (2020). Elevated levels of IL-6 and CRP predict the need for mechanical ventilation in COVID-19. *Journal of Allergy and Clinical Immunology, 146*(1), 128-136. doi: 10.1016/j.jaci.2020.05.008

42. Karki, R., & Kanneganti, T. (2021). The 'cytokine storm': molecular mechanisms and therapeutic prospects. *Trends in Immunology, 42*(8), 681–705. doi: 10.1016/j.it.2021.06.001

43. Hayney, M. S., Henriquez, K. M., Barnet, J. H., Ewers, T., Champion, H. M., Flannery, S., & Barrett, B. (2017). Serum IFN-γ-induced protein 10 (IP-10) as a biomarker for the severity of acute respiratory infection in healthy adults. *Journal of Clinical Virology, 90*, 32–37. doi: 10.1016/j.jcv.2017.03.003

44. Moderbacher, C. R., Ramirez, S. I., Dan, J. M., Grifoni, A., Hastie, K. M., Weiskopf, D., Belanger, S., Abbott, R. K., Kim, C., Choi, J., Kato, Y., Crotty, E. G., Kim, C., Rawlings, S. A., Mateus, J., Tse, L. P. V., Frazier, A., Baric, R., Peters, B., & Crotty, S. (2020). Antigen-Specific Adaptive Immunity to SARS-CoV-2 in Acute COVID-19 and Associations with Age and Disease Severity. *Cell, 183*(4), 996-1012. doi: 10.1016/j.cell.2020.09.038

45. Dan, J. M., Mateus, J., Kato, Y., Hastie, K. M., Yu, E. D., Faliti, C. E., Grifoni, A., Ramirez, S. I., Haupt, S., Frazier, A., Nakao, C., Rayaprolu, V., Rawlings, S. A., Peters, B., Krammer, F., Simon, V., Saphire, E. O., Smith, D. M., Weiskopf, D., & Crotty, S. (2021). Immunological memory to SARS-CoV-2 assessed for up to 8 months after infection. *Science, 371*(6529). doi: 10.1126/science.abf4063

46. Tsalik, E. L., Henao, R., Montgomery, J. L., Nawrocki, J. W., Aydin, M., Lydon, E. C., Ko, E. R., Petzold, E., Nicholson, B. P., Cairns, C. B., Glickman, S. W., Quackenbush, E., Kingsmore, S. F., Jaehne, A. K., Rivers, E. P., Langley, R. J., Fowler, V. G., McClain, M. T., Crisp, R. J., & Woods, C. W. (2021). Discriminating bacterial and viral infection using a rapid host gene expression test. *Critical Care Medicine, 49*(10), 1651–1663. doi: 10.1097/ccm.0000000000005085

47. Shen, C., Wang, Z., Zhao, F., Yang, Y., Li, J., Yuan, J., Wang, F., Li, D., Yang, M., Xing, L., Wei, J., Xiao, H., Yang, Y., Qu, J., Qing, L., Chen, L., Xu, Z., Peng, L., Li, Y., & Liu, L. (2020). Treatment of 5 critically ill patients with COVID-19 with convalescent plasma. *JAMA, 323*(16), 1582. doi: 10.1001/jama.2020.4783

48. Chiu, C. Y., & Miller, S. A. (2019). Clinical metagenomics. *Nature Reviews Genetics, 20*(6), 341–355. doi: 10.1038/s41576-019-0113-7

49. Kellner, M. J., Koob, J. G., Gootenberg, J. S., Abudayyeh, O. O., & Zhang, F. (2019). SHERLOCK: nucleic acid detection with CRISPR nucleases. *Nature Protocols, 14*(10), 2986–3012. doi: 10.1038/s41596-019-0210-2

50. FDA. (2018). Biomarker Qualification: Evidentiary Framework Guidance for Industry and FDA Staff. Retrieved from https://www.fda.gov/media/119271/download

51. Matsumoto, H., Kikuchi, S., Murata, S., Ohshita, M., Harima, Y., Annen, S., Mukai, N., Nakabayashi, Y., Ogawa, S., Okita, M., Takeba, J., & Sato, N. (2023). Dynamic coagulofibrinolytic responses under long-term VV-ECMO management without anticoagulation in a COVID-19-ARDS patient: A case report. *Medicine, 102*(4), 32817. doi: 10.1097/md.0000000000032817