

9 Challenges and Future Perspectives

9.1 Lack of standardized microbial health indicators

The largest conceptual obstacle is the absence of a universal microbial definition of health. Species identity, developmental stage, diet, tissue, salinity, geographic location, season, and farm design all influence microbiome composition. Reviews of aquaculture dysbiosis consistently find recurring patterns, such as pathogen enrichment and community restructuring, but not a single taxonomic profile that defines healthy fish across systems (Mougin and Joyce, 2023; Xavier et al., 2024).

Standardization should therefore focus first on measurement and interpretation rather than universal taxa. Comparable sampling procedures, metadata, controls, sequence processing, absolute abundance measurements, and outcome definitions would make it easier to identify indicators that genuinely transfer across farms.

9.2 Relative abundance versus absolute quantification

Amplicon sequencing is compositional. If one organism expands strongly, the apparent relative abundance of all other organisms may decline even when their absolute cell numbers remain unchanged. This makes relative abundance alone potentially misleading for health decisions.

Combining community sequencing with total-cell measurements, spike-in standards, qPCR, or digital PCR can address part of this problem. Digital PCR is particularly attractive for operational biomarkers because it provides absolute target quantification, while viability treatments can further distinguish viable-cell signals in some applications (Yang et al., 2023). Future health indices should report absolute microbial loads whenever the biological question concerns infection pressure rather than community composition alone.

9.3 From association to causality

A dysbiotic microbiome may be a cause of disease, a consequence of disease, or a correlated response to the same environmental stressor. Distinguishing these possibilities requires temporal and experimental evidence. Shrimp research using microbiota manipulation has provided unusually strong evidence that dysbiosis can contribute to disease processes, but such causal demonstrations remain far less common than cross-sectional associations (Huang et al., 2020).

Future biomarker development should therefore proceed in stages: longitudinal association, independent replication, mechanistic testing, and finally prospective prediction. Organisms identified only because they dominate moribund animals should not be promoted immediately as early-warning biomarkers.

9.4 Need for long-term and multi-farm monitoring

A model that performs well on samples from one farm may simply recognize that farm. Seasonal patterns can create the same problem: an algorithm may inadvertently learn sampling date rather than disease risk. The Sanggou Bay study demonstrates how strong seasonality can be relative to culture-mode effects, while barramundi hatchery observations reveal persistent differences among tanks managed within the same facility (Lu et al., 2025; Bui et al., 2026).

Robust validation therefore requires several production cycles, multiple farms, independent cohorts, and deliberately separated training and test data. Shared longitudinal datasets linking microbiomes with environmental variables and standardized health outcomes would probably advance the field more than increasingly complex algorithms trained on small local datasets.

9.5 Standardization and cost reduction

Differences in filtration volume, sample storage, DNA extraction, primer choice, sequencing depth, bioinformatic pipelines, and taxonomic databases can all change the apparent microbiome. Operational monitoring adds further constraints: methods must be fast, reproducible, affordable, and usable by non-specialist personnel.

One realistic route is a two-tier technology system. Research laboratories could use sequencing and multi-omics to discover and validate indicators, while farms employ simplified qPCR, dPCR, flow-cytometry, LAMP, or biosensor