

illustrating how viability and absolute quantification can be incorporated into environmental surveillance (Yang et al., 2023).

The limitation of targeted assays is equally clear: they detect what investigators decide to search for. A well-designed qPCR panel can be highly useful when key pathogens and virulence markers are known, but it cannot describe an unexpected community-wide shift. For health management, targeted PCR is therefore best positioned as a rapid confirmation layer within a broader monitoring program.

3.3 16S rRNA gene amplicon sequencing

16S rRNA gene sequencing remains one of the most accessible approaches for characterizing bacterial and archaeal communities. It can compare alpha diversity, community dissimilarity, taxonomic composition, succession, and candidate indicator taxa across time, farms, tissues, or health states. Longitudinal shrimp, salmon, oyster, and barramundi studies demonstrate how these profiles can reveal microbial changes that are invisible to routine culture or single-target assays (Kim et al., 2022; Cram et al., 2024).

Its interpretation requires restraint. Amplicon sequencing normally reports relative rather than absolute abundance; taxonomic resolution can be insufficient to distinguish pathogenic and non-pathogenic strains; primer choice and DNA extraction introduce bias; and detection of bacterial DNA does not prove viability or virulence. Bui et al. (2026) found that 16S sequencing and flow-cytometric community fingerprints captured complementary aspects of microbial dynamics in commercial barramundi larviculture, reinforcing the value of combining rather than substituting methods.

3.4 Metagenomic and metatranscriptomic approaches

Shotgun metagenomics extends monitoring from marker genes to the broader genetic potential of a community. It can provide improved taxonomic resolution and detect genes related to metabolism, virulence, nutrient cycling, and antimicrobial resistance. In marine cage aquaculture, metagenomic analysis has revealed differences in both taxonomic composition and carbon-, nitrogen-, and sulfur-related functional profiles between aquaculture and nearby non-aquaculture waters (Liu et al., 2024). Such information may eventually help distinguish a harmless taxonomic shift from a functionally consequential one.

Metatranscriptomics adds another dimension by measuring expressed RNA and thus providing a closer view of microbial activity. Its practical barriers are cost, RNA instability, bioinformatic complexity, and the difficulty of separating biologically meaningful signals from short-term environmental responses. These approaches are therefore most valuable today for biomarker discovery and mechanistic research rather than routine monitoring of every production unit.

3.5 Multi-Omics approaches to microbial health monitoring

No single omics layer fully describes host–microbe–environment interactions. Metagenomics indicates functional potential, metatranscriptomics identifies active expression, metabolomics characterizes chemical outputs, and proteomics can reveal expressed proteins. Integrating these layers may therefore separate taxonomic change from functional change and identify mechanisms that would remain hidden in a 16S dataset alone.

This potential is illustrated by integrated metagenomic and metabolomic analysis of *Litopenaeus vannamei* exposed to microcystin-LR, where microbial functional shifts were examined together with altered intestinal metabolites (Duan et al., 2022). The study also illustrates a broader caution: multi-omics produces many associations, but candidate biomarkers still require experimental validation before becoming operational health indicators.

3.6 Rapid and on-site microbial detection technologies

Farm health decisions often require information faster than conventional sequencing can provide. Isothermal amplification, portable nucleic-acid platforms, biosensors, microfluidics, and high-throughput flow cytometry are attractive because they can reduce analytical turnaround. A LAMP assay developed for *Vibrio harveyi* demonstrated rapid target detection under isothermal conditions, and a more recent direct LAMP workflow for *V.*