Salient Characteristics

Compatibility with Existing Assays and Laboratory Workflows.

- The system must support TaqMan® hydrolysis probe assays, SYBR™ Green dye-based assays, and standard PCR master mixes routinely used in the laboratory.

Analytical Performance.

- The system must reliably quantify targets over at least a 7-log dynamic range (approximately 10^1–10^7 template copies) with quantitative accuracy ($R^2 \geq 0.99$ on standard curves) and detect low-copy targets approaching 10 copies per reaction, depending on assay performance, to detect subtle differences in gene expression associated with wildlife health, disease progression, and environmental stressors.
- It must support multiplex detection of up to six fluorescent targets within a single reaction, allowing simultaneous measurement of multiple biomarkers while maintaining high quantitative precision.
- The instrument must provide independently controlled thermal zones capable of optimizing annealing temperatures across a range of approximately 50–70°C with temperature uniformity sufficient to minimize well-to-well variation.
- The system must support reaction volumes from 10–50 µL in 384-well optical plates.
- Six-color multiplex fluorescence detection
- Excitation/emission filters compatible with FAM, VIC, JUN, ABY, TAMRA, Cy5, and ROX (or equivalent fluorophores).
- 384-well throughput.
- Ability to perform absolute quantification, relative quantification (ΔΔCt), melt curve analysis (if using SYBR), and allelic discrimination.

Software and Data Management.

The instrument must include integrated software for experiment design, instrument control, quality assurance, data analysis, and export of results. The software must support standardized analysis workflows that are compatible with existing laboratory data processing pipelines and enable consistent comparison of newly generated data with historical datasets. The system should minimize additional staff training and support efficient transfer of data into downstream statistical and bioinformatic analyses.

- The software must be compatible with Applied Biosystems QuantStudio Design and Analysis Software and support export formats compatible with existing R-based statistical analysis workflows and laboratory quality-control procedures.

Long-Term Data Standardization.

The laboratory conducts long-term wildlife health monitoring and transcriptomic studies that require direct comparison of gene expression data collected over multiple years. The system must provide highly reproducible performance and standardized analytical outputs to minimize instrument-related variability and ensure continuity across federally funded monitoring programs and longitudinal datasets.

Operational Efficiency and Reliability.
The system must be capable of continuous daily operation supporting multiple consecutive 384-well runs (8–12 hours/day) without loss of performance or calibration. It should include automated calibration, quality control, and performance monitoring capabilities that reduce instrument downtime and ensure reliable operation in a research laboratory environment.

Service and Technical Support.
The manufacturer must provide comprehensive technical support, instrument calibration, software maintenance, and repair services throughout the operational life of the instrument. Factory-authorized service and readily available replacement parts are necessary to maintain instrument performance, minimize downtime, and ensure uninterrupted support of ongoing research activities.

A real-time PCR system meeting these functional requirements is essential for supporting current and future research in wildlife disease surveillance, transcriptomic biomarker validation, and molecular diagnostic assay development while maintaining consistency with established laboratory methods and long-term monitoring objectives.

- Optical detection calibrated for low-abundance transcript detection.
- Calibration traceable to manufacturer standards.