Application Information Bulletin: AQUIOS CL System

Error Prevention on the AQUIOS CL System for the Tetra Application

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INTRODUCTION

Laboratories follow procedures defined by laboratory standards and regulations set forth by regulatory bodies such as the Centers for Disease Control (CDC) among others. The procedures are in place as safety countermeasures and as error prevention methods. The work behind error prevention can be expensive and time consuming. The AQUIOS CL flow cytometer design has taken this into account in as many areas as possible. The AQUIOS CL flow cytometer has several systems built-in to prevent failures.

The AQUIOS CL flow cytometer has error prevention systems to assist in:

- General workflow
- Startup, cleaning, and worklist generation
- Sample preparation
- Quality control

The sections below provide an overview of each of the error prevention systems in the . . . *at a Glance* tables at the start of each section. Each error prevention is then explained in further detail below the table.

ERROR PREVENTION FROM START TO FINISH ON THE AQUIOS CL SYSTEM

The AQUIOS CL system employs several features that work to prevent errors throughout the entire workflow from start to finish. These prevention methods can be broken down into the following:

Table 1 – Error Prevention from Start to Finish at a Glance

Error Prevention Method	Traditional Flow Cytometer	AQUIOS CL Flow Cytometer	Description
Specimen barcodes	Manual scanning	Automated scanning	**Traditional flow cytometer:** Many traditional flow cytometers do not use barcodes. Others use a handheld barcode scanner requiring the user to manually scan each barcode. **AQUIOS CL flow cytometer:** Within the given specifications, the tube barcode reader can automatically read barcodes without the use of a handheld barcode scanner.
Positive Specimen ID (patient and specimen) monitoring	Manual	Automated tracking	**Traditional flow cytometer:** The specimen ID must be linked to the daughter tube ID. These barcodes are traditionally entered into the system manually and tracked. The operator is responsible for identifying and tracking the tube throughout the sample run. **AQUIOS CL flow cytometer:** The AQUIOS CL flow cytometer automatically matches the barcode on the tube to the LIS request. The Specimen ID is recorded immediately before specimen aspiration to prevent misidentification. This Specimen ID is automatically tracked throughout the run.
Remote User Alerts	N/A	Automated	**Traditional flow cytometer:** Remote user alerts are not available on traditional flow cytometers. **AQUIOS CL flow cytometer:** Once this feature is set up, the system automatically sends text and e-mail alerts when the system encounters an error.
Smart-Track reagent monitoring	N/A	Automated tracking	**Traditional flow cytometer:** Smart-Track reagent monitoring is not available on traditional sample prep devices. Traditional systems do not detect if an incorrect antibody bottle was used. **AQUIOS CL flow cytometer:** AQUIOS reagents are labeled with 2D barcodes for smart-track reagent monitoring. The system will not run a Test unless it finds the necessary antibody vials.
Scanning control assay ranges	Manual entry OR N/A	Barcode scanning OR Automated	**Traditional flow cytometer:** The user must manually enter the control ranges. **AQUIOS CL flow cytometer:** Control ranges are scanned to minimize data entry errors.
Locked system	Open	Locked	**Traditional flow cytometer:** In some cases users can change voltages and gains, and more. In other cases, systems have features where these settings can be automatically changed on a daily basis. **AQUIOS CL flow cytometer:** The system is locked to ensure that the instrument setup is consistent.

Specimen Barcodes

Unlike traditional flow cytometers, all compatible barcodes on the AQUIOS CL flow cytometer can be automatically read by the tube barcode reader without the need for a handheld barcode scanner, reducing the need to manually scan specimens. Refer to Appendix D, Barcode Specifications in the AQUIOS CL Flow Cytometer Instructions for Use (PN B21496).

Positive Specimen ID (Patient and Specimen) Monitoring

On a traditional flow cytometer, barcode scanners are used to scan both the daughter tube and the specimen into the system. This workflow has the potential for sample misidentification if an operator scans the incorrect daughter tube and/or specimen and places it in the incorrect location in the rack or carousel.

The AQUIOS CL flow cytometer reduces the possibility of sample misidentification by scanning the specimen tube right before aspiration. This form of error prevention, although new to flow cytometry, was borrowed from hematology instruments such as the DxH800.

Remote User Alerts

In current flow cytometry laboratories [1,2], an instrument operator sets up the cytometer and monitors it as the sample is run. This also implies that instruments typically have dedicated operators who need to be available to periodically monitor the instrument.

The AQUIOS CL flow cytometer monitors the sample in real time and, if three consecutive runs are flagged, it can be set up to notify the operator by a text message or an e mail. Once notified, the operator can address the issue right away. This ability allows an operator to run multiple instruments, perform other tasks in the laboratory as samples are being processed and walk away knowing that the flow cytometer can notify the operator of any significant issues. This is unique to the AQUIOS CL flow cytometer and not available on any other flow cytometer.

The AQUIOS CL flow cytometer also tracks a variety of errors throughout the workflow. Refer to Chapter 9, Troubleshooting in the AQUIOS CL Flow Cytometer Instructions for Use for a list of errors and the relevant messages tracked by the system.

Smart-Track Reagent Monitoring

Smart-Track reagent monitoring is not available on traditional sample prep devices. Traditional sample prep systems do not detect if an incorrect antibody bottle was used. Conversely, the AQUIOS system has reagents that use a unique 2D barcode identity for tracking the expiration date, on board expiration, lot, and container numbers and links this information to the sample. The reagent consumption and plate usage are monitored by the system as the samples are processed. Updates of new reagents, plate usage, reagent availability and reagent location are displayed after opening and closing the reagent door.

NOTE: The AQUIOS CL Flow Cytometer system does not track the AQUIOS Sodium Hypochlorite Solution. You must track the expiration date of the AQUIOS Sodium Hypochlorite Solution upon use.

Consumables maintained on board are automatically read into the system when placed onboard. Barcoded reagents that are not housed onboard can be read into the system by presenting the barcode on the label to the external barcode reader.

Expiration dates and reagent quantity are both measured in real time. The Reagent Status page displays the amount of remaining reagent and the number of tests remaining or the remaining reagent volume (in %). Upon startup, the system displays how many tests remain before replacement is required. When a replacement is required, the system displays one of two warnings: the reagents are either low or depleted.

Scanning Control Assay Ranges

Traditional systems require that control assay ranges be input manually. This is time consuming and there is a risk of data entry errors. Since the AQUIOS CL system monitors and tracks the barcodes in real time by scanning upon loading and scanning before aspiration, samples can be loaded into the cassettes in any order and the cassettes can be loaded onto the system in any order. There is no longer a need to track a complex worklist with multiple tubes for the specimen and the daughter tube. And, there is no need to manually track the location of each daughter tube in the system. This reduces the risk of sample misidentification.

Locked System

All settings are locked except for the following: ability to adjust gates and regions* and ability to adjust compensation (not done on a regular basis). A locked system prevents user error due to erroneous setting selections or any unintended changes to the settings. A locked system makes it easy to use and allows less experienced operators to be trained to run the system.

** When adjusting regions, the system keeps a history of changes made and those changes can be undone.*

ERROR PREVENTION STARTUP/CLEANING/WORKLIST GENERATION ON THE AQUIOS CL SYSTEM

The AQUIOS CL system employs several features that work to prevent errors during startup, cleaning, and worklist generation. These prevention methods can be broken down into the following:

Table 2 - Error Prevention Startup/Cleaning/Worklist Generation at a Glance

Error Prevention Method	Traditional Flow Cytometer	AQUIOS CL Flow Cytometer	Description
Autocleaning	N/A	Automated	**Traditional flow cytometer:** Although traditional flow cytometers do not support a detection system to determine when to clean, some traditional flow cytometers offer an automatic priming function. **AQUIOS CL flow cytometer:** The system automatically monitors and detects potential clogs and/or bubbles in the flow cell. When a clog is identified, the run is automatically aborted and the system begins an autocleaning cycle. Once complete, the system will rerun the prepared sample.
Test Requests to and from the LIS	Manual/ Semi-Automated	Automated	**Traditional flow cytometer:** Requires either manual entry or middleware to transfer test requests from the LIS. When using middleware, the user is responsible for manually linking the specimen to the test request. **AQUIOS CL flow cytometer:** Test requests can be transferred to and from the LIS in three ways: • Manually: Used for controls. • Broadcast download: All appropriate test requests are automatically sent from the LIS to the AQUIOS CL flow cytometer. When the system reads the specimen barcode, it will link it to the test request that is already on the AQUIOS CL system. • Host Query/Dynamic Download: Once the system reads the specimen barcode, the system will query the LIS to read the specific test request.

Error Prevention Method	Traditional Flow Cytometer	AQUIOS CL Flow Cytometer	Description
Creating worklists	Semi-Automated	Automated	**Traditional flow cytometer:** Worklists must be manually copied into the system or require middleware to automatically generate worklists. Manually created worklists typically are verified by a second user for accuracy. **AQUIOS CL flow cytometer:** When the LIS sends test requests to the AQUIOS CL system, it captures the information provided and automatically generates a worklist.

Autocleaning

Autocleaning is a function integrated directly into the AQUIOS system. The AQUIOS CL flow cytometer continually monitors and detects potential clogs and/or air bubbles in the flow cell. Upon detection of a potential clog or bubble in the flow cell, the system aborts the sample run and activates an autocleaning cycle. Once this cycle is complete, the system will rerun the aborted sample. Autocleaning reduces the potential for low precision and erroneous results. In addition, autocleaning reduces the need for sample reruns.

Test Requests to and from the LIS

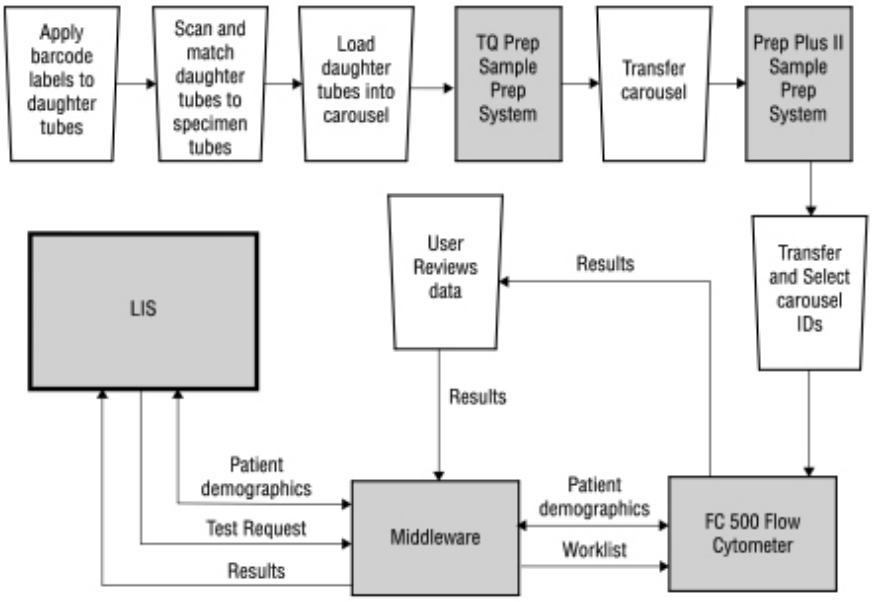
Traditional flow cytometers do not automatically transfer test requests to and from the LIS without the added expense of middleware. If the LIS connection is enabled on the AQUIOS CL flow cytometer, the system automatically retrieves the test request from the LIS. Users with Reviewer access are authorized to review and transmit samples to the LIS. Reviewer users are able to generate and perform test requests, and review and release results.

Not only does the built-in LIS ensure automatic transfer of test requests reducing transcription errors, but no middleware is required. Different user access levels ensure that operators can only work on areas that align with their level of experience with the system.

Creating Worklists

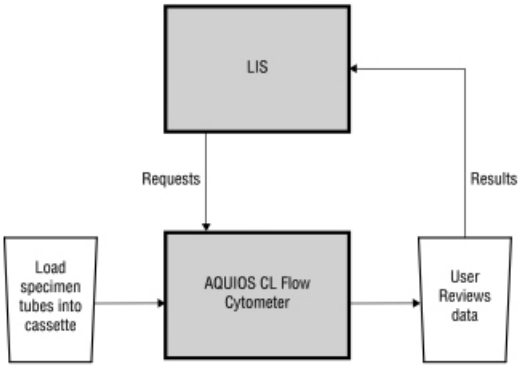
On a traditional flow cytometer, there is no direct connection from the worklist to sample preparation system, or sample preparation system to the analyzer. Traditional flow cytometers automatically send the results from the analyzer to the LIS and the test request from the middleware to the analyzer when using middleware. Potential errors may result because everything is not linked together. Refer to Figure 1 for a visual representation of an LIS connection using the FC 500 flow cytometer and TQ-Prep sample prep system.

Figure 1 - LIS Connection for FC 500 Flow Cytometer and TQ-Prep Sample Prep System



Conversely, the AQUIOS CL flow cytometer is one seamless system; no middleware is required. However, middleware can be used if desired. The system is able to send test requests to the analyzer and capture the information to automatically generate a worklist. Refer to Figure 2 for a visual representation of the LIS connection for the AQUIOS CL flow cytometer.

Figure 2 - LIS Connection for the AQUIOS CL Flow Cytometer



ERROR PREVENTION SAMPLE PREP ON THE AQUIOS CL SYSTEM

The AQUIOS CL system employs several features that work to prevent errors during sample prep. These prevention methods can be broken down into the following:

Table 3 - Error Prevention Sample Prep at a Glance

Error Prevention Method	Traditional Flow Cytometer	AQUIOS CL Flow Cytometer	Description
Flow cell clogging prevention	N/A	Built into the design	**Traditional flow cytometer:** Traditional flow cytometers use the prime and flush functions to mitigate flow cell clogs. **AQUIOS CL flow cytometer:** In addition to the prime and flush functions used to mitigate flow cell clogs, four additional features on the AQUIOS CL flow cytometer work to reduce the possibility of flow cell clogging: • Blunt probe reduces coring of septa pierceable cap. • 2 holes in the probe prevent probe clogs. • A filter before the flow cell prevents large particles from reaching the flow cell. • Real-time monitoring of the sample flow rate.
Reducing tandem dye degradation/ evaporation/ condensation	N/A	Built into the design	**Traditional flow cytometer:** Antibody vial caps must typically be removed for use exposing the antibody to light and other environmental contaminates such as dust and dirt. The open vial allows for antibody evaporation and condensation. **AQUIOS CL flow cytometer:** Sealed antibody vials help prevent tandem dye degradation due to light exposure, and evaporation or condensation in the vial. Since these vials are cap-pierced, this also helps to prevent contamination.

Error Prevention Method	Traditional Flow Cytometer	AQUIOS CL Flow Cytometer	Description
Absolute counting	Manual	Automated	**Traditional flow cytometer:** Absolute counts are usually determined using one of two methods: • Single platform: Add fluorospheres to the sample or use daughter tubes coated with beads to determine the absolute count. • Dual platform: Uses counts from a hematology instrument to calculate the counts. **AQUIOS CL flow cytometer:** The AQUIOS CL flow cytometer uses volumetric absolute counting. No absolute count tubes or beads are used for the Tetra application. And, there is no risk of manual pipetting errors.
Specimen Mixing	Semi-Automated	Automated	**Traditional flow cytometer:** On most systems, samples are mixed using a rocker not included on the instrument. This requires the operator to manually transfer samples from the rocker to the sample prep module and from the sample prep module to the analyzer. **AQUIOS CL flow cytometer:** The rocker is incorporated directly in the system and mixes as soon as the cassette is loaded onto the rocker and then again between sample aspirations. Since the mixing module automatically mixes, pierces, and aspirates, the user does not open specimen tubes or extract samples from specimen tubes, thereby reducing operator exposure to blood samples.
Sample Prep	Semi-Automated	Automated	**Traditional flow cytometer:** Sample prep is generally done either manually or on a stand-alone sample prep system, then manually transferred and tracked by the operator to the analyzer. **AQUIOS CL flow cytometer:** The AQUIOS flow cytometer automatically performs the entire sample preparation process without user intervention, then automatically aspirates and analyzes the prepared sample.
Daughter tubes	Required	N/A	**Traditional flow cytometer:** Daughter tubes must be labeled, positioned, transported, and tracked throughout the entire workflow. **AQUIOS CL flow cytometer:** Eliminates the need for the daughter tubes through the use of the microplate. And, since everything is done on one system, there is no need to transfer anything.

Error Prevention Method	Traditional Flow Cytometer	AQUIOS CL Flow Cytometer	Description
Blood aspiration detection	Manual	Automated	**Traditional flow cytometer:** There is no strategy to ensure the amount of blood aspirated, or if any was aspirated at all. Proper blood aspiration can only be inferred through aberrant results and system troubleshooting including visual inspection. If the specimen tube has been previously opened, the user may be required to vent the specimen tubes prior to aspiration to account for pressure and vacuum differences. **AQUIOS CL flow cytometer:** Automatically detects that blood was actually aspirated and ensures the correct volume is aspirated. The system includes an extra buffer in the total blood aspiration to account for the presence of vacuum/ pressure in the specimen tube. Venting is not required.

Flow Cell Clogging Prevention

Traditional flow cytometers use the prime and flush functions to mitigate flow cell clogs.

In addition to the prime and flush functions used to mitigate flow cell clogs, the AQUIOS CL flow cytometer incorporates four built-in features to prevent clogs:

- **A blunt probe.** Using the specimen tube more than once can cause coring of the septa cap which can contaminate the sample and clog the flow cell. To minimize this from happening, the AQUIOS CL flow cytometer uses a blunt sample probe.
- **2 holes in the sample probe.** Two holes in the sample probe reduces the probability of a probe clog.
- **A filter before the flow cell.** The filter works to keep large particles from reaching the flow cell.
- **Real-time monitoring of the sample flow rate.** The system monitors the sample flow rate for potential changes from partial clogs and bubbles and triggers an auto-repeat. Flow rate instability is measured by data rate and channel number. For example, if the channel is drifting, there is likely a clog in the system. The system also looks for insufficient data since an aperture clog could trigger this condition. When an auto-repeat is triggered, the system automatically aborts the run, runs a cleaning cycle, re-aspirates the sample from the 96-Deep Well Plate and re-analyzes the sample.

By combining these features with autocleaning (refer to Table 2), the AQUIOS CL flow cytometer minimizes the need to monitor the system for clogs and is part of what makes it a true Load & Go system.

Reducing Tandem Dye Degradation/ Evaporation/ Condensation

Unlike the emission spectra of most single fluorochromes, tandem dyes are known to display a great deal of variability in "fluorescence intensities within different regions of each emission spectrum." [3] The degradation of tandem dyes is largely due to light exposure that causes "photon-induced oxidation."[3]

Degredation of tandem dyes can occur "within minutes of ambient light exposure." [3] When cell-labeling procedures involve large batch numbers or are conducted in "circumstances where light protective measures are difficult to maintain," a large degree of variability and degradation are introduced. [3] Monoclonal antibodies used on traditional flow cytometers typically use amber or opaque white vials to eliminate light exposure. On these traditional flow cytometers, the vials are typically capped for transport and storage, but must be uncapped for preparation and analysis. It is well-known that sample preparation can introduce variability due to light exposure. [3] Many labs combat this potential for variability by conducting sample preparation activities in a dark laminar flow hood. [3]

The AQUIOS CL system uses similar methods to combat tandem dye degradation, but takes it one step further. Monoclonal antibodies are packaged in amber vials and remain capped throughout transport and storage just like traditional flow cytometers. But, since the AQUIOS monoclonal antibody caps are pierceable, this cap also remains on the vial during preparation and analysis further reducing the risk of light exposure. By keeping the cap on throughout the entire workflow, the AQUIOS CL system also reduces the risk of evaporation, condensation, and contamination. In addition, since sample preparation is done on the system in a closed environment, this further reduces the risk of degradation due to light exposure.

By reducing tandem dye degradation to a point where spectral compensation is minimally affected, the AQUIOS CL system is able to implement "predetermined compensation matrices and analysis templates with preset regions and gating strategies." This simplified approach is ideal for a clinical setting as long as the cell populations meet the system's required separation. [3] With the innovative "bottom-up" gating strategy that the AQUIOS CL system employs, you can rely on good isolation of the desired populations. Refer to the AQUIOS Tetra Application Gating Strategy product bulletin for more information on the gating strategies used on the AQUIOS CL system.

Absolute Counting

Traditional flow cytometers are not volumetric; instead, they rely on fluorospheres for accurate counts. The beads have a specific concentration. These instruments take the concentration plus the known volume added to the sample and calculate the count based on the 1:1 dilution. But, if there is any deviation in the volume of either blood or beads, this can skew the counts.

Volumetric absolute counting is a method used on the AQUIOS CL flow cytometer Tetra Tests that uses calibrated syringes to deliver a specific amount of fluid volume within a specific time. The system is calibrated upon installation by a Beckman Coulter Representative. Once calibrated, the system automatically determines the absolute count.

One of the benefits of this method is that it eliminates the need for daughter tubes and pipetting, both of which have known risks associated with mislabeling, misidentification, and pipetting errors.

Sample Mixing

If using the autoloader, the cassette with the samples is rocked prior to cap-piercing each of the tubes. Simply load the cassettes with the samples in the autoloader. Samples for the Single-tube Loader must be mixed by the user prior to loading the sample.

Sample Prep

When sample preparation is not standardized, it increases the risk of error. Sample misidentification and time are the two most critical variables when dealing with the disconnect in the process between a sample prep system and a flow cytometer. The need to manually transfer samples between the systems leaves room for misidentification errors including mislabeling of daughter tubes, and misplacement of samples. The AQUIOS CL eliminates the risk of misidentification by incorporating the sample prep process directly into the flow cytometry system.

Sample preparation also supports various tasks that rely on consistent time windows. For example, if you batch 40 samples, the timing on a typical flow cytometer is not consistent. On a traditional flow cytometer, since sample preparation is done outside of the system, sample 1 is processed immediately, but sample 40

remains in the queue on the system until all of the other samples are processed. This allows for an inconsistency in the preparation time to processing time. The user is responsible for monitoring this. There is no automatic notification if any step in the process is beyond the time limit.

The AQUIOS CL flow cytometer integrates sample prep directly into the system; samples are aliquotted, stained, incubated and lysed onboard. The AQUIOS CL flow cytometer, therefore, treats every sample the same way. A software scheduler ensures that all samples are prepared and analyzed within the required time. Since sample preparation is done on the system, the instrument is able to schedule preparation times to keep the time variable between sample preparation and sample processing consistent. As the first sample is incubating, the second sample is preparing. Every step has a predefined time window which eliminates time differences that can lead to under and over lysing and inconsistent sample suspension due to settling. Refer to Figure 3 for the sample preparation stations.

Figure 3 – AQUIOS CL Flow Cytometer Sample-Preparation Stations

❶ The automatic **rocking station** which contains the cassette with up to five closed vial specimen tubes.

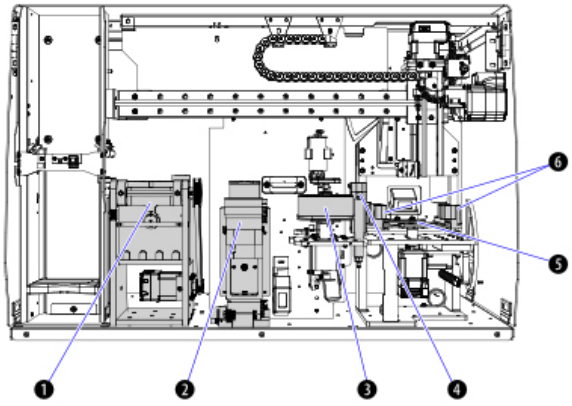
❷ The **single-tube loader station** which holds a single specimen tube that can be open vial or closed vial. The user must thoroughly mix the specimen tube prior to inserting the tube into the station for sampling.

❸ The **reagent carousel** which holds up to ten sealed monoclonal antibody reagent vials.

❹ The **sample-prep probe wash station** which cleans the sample-prep probe after each operation.

❺ The **plate holder** which holds a 96-Deep Well Plate.

❻ Four lysing **reagent holders**, for Lyse A and Lyse B.



Daughter Tubes

The AQUIOS CL system continually tracks where samples are in the process eliminating the need to manually track daughter tubes. Specifically, the system provides traceability between the specimen ID and the position of the prepared sample reducing the possibility for sample misidentification.

Blood Aspiration

Blood Aspiration Detection

On the AQUIOS CL flow cytometer, there is a sensor on the instrument that uses spectrophotometry to determine the difference between blood, air, and sheath fluid based on color. The system uses this sensor as a means to determine if blood is actually aspirating. The system displays a short draw flag to notify the user if a sufficient amount of blood was not aspirated.

Specimen Tube Vacuum/Pressure

The AQUIOS CL flow cytometer borrows the concept of an aspiration buffer from Hematology as a means of negating the impact of vacuum and pressure on aspiration. On other flow cytometry instruments, users would account for the impact of varying vacuum and pressure by venting the tubes before aspiration if the specimen tube was previously opened. This is no longer necessary on the AQUIOS CL system.

ERROR PREVENTION DURING QUALITY CONTROL ON THE AQUIOS CL SYSTEM

The AQUIOS CL system employs several features that work to prevent errors during quality control. These prevention methods can be broken down into the following:

Table 4 - Error Prevention Quality Control at a Glance

Error Prevention Method	Traditional Flow Cytometer	AQUIOS CL Flow Cytometer	Description
Automatically Pause when QC Fails	N/A	Automated	**Traditional flow cytometer:** Auto pause is not applicable on traditional flow cytometers since these instruments are not designed as Load and Go instruments. User presence is needed to verify that QC passed before patient samples can be loaded onto the system. **AQUIOS CL flow cytometer:** Since the AQUIOS CL flow cytometer is a true Load and Go instrument, the system has built in auto pause features that the user can enable to automatically proceed with patient samples once QC has passed. If the feature is enabled and QC fails, the instrument will not process patient samples and the user can be automatically alerted via e-mail or text.

Error Prevention Method	Traditional Flow Cytometer	AQUIOS CL Flow Cytometer	Description
QC checks	Multiple Reagents Required	Two Controls Required	**Traditional flow cytometer:** Most flow cytometry systems use a variety of material to ensure the system is functioning properly. These materials include: various fluorospheres, each of which measure different aspects of QC, and blood-based controls. **AQUIOS CL flow cytometer:** The AQUIOS CL flow cytometer automatically checks, not just the assay values, but several other internal criteria to automatically determine if the system passes QC. This is achieved by running only two controls daily.

Automatically Pause when QC Fails

Refer to *Laboratory SOP: Pause for QC in the Daily Quality Control on the AQUIOS CL System for the Tetra Application* product bulletin for more about the Pause for QC function.

QC Checks

Unlike traditional flow cytometers the AQUIOS CL flow cytometer does not require additional reagents such as fluorospheres to perform QC checks when running the Tetra application. The system's advanced algorithm checks a variety of internal criteria and will simply display pass or fail. The AQUIOS relies on quantitative data to determine the pass/fail parameters. There are four categories of QC checks on the AQUIOS CL flow cytometer:

- **Daily QC (Controls)**

  Daily QC is performed to ensure optical, fluidics and electronic stability within the system are operating within the instrument specifications. When enabled, the system provides a reminder to run controls. Refer to the *Daily Quality Control on the AQUIOS CL System for the Tetra Application* Product Bulletin for a detailed look at the QC process on the AQUIOS CL flow cytometer.

- **Real Time QC (Patient Sample)**

  Real time QC for the Patient Sample automatically monitors several variables during a sample run including: separation quotient (SQ), mean channel, stable data rates, and compensation checks. Since there is no range established for a given sample, the system will not flag it. However, the system will display a generic notification if there are any inconsistencies.

- **Application-Specific Quality Checks (Patient Sample)**

  AQUIOS Tetra Application. The AQUIOS Tetra application consists of three tests: the Tetra 1 test (CD45-FITC/CD4-RD1/CD8-ECD/CD3-PC5), the Tetra-2+ test (CD45-FITC/(CD56+CD16+) RD1/CD19-ECD/CD3-PC5), and the Tetra Combo test (AQUIOS Tetra 1 test followed by AQUIOS Tetra 2+ test).

  The AQUIOS Tetra 1 Panel reagent also functions as an analytic reliability check for a specimen by monitoring the Total CD3+ absolute count. The CD3+ Reliability Check is the sum of the percentages of CD3+CD4+ and CD3+CD8+ cells and should be within ±5% of the total percentage of CD3+. [5]

  The combination of AQUIOS Tetra 2+ Panel monoclonal antibody reagents can function as a quality control check for a specimen in terms of total lymphocyte percentage determined using the following formula: [5]

$$\text{Total lymphocyte percentage (\%)} = \begin{array}{l}\text{\% CD3+ (T) lymphocytes + \% CD19+ (B) lymphocytes +} \\ \text{\% CD3-/CD56+ and/or CD16+ (NK) lymphocytes}\end{array}$$

  The combination of monoclonal antibody reagents in the Tetra Combo test can function as a quality control check for a specimen in terms of total lymphocyte percentage determined using the same formula described above.

  CD3+ Intrapanel Check in the Tetra Combo test is the variability between AQUIOS Tetra 1 and AQUIOS Tetra 2+ for CD3+ and serves as an internal control. Differences between replicate CD3 percent positive results should be ±3.5%.

- **System Checks**

  The system automatically verifies the laser power and that the vacuum is acceptable before each sample is run. The system also verifies internal communications and that the hardware is functioning correctly at each step of the process. In addition, the system performs a fluidic check at the start of each run to ensure there is enough AQUIOS Sheath Solution available for the requested run.

  With the AQUIOS CL flow cytometry system, QC checks associated with instrument performance essentially follow the same principles that are behind the fluorosphere-based QC checks performed by general purpose flow cytometers. The key difference is that the AQUIOS CL flow cytometer is designed to perform these QC checks automatically without the need for additional reagents. This helps to simplify the process making QC checks associated with the instrument quick and efficient.

CONCLUSIONS

Laboratory standards and regulations set forth by governmental bodies such as the Centers for Disease Control (CDC) help to define the procedures that laboratories follow. These procedures are used in an effort to standardize the workflow and are used as safety countermeasures and help to prevent errors. Error prevention, however, is costly and time consuming.

The AQUIOS CL flow cytometer utilizes several error prevention features that are cost effective and in many cases automatically helping to improve safety and reduce the cost and time that often goes hand-in-hand with error prevention methods.

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ACRONYMS

CDC:	Centers for Disease Control
FDA:	Food and Drug Administration
ID:	identification
QC:	quality control
SQ:	separation quotient

TRADEMARKS

CLASS 1 LASER PRODUCT