ABRAXIS® ACE Microcystins Kit with MCT-ADDA ELISA on CAAS Cube

1. Intended Use

For the use of the CAAS Cube instrument for performing automated downstream ABRAXIS® Microcystins ADDA ELISA (PN 520011) analysis on standards and samples extracted with the ABRAXIS® Affinity Capture and Extraction (ACE) – Microcystins Kit (PN 520100) and either the ABRAXIS® Uri-Standards Set – Microcystins (PN 520101) or ABRAXIS® Seri-Standards Set – Microcystins (PN 520102).

2. Safety Instructions

Urine and blood serum samples may contain biohazards. Use appropriate protective equipment (including but not limited to gloves, lab coats, and safety glasses) when working with extracted samples. Discard extracted samples according to local, state, and federal regulations.

3. Limitations, Possible Interferences

See the User Guides for the ABRAXIS® ACE Microcystins Kit and Uri- and Seri- Standard Sets - Microcystins for information on required dilutions and matrix interferences. Additional sample diluent is included with Uri- and Seri-Standard Sets to allow for dilution of samples with interferences or which test higher than Standard 5 (0.4 ppb).

4. Warnings and Precautions

Users working with the Gold Standard Diagnostics CAAS Cube instrument must be fully trained on its operation by GSD Technical Support. The instrument must be maintained and operated as instructed. The instrument must be primed and readied as instructed prior to testing.

5. Sample Collection and Handling

ABRAXIS® Uri- and Seri-Standard Microcystin standard curves, and urine and blood serum samples, extracted with the ABRAXIS® ACE Microcystins Kit must be tested within 24 hours of extraction. Store samples at 2-8°C if necessary and bring to room temperature before testing.

6. Materials Required

6.1 ACE Standards 0, 1, 2, 3, 4, 5, and Control from the ABRAXIS® Uri-Standards Set – ACE Microcystins (PN 520101) OR the ABRAXIS® Seri-Standards Set – ACE Microcystins (PN 520102) extracted using the ABRAXIS® Affinity Capture and Extraction (ACE) – Microcystins Kit (PN 520100)

6.2 Urine or blood serum samples extracted alongside the corresponding Standards Set using the ABRAXIS® Affinity Capture and Extraction (ACE) – Microcystins Kit (PN 520100)

6.3 ABRAXIS® Microcystins/Nodularins (ADDA) ELISA Kit (PN 520011)

6.4 Gold Standard Diagnostics CAAS Cube System (PN 475006)

6.5 Pipette and tips capable of 50 µL volume

7. Procedure

7.1 Bring reagents, standards, and samples for the assay to room temperature

a. Remove the Antibody, Conjugate, Color, Stop, 5X Wash, and Laboratory Reagent Blank (LRB) reagents as well as the Microtiter Plate from the ADDA ELISA kit and let them warm up to room temperature for ~45-60 minutes (depending on the temperature of the room, this could differ). DO NOT USE the standards and control in the ADDA kit, these will be replaced by the standards and control extracted with the ACE Kit!

b. Ensure the extracted Uri- or Seri- standard curve, control, and urine or blood serum samples being run have been properly extracted according to ACE Kit procedure and brought to room temperature prior to analysis. Ensure extracted samples have been stored at 2-8°C for fewer than 24 hours.

c. Dilute the ABRAXIS® Wash Buffer (5X) Concentrate (from the ABRAXIS® Microcystins/Nodularins (ADDA) ELISA Kit (PN 520011)) at a ratio of 1:5 with deionized or distilled water. If using the entire bottle (100 mL), add to 400 mL of deionized or distilled water and mix thoroughly. Place Cube probe into diluted 1X Wash Buffer bottle.

7.2 Setting up the Cube to run an assay

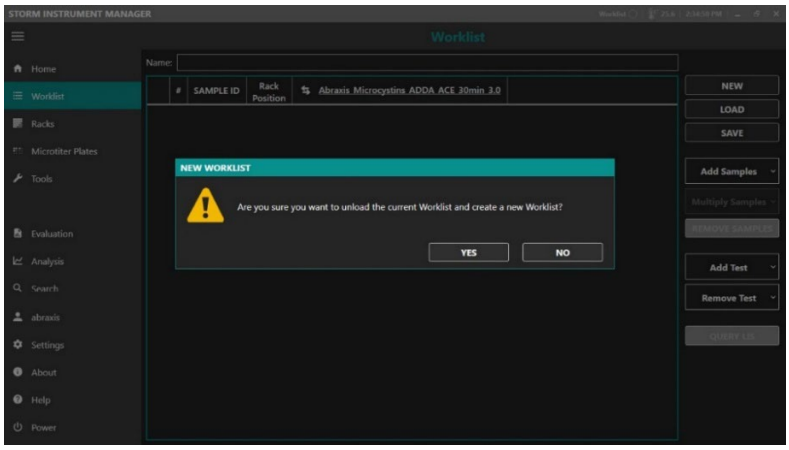
a. Open the Storm Instrument Manager software

b. Ensure all daily, weekly, and monthly maintenance is properly completed prior to running the test – Prompt will generate if needed

c. Ensure DI water and 1X Wash Buffer are full and properly prepared, and waste container is empty

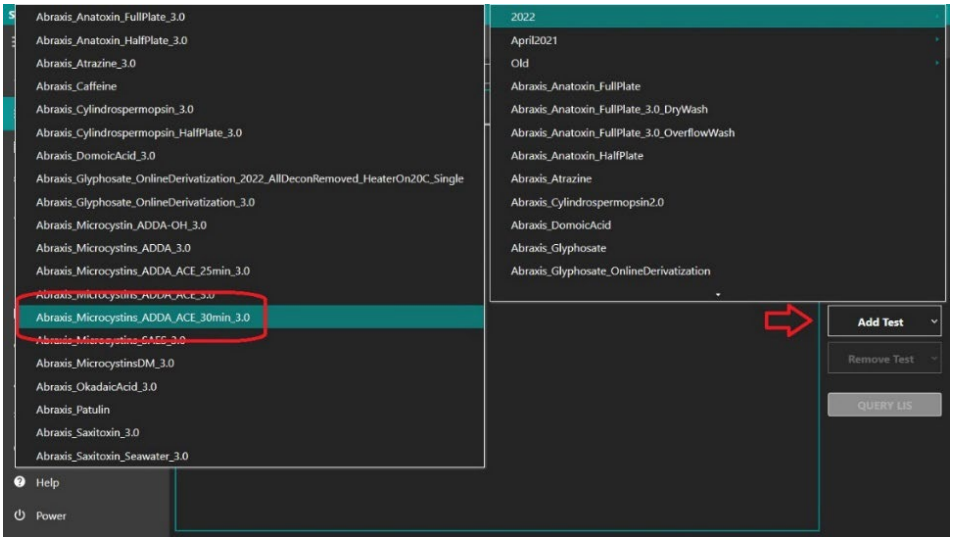
d. Navigate to the worklist tab

e. If a worklist is already loaded, select “New” and then “Yes”

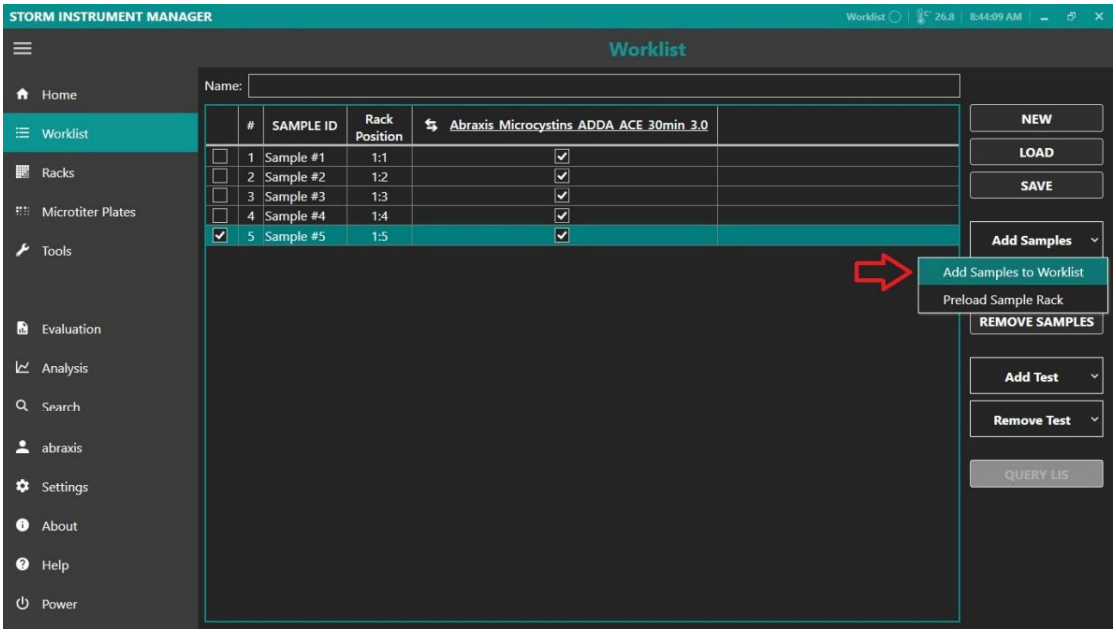


f. Enter the name of the run (typically the name of the assay being run and the date) in the Name bar

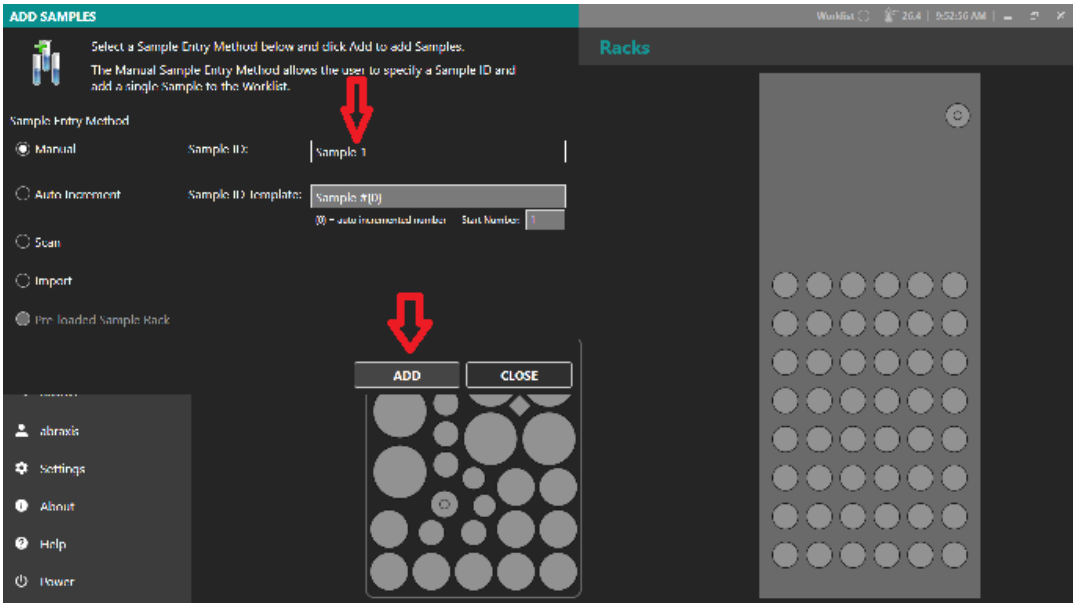
g. Add the test to be run by clicking on “Add Test” and selecting “Method” from the dropdown. Select Abraxis_Microcystins_ADDA_ACE_ 30min_3.0



h. Click on the “Add Samples” and then the “Add Samples to Worklist” buttons



i. Select sample entry method (manual, auto-increment, scan or import)
j. Enter sample IDs, click “Add”

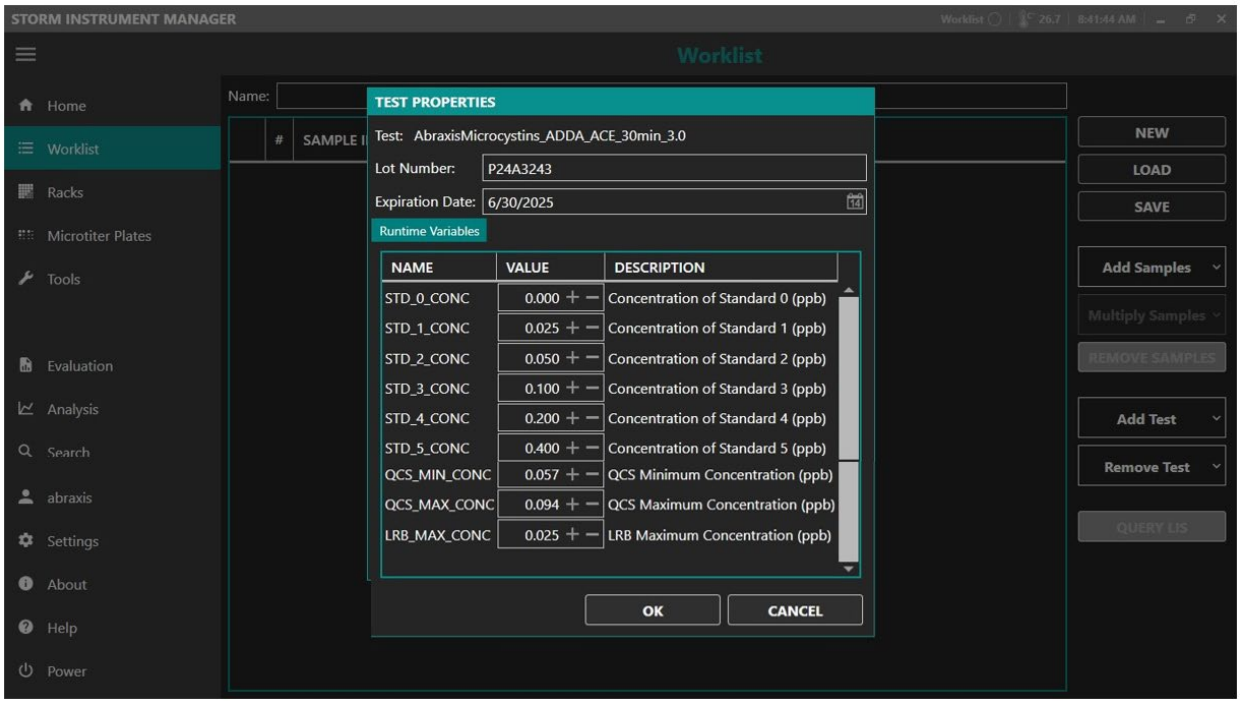


k. Select “Done”. DO NOT physically add sample vial to rack – samples will be pre-loaded directly onto the microtiter plate in step 7.3.a
l. Repeat this step until all sample names are added, and then select “Close”

m. Now on the worklist tab, check the box to the right of each sample ID to be run. Ensure that the final image looks as it does in figure 7.2.h with all samples selected

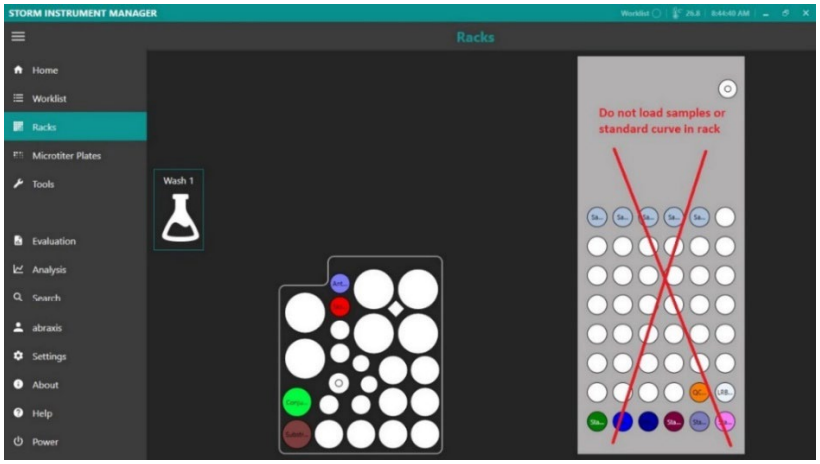
n. Click on the test name (Abraxis_Microcystins_ADDA_ACE_ 30min_3.0) and update with the ADDA kit lot number and expiration date. Change the concentration of the samples to match the following, then select "OK" to save the worklist:

STD_0_CONC	0.000
STD_1_CONC	0.025
STD_2_CONC	0.050
STD_3_CONC	0.100
STD_4_CONC	0.200
STD_5_CONC	0.400
QCS_ MIN_CONC	0.056
QCS_ MAX_CONC	0.094
LRB_MAX_CONC	0.025



NOTE: Standard concentrations will only need to be input the first time the assay is run. Concentrations will be saved in the software for succeeding runs when using the same test method. However, it is always a good idea to double-check that standard concentrations match the above.

o. Navigate to the "Racks" tab and ensure the 1X Wash Buffer is connected and that the Antibody, Conjugate, Color, and Stop solutions ONLY are placed in the correct places in the rack with the lids removed. DO NOT physically load standard curve or samples in rack even though the software displays this on the screen



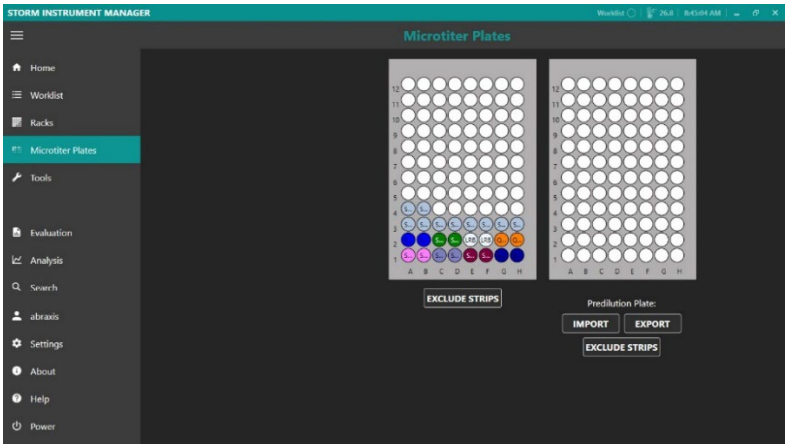
7.3 Load the ELISA plate and begin the automated assay:

a. With the room temperature extracted standard curve, LRB, control, samples, and microtiter plate from step 7.1, pipette 50 µL of each into duplicate wells on the microtiter 8-strips according to the following scheme. Seal remaining non-used microtiter plate 8-strips back in the plate bag for later use

	1	2	3	4	5	6	7	8	9	10	11	12
A	Std 0	Std 4	Samp 1									
B	Std 0	Std 4	Samp 1									
C	Std 1	Std 5	Samp 2									
D	Std 1	Std 5	Samp 2									
E	Std 2	LRB	etc.									
F	Std 2	LRB	etc.									
G	Std 3	Control										
H	Std 3	Control										

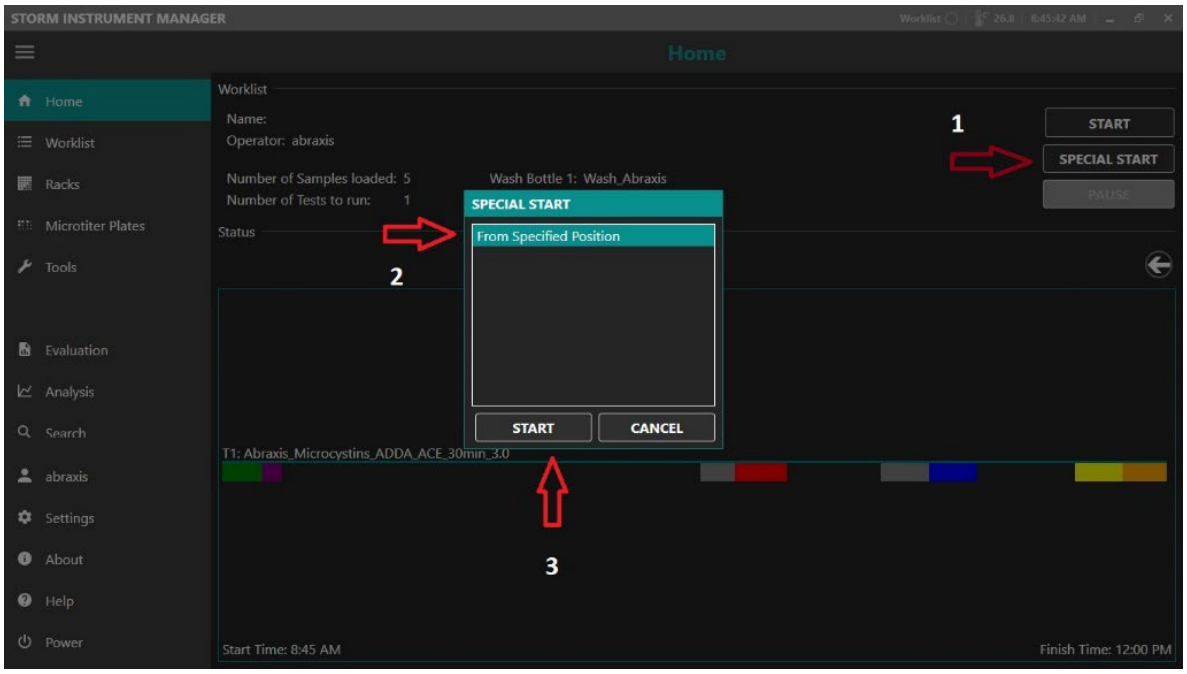
b. Navigate to the “Microtiter Plates” tab

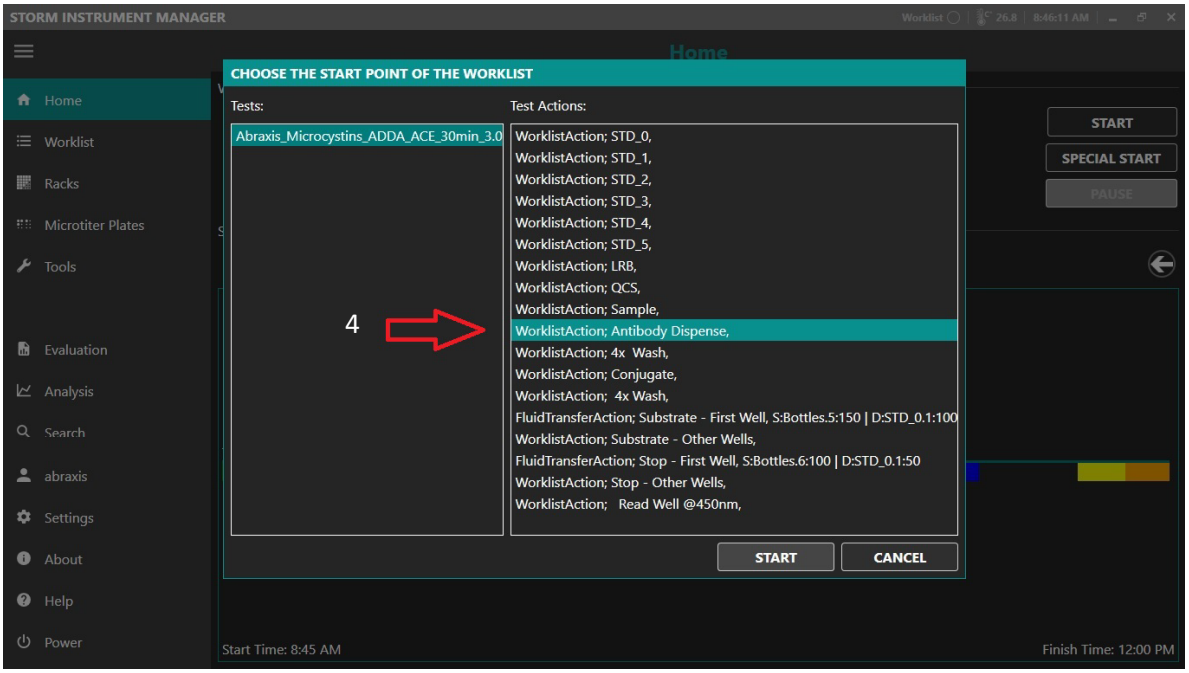
c. Place the loaded ELISA plate into the holder securely according to the picture below. Make sure the ELISA plate has the proper amount of strips and is loaded in the correct plate holder. Be careful when loading to not spill or splash the contents!



d. Return to the home screen and select "Special Start" (1). Select "From Specified Position" (2) then select "Start" (3) and then "WorklistAction; Antibody Dispense" (4). Hit "Start" and "Continue" to begin the run. This will ensure that the program begins with the addition of antibody to the pre-loaded standard curve and samples

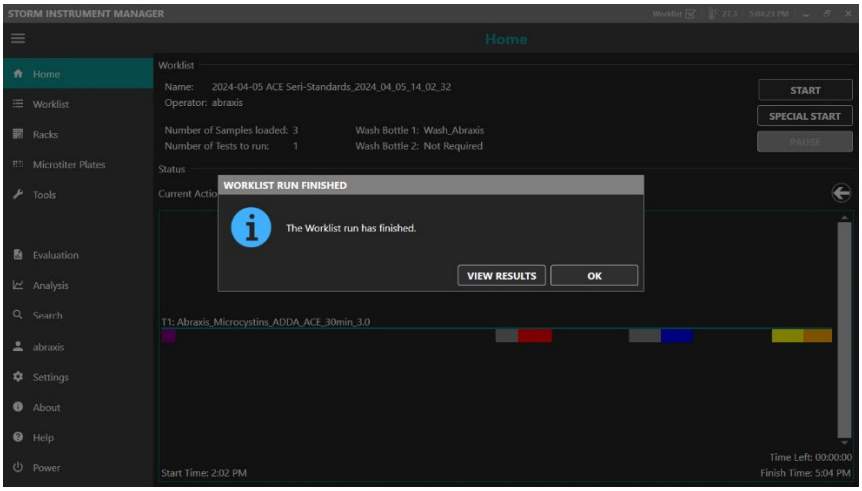
NOTE: The CTRL button may be held down while executing the above steps in order to allow the Cube door to be open during the run, if desired





7.4 Evaluation of results:

a. Once the run is complete, click “View Results” or navigate to the Evaluation tab to see the worklist report



b. Remove and recap all reagents and store properly

c. Ensure end-of-day cleaning is followed according to prompts and instructions

8. For ordering or technical assistance contact:

Gold Standard Diagnostics
795 Horsham Rd
Horsham, PA 19044
WEB: www.abraxiskits.com

Phone: (215) 357 3911
Fax: (215) 357 5232
Ordering: info.abraxis@us.goldstandarddiagnostics.com
Technical Support: support.abraxis@us.goldstandarddiagnostics.com

Date this Technical Bulletin is effective: 09APR2024 Version: 01