Life Technologies
Attune® NxT Software version 1.1.0 Release Notes

Customer ("you") must review these notes prior to installing or operating the Attune® NxT Software version 1.1.0 ("Attune Software v1.1.0").

System Requirements
Recommended: Quad core processor, 16 GB RAM, 500 GB disk space available. Windows 7 64 bit Professional with Service Pack 1.

Attune Software v1.1 Installation
NOTE: You must have Administrator privileges to install this software

1. It is recommended that no other applications are running while completing these steps.
2. Unzip AttuneNxT.zip and run "setup.exe". Follow the instructions and accept all defaults.
3. After install is complete, open a Windows Explorer window and navigate to the Attune NxT install folder, default <C:\Program Files\LifeTechnologies\AttuneNxT\>
4. From this folder, install the drivers and database as described below.

1. DESKey Driver installation
   - 1.1. Navigate to the Attune installation directory, DESKey driver folder. The default is <C:\Program Files\LifeTechnologies\AttuneNxT\DESkey Driver>
   - 1.2. Run <dk2wn3264_7.34.0.57.exe> and follow the instructions to install the DESKey drivers.
2. Attune® NxT Driver installation
   - 2.1. Navigate to the Attune installation directory, USBDriver folder. The default is <C:\Program Files\LifeTechnologies\AttuneNxT\UsbDriver>
   - 2.2. Run < AttuneNxTDriverInstaller_x64.exe> and follow the instructions to install the Attune® NxT Cytometer USB driver.
3. Database Engine Installation
   - 3.1. Navigate to the Attune installation directory. The default is <C:\ProgramFiles\LifeTechnologies\AttuneNxT\dbinstall>
   - 3.2. Right click "database_engine_install.cmd", then select "Run as Administrator".
   - 3.3. This will take some time to run, hit the enter key when finished.
4. Attune® NxT Datastore Installation (Ensure that step 3 is completed before performing step 4)
   - 4.1. Navigate to the Attune database installation directory. The default is <C:\ProgramFiles\LifeTechnologies\AttuneNxT\dbinstall>
   - 4.2. Run <AttuneNxtDatabaseSetup.exe> and follow the instructions to setup and initialize the Attune NxT Datastore
5. Migrate Existing MySQL data
   - 5.1. Navigate to the Attune database installation directory. The default is <C:\ProgramFiles\LifeTechnologies\AttuneNxT\dbinstall\MySQL Import>
   - 5.2. Run <Import_MySQL_to_Postgres.cmd> to import existing MySQL data.

Features of Software

Instrument Controls

Automated Performance Testing - Baseline and PT (Report)
Automated Maintenance - Start up, Shutdown, Rinse, Deep Clean, Sanitize
Data Collection from tubes

Visualization Tools

Dot Plot, Density Plot, Histogram
Histogram marker, quad marker, Precedence Density Plots
Customized logic gates
Data Display-Log, Linear
HyperLog transform
Statistics
Filmstrip Zoom Tool
Plot Previews

Experiment Set Up

Auto-compensation from tubes
Auto layout and Freeform Workspace Layouts

File Management

Import/ Export of FCS Files and experiments
Files Stored as FCS3.1 format

User Management & Security

3 accounts – single service, admin and user account

Known Issues:

Instrument Start Up/Performance Test

- Power on the instrument prior to launching the software
- Wait at least 1 minute after powering instrument on before launching software to allow ample time for system self-test to complete.
- If refilling fluids prior to performing start up, please make sure that the bottles are completely removed from the instrument prior to filling the bottles. When the bottles are placed back on the instrument, please ensure that all sensor cables are plugged in tightly.
- If sensor cables are not plugged in correctly, start up function may not be activated and lights for the fluid bay will flash. In this case, check all sensor cables, power cycle the instrument, and restart the software.
- For performance test, ensure that you are using the right concentration of beads (3 drops/2mL) otherwise the algorithm may time out.
- Run performance test directly after starting application. If you open an experiment and then run performance test you may experience a failure. Restart application and repeat performance test.

Artifact #	Description	Suggested Action
artf54561	Status bar does not update performance test status	Check performance test report to determine performance test status daily

Setting up an experiment

- Use rectangle or polygon gates whenever possible
- Do not use quadrant gates as a parent gate.

Artifact #	Description	Suggested Action
artf52800	When changing plot axis parameters using the plot context menu some parameters are	When using the plot context menu make sure to check the labels

	not in the same order	before selecting the parameter as yellow laser labels go from largest to smallest, while all other laser parameters go from smallest to largest
artf53358	When scaling a polygon using the bounding rectangle, the polygon collapses to a line. The vertices do not maintain their relative positions.	Avoid scaling polygon gates.
artf53507	Hyperlink to set populations not available on the top of the plot	On customize menu, deselect experiment and or sample name to visualize the hyperlink. If names have too many characters, hyperlink will display out of the plot area that is visible
artf53424	Changing scatter histogram plots to dual parameter plots using customize menu doesn't set axis to default linear scale	If you change plot types using the customize menu, ensure that x and y axis scale ranges are set appropriately
artf54546	Plots cannot be added when gate is selected	Deselect the gate prior to adding a plot

Experiment Explorer

artf52612	Duplicating Experiments experiment names cannot be immediately changed	Double click on the newly created experiment to activate experiment, right click on experiment to rename
artf53466	Invalid characters cannot be used in experiment explorer	Limit experiment, group, and sample names and comment fields to either letters or numbers, avoid use of special characters

Compensation

- Compensation files must be created from scratch.
- If you make an error in setting up your compensation i.e. wrong controls or auto-fluorescence mode, you must create a new experiment and start over.
- Compensation control run protocols will default to a set volume, flow rate, and number of events even if you input specific values into the run protocol.
- Compensation gates need to be manually set for each individual control, apply gate to all controls is not available at this time.
- For experiments that have compensation applied, occasionally you might see FSC and SSC voltage boxes flashing, which will not allow you to adjust the voltages by typing a value into the box. You can adjust voltages by using small increments on the slider bars.

Artifact #	Description	Suggested Action
artf53408	The compensation matrix dialog's reset button only reverts changes made to the dialog and not to the original matrix state as determined by comp controls	Prior to making adjustments on the compensation matrix, make note of the calculated values so that you can reset them manually if necessary.
artf54564	In compensation set up changing parameters prior to selecting background mode and Area/Height doesn't persist	Choose Area/Height parameter and auto-fluorescence mode prior to selecting parameters for

		controls to ensure persistence
	Scatter population disappears while adjusting histogram gates during compensation sample recording	Allow compensation recording to complete prior to adjusting histogram gate. If population disappears, open another compensation tube by double clicking on experiment explorer and then re-open control where error occurred
artf54796	Compensation control is not highlighted in voltage adjustment panel	When adjusting voltage sliders when collecting compensation controls, ensure that you are adjusting the correct channels per control selected on the experiment explorer
artf54815	Not all parameters are available to adjust on compensation controls	When adjusting the axis labels for compensation parameters some parameters may be missing. If parameters are missing, create new experiment.
artf55007	Adjustments in values on compensation matrix occasionally will not update correctly	If compensation doesn't seem to adjusting when typing values directly into the matrix, adjust the marker on all of the controls to force the compensation matrix to refresh.
artf55053	Voltages can not be adjusted from a deleted compensation controls that has data previously recorded	If fluorescence voltages need to be adjusted on any compensation control that has data recorded, delete compensation and restart voltage optimization and then record.

Acquiring Samples

- Keep the event rate under 9,999 events/sec and minimize the number of parameters to only those that you need (for example, only collect data in height or area not both and remove width). Keep total event file size below 1 million total events. Ensure that the USB 3.0 cable is plugged into the SS USB port prior to acquiring a file.
- On occasion data may not be displayed on the workspace when run or record is pressed. Often stopping the system and restarting solves the issue. If it is persistent power cycle the instrument and software.
- Turn off the automatic back gating.

Artifact #	Description	Suggested Action
artf53353 artf53369	Run protocol is not persisted if you adjust the flow rate or if you apply to experiment	Adjust the sample flow rate prior to entering the acquisition volume and the stop options. If adjustment is needed, remember to go back and check and or change acquisition volume/stop options or it will revert to default settings
artf54239	Number of events displayed may not be the	

	number selected in stop criteria	
artf55949	Changing plots during acquisition on large event files can cause a large lag or system crash	If collecting large event files >100,000 events, do not adjust any of the plot parameters during acquisition. After acquisition, parameters can be adjusted, however depending on the total file size there may be some delay.

Workspace (Gates, Plots, Stats)

- Use less than 31 gates if you are inserting experiment statistics
- Do not use quad gates as parent gates.
- It is recommended not to copy and paste gates
- Use less than 50 characters when naming gates.
- Copy and paste of plots outside of application is not available at this time.

Artifact #	Description	Suggested Action
artf50038	Alignment of plots, aligns to the object to the furthest side of the page (Left aligns to the most left object, right to the most right, top to the most top)	Manually align plots or use auto layout mode to automatically align plots in grid fashion
artf50040	Plot resizing (Make same height" and "Make same width) will vary based on insertion method	To resize use the plot resizing tools to resize to the plot that was inserted last
artf52417	Plots blur at low resolutions	Keep plot resolution greater than 256x256 for best visualization
artf53509	Gate labels can be a different from the gate if selected from the gate customization menu	Ensure that the gate you are looking at matches the label do not depend upon color of label alone
artf54881	Quad gate names truncated after 31 characters	Modify quad gate names to be less than 31 characters
artf54867	Sample context menu is not working to open sample	
artf54918	Pasting gates between plots doesn't work using short cut or context menu	Do not copy gates from plots; add plots individually using the workspace ribbon.
artf55024	Hyperlink may disappear when workspace is shrunk using the slider bar size adjustment scale	Increase size of workspace using the size adjustment slider bar
artf55025	Gate color is limited to 9 colors	After 9 gate colors all additional gates are colored black
artf55053	Unable to delete compensation controls	You cannot delete a compensation control, you will need to create a new experiment and then recreate compensation. You can delete the recorded file and re-record.
artf55170	In results tab, concentration can show 0.00 even when the concentration is not 0.00.	Concentration stat not available at this time.
artf55203	Revising the name of a group will not be displayed in stats	Open another sample within the group to update the new group name.
artf55224	Derived gates can only be created using	If creating derived gates, be

	regions	aware that they can only be created using regions.
artf55595	Customize panel does not update properly when film strip view is open	Use navigation arrows to force customize panel to update its contents

Data Analysis / Data Display

- On the results view, right clicking on a column header will display a list of statistics some of which are most not currently available.
- When viewing files within experiments, sometimes there can be a parameter mismatch error or file error, click off the sample you are trying to activated and then reselect the sample and it should display correctly without errors.

Artifact #	Description	Suggested Action
artf52579	Plot Statistics on workspace does not show relevant gates if plot for stat box is a daughter plot	Statistics for plots with a region used to set the population will only display all events.
artf54100	System locale settings not persisted in statistics tables, stats displayed on plots and results tables	

FCS files

Artifact #	Description	Suggested Action
artf52544	Time stamp on FCS file is not accurate- it does not show the system locale time it is based on a universal clock	

Shutdown/Instrument Maintenance /Instrument error states

- It is recommended to keep the computer on when performing shutdown that way if there are any errors detected during the process they will be captured on the screen.
- System Test button in Instrument Tab missing
- Decontamination: At all steps that require removing bottles, the tube lifter should be lowered before the bottles are removed and raised only after the bottles are re-installed and the step is acknowledged by selecting OK. If the bottles are disconnected while the tube lifter is up, there is a high probability that the decontamination function will not complete. If this happens, cycle power on the instrument and restart the decontamination function.

Artifact #	Description	Suggested Action
artf54606	After fluid error state, instrument will begin running if sensor cable is plugged in before bottle cable	Plug bottle cable in first prior to plugging in sensor cable
artf54707	Instrument doesn't wake up after instrument shutdown	Once shutdown is complete power down instrument or power cycle instrument prior to restarting software and running startup
artf55261	Cancelling Lower Lid Error state dialog will not resolve the error state	If cancelling error "close lid" restart software