CE

# EVE™ HT A26

## User Manual




NanoEntek

**EVE™ HT A26 User Manual**

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The information in this user manual is described as accurately as possible.

Firmware and software changes may occur without prior notification.



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# Introduction

EVE™ HT A26, developed by NanoEntek, is a fully automated cell counting system designed to integrate the entire sample preparation and cell counting workflow into a single platform. By combining hands-free pipetting, aliquoting, diluting, staining, and counting into one seamless process, the system allows users to save time, minimizes user dependent variability, and delivers highly reproducible results. EVE™ HT A26 is intended for use in research laboratories and bioprocessing facilities requiring high-throughput cell analysis.

The system is capable of processing up to 96 samples in just 10 minutes using only 20 µL of sample volume per channel. Once consumables and samples are loaded onto corresponding positions in the deck, EVE™ HT A26 completes the rest of steps to complete cell counting workflow — aliquoting, diluting, mixing, loading, and counting — without requiring further user input.

For detection, EVE™ HT A26 supports both bright field based and fluorescence based cell counting capabilities and works with Trypan Blue, Erythrosin B, and AO/DAPI reagents. The fluorescence-based imaging option distinguishes nucleated cells from debris, ensuring accurate viability measurements with samples having tissue debris or red blood cells. Furthermore, the auto-darkening front door blocks external light interference during counting, ensuring accurate and reliable fluorescence-based measurements. These features ensure consistent, user-independent results across varied operating conditions.

# Product Components

EVE™ HT A26 consists of the following components.

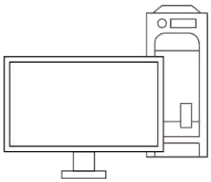
If any of the components are missing or damaged, please contact your local sales representative or send an email to **sales@nanoentek.com**.

**EVE™ HT A26**
**Instrument**

1 EA

**EVE™ HT A26**
**desktop PC**

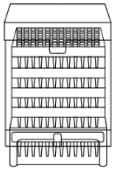
1 EA



**EVE™ HT A26**
**counting kit**

1 EA

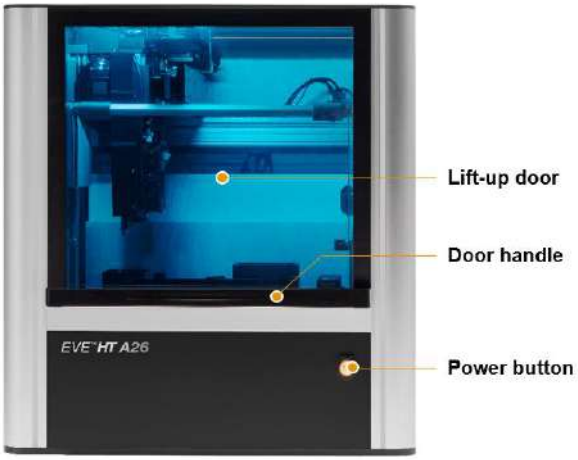
**EVE™ HT A26**
**Disposable Pipette Tips**



**EVE™ HT A26**
**21 CFR part 11 software**
**(optional)**

# Product Description

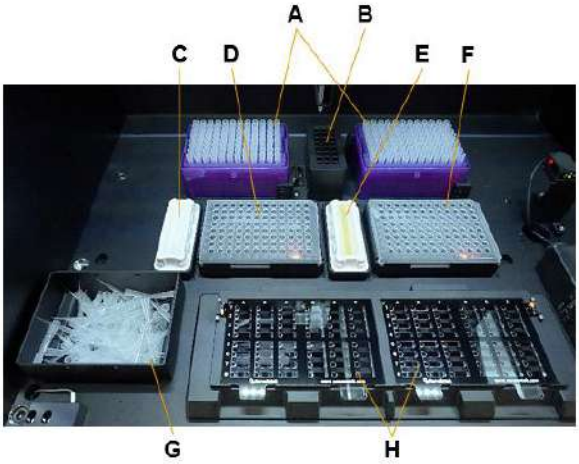
## Front view




| Part name | Description |
|---|---|
| Lift-up door | Door that locks during operation and automatically becomes dark during fluorescence imaging. |
| Door handle | Handle for opening the lift-up door. Pull the handle outward, then lift to open the door. |
| Power button | Button to turn the instrument on or off |

# Product Description

## Interior view




| | Part name | Description |
|---|---|---|
| **A** | Pipette tip box holder | Holds pipette tip boxes; supports up to 2 boxes |
| **B** | Pipette tip settler | Helps secure tight sealing of pipette tips onto the pipette head |
| **C** | Reservoir D slot | Holds a reservoir for diluent |
| **D** | Sample plate holder | Holds the sample plate in place |
| **E** | Reservoir R slot | Holds a reservoir for reagent |
| **F** | Mixing plate holder | Holds an empty 96 well plate for mixing |
| **G** | Tip waste bin | Holds a bin to collect used pipette tips |
| **H** | Counting plate holders | Hold up to two EVE™ HT A26 counting plates |

# Installation

## Environmental Requirements

For best performance, please review the following recommendations before setting up EVE™ HT A26:

- Use at room temperature.
- Do not expose instrument to direct sunlight.
- Do not expose instrument to continuous vibration.
- Do not expose instrument to intense magnetic or electromagnetic fields.
- Do not install instrument in high-humidity environment.
- Do not install instrument near corrosive gases or substances.
- Minimize contact with dust or airborne particles.
- Make sure to have at least 10 cm (4 inches) of free space around instrument for proper airflow.

⚠ ***WARNING***

*Installation must be performed only by NanoEntek certified engineers. Unauthorized installation voids the product warranty and may result in personal injury or damage to equipment.*

# Sample & Consumables preparation

## Recommended Actions

To obtain best results, follow these recommendations:

**1. Handling sample**

① Wear personal protective equipment while handling samples.

② Use 96-well plates in EVE™ HT A26 kit to prepare sample plate. Other 96-well plates are not compatible.

③ Start measurements within 30 minutes after preparing sample plate to avoid aggregation and evaporation of samples.

**2. EVE™ HT A26 Operation**

① Turn on the equipment first, then launch the software. The software will check the equipment status when it launches.

② Before turning on EVE™ HT A26, make sure that nothing inside obstructs pipette head, and the lift-up door is closed.

③ The lift-up door locks automatically once EVE™ HT A26 starts its operation and can't be opened while it is moving. To stop operation, press pause or cancel button in the EVE™ HT A26 software.

④ Do not touch the pipette head inside the equipment. This may cause misalignment or malfunction.

**3. Consumables Check and Refill**

① Before starting measurements, make sure to replenish consumables including mixing plate, EVE™ HT A26 counting plates and pipette tips. Once consumables are replenished, make sure to update consumable quantities in the software to match the actual amount.

② Before starting measurements, make sure to add sufficient amount of EVE™ HT reagent in the reservoir 'R'. If dilution function is activated, make sure to add sufficient amount of diluting media in the reservoir 'D'. Avoid spilling inside when refilling consumables. Pay special attention to the area below the counting plate holder, as it has holes for imaging through which consumables can fall.

③ When installing EVE™ HT A26 counting plates, hold the handles in the plate and avoid touching any other part of the counting plates.

# Sample & Consumables preparation

## Preparing samples

Follow the steps below to prepare samples in a 96-well plate provided in the EVE™ HT A26 kit. EVE™ HT A26 will handle all subsequent steps to provide cell counting results.

1. Prepare samples in growth medium or PBS. Make sure that cells are well dispersed. If needed, vortex samples before adding samples into sample well plate.

☛ ***NOTE***

*Recommended cell concentrations for EVE™ HT A26 are as below:*

*Detectable range: $1 \times 10^4$–$2 \times 10^7$ cells/mL*

*Optimal range: $1 \times 10^5$–$1 \times 10^7$ cells/mL*

2. Add well-mixed samples into a sample well plate. Refer to the table below regarding the volume of samples to be added. Make sure to record the arrangements of samples within the well plate so that the arrangement can be updated in EVE™ HT A26 software.

☛ ***NOTE***

*Sample well plates (similar to standard PCR plates) are included in EVE™ HT A26 kit. Sample well plates and mixing well plates are same.*

**Required sample volume by mode**

| Mode | Sample volume |
|---|---|
| **Direct mode** | Exactly **20 µL** per well |
| **Full mode**<br>(Without aliquot and dilution) | At least **30 µL** per well (Max. 100 µL) |
| **Use Aliquot** | **30 µL × (number of aliquots)**<br>(Max. 6 times / up to 180 µL) |
| **Use Dilution** | **at least 50 µL** per well<br>(Exception: load **60 µL** if Dilution Factor is 2) |

3. Aliquot and dilution functions are done within sample well plate. Therefore, it is important to make sure that there are sufficient empty wells in sample well plate for dilutions or aliquots.

☛ ***NOTE***

*Aliquot or dilution is performed from left to right. For detailed settings, refer to page 23.*

## Preparing samples

4. Place the prepared sample well plate onto the holder inside EVE™ HT A26.

☛ ***NOTE***

*It is important to start cell counting within 30 minutes after preparing sample well plate. Waiting more than 30 minutes may affect counting results.*

☛ ***NOTE***

*Refer to the image below for the location of the sample well plate holder. Ensure the sample well plate is not placed in the mixing well plate holder.*

# Sample & Consumables preparation

## Preparing consumables

Consumables for EVE™ HT A26 must be loaded into the instrument. Check the remaining quantity of each consumable in the EVE™ HT A26 software, then add or replace them as needed.

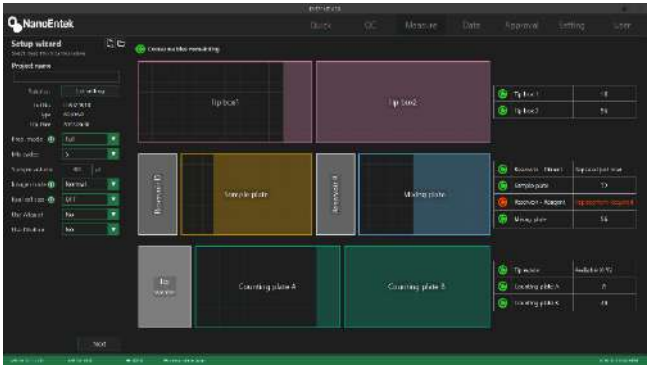
☛ ***NOTE***

*The number of consumables required varies depending on experimental design. The exact quantities can be confirmed in Step 3 of the Setup Wizard.*

⚠ ***CAUTION***

*Consumables cannot be replaced while the instrument is in operation. Ensure that all necessary consumables are prepared before starting a run.*

To check the current consumable status, click the Measure tab in the software.

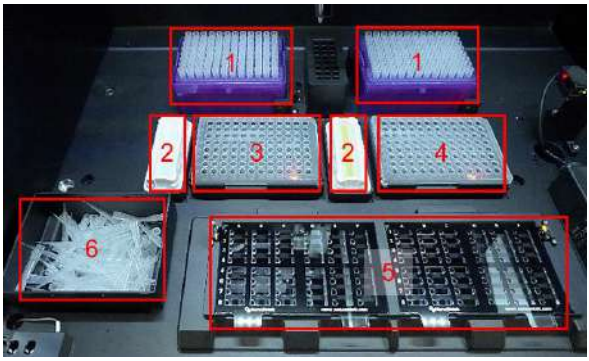



EVE™ HT A26 automatically tracks the number of consumables used during each measurement, and displays remaining quantity of each consumable so that they can be replenished as needed.

After replenishing consumables, you must click the refresh button to apply the change. Alternatively, you can right-click on each consumable to apply a refill or edit the quantity manually.

# Sample & Consumables preparation

## Preparing consumables




⚠ ***CAUTION***

*Always use consumables validated for use with EVE™ HT A26. Use of non-validated or incompatible products may result in malfunction or damage to the instrument.*

**1. Tip boxes**

Pipette tips for EVE™ HT A26 are provided in a pack of 5 trays which includes one empty tip box. Pipette tips for EVE™ HT A26 can be purchased through registered vendors. To install a new tray, remove empty tray from tip box, then place new pre-loaded tray of pipette tips into empty box.

**2. Reservoirs**

EVE™ HT A26 has two positions for reservoirs.

**Reagent reservoir (Reservoir R)**

Reservoir between the sample plate position and the mixing plate position is for reagent. First put an empty reservoir in the position for 'Reservoir R', and fill with sufficient amount of reagent. You may use a pipette or pour directly from reagent bottle.

**Diluent reservoir (Reservoir D)**

When using the dilution function, place an empty reservoir in the position for 'Reservoir D', and fill it with sufficient amount of diluent.

⚠ ***CAUTION***

*Avoid overfilling the reservoir or spillage onto surrounding areas.*

**3. Sample well plate**

Prepare a sample well plate as described in 'Preparing Samples' (page 11), then place it in the sample plate position in the correct orientation, with the well 'A1' positioned at the top-left corner.

# Sample & Consumables preparation

## Preparing consumables

**4. Mixing well plate**

Place an empty well plate provided in EVE™ HT A26 kit in the mixing plate position. EVE™ HT A26 will automatically dispense both samples and reagent into the mixing well plate.

If there are remaining empty wells in a mixing well plate, it can be reused. Make sure to mark empty well positions in EVE™ HT A26 software.

☛ ***NOTE***

*In Direct Mode, reagents will be added directly to sample plate, therefore, mixing well plate is not required.*

**5. Counting plates**

Use only the counting plate made exclusively for EVE™ HT A26. There are two positions for counting plates, and each counting plate is designed to count up to 48 samples. Therefore, 96 samples prepared in a 96-well plate can be counted simultaneously.

When handling counting plates, use only the handle and avoid touching other areas. After placing counting plates in the counting plate positions, make sure that they sit properly on the counting plate holder.

⚠ ***CAUTION***

*Do not load the counting plate upside down. The side with the logo must face upward.*

**6. Tip waste**

Waste bin collects used pipette tips. Before each run, make sure to empty bin.

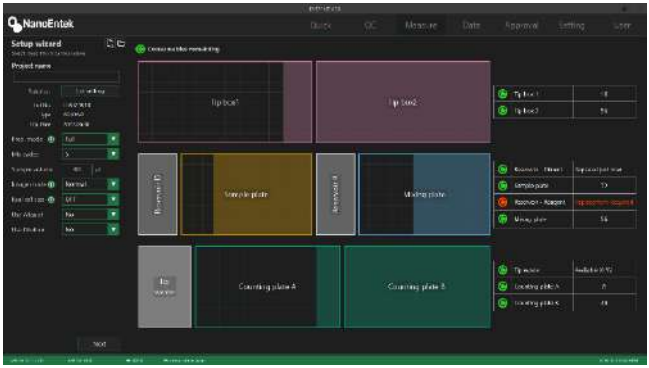
☛ ***NOTE***

*Used consumables, including pipette tips, must be disposed of in accordance with applicable local regulations.*

# Measure

## Setup wizard – Step 1

The **Setup Wizard** available in the **Measure** tab provides a helpful tool for users to set up a cell counting project.
In general, the Setup Wizard menu splits in two panels. The left panel lists available parameters with options, and the right panel demonstrates the arrangement of consumables.



In **Step 1**, users can define modes and options of a cell counting project, and check the remaining quantity of consumables.

**Step 1 - Left panel**

# Measure

## Setup wizard – Step 1

A. **Project Name**: Enter a unique name for a cell counting project.

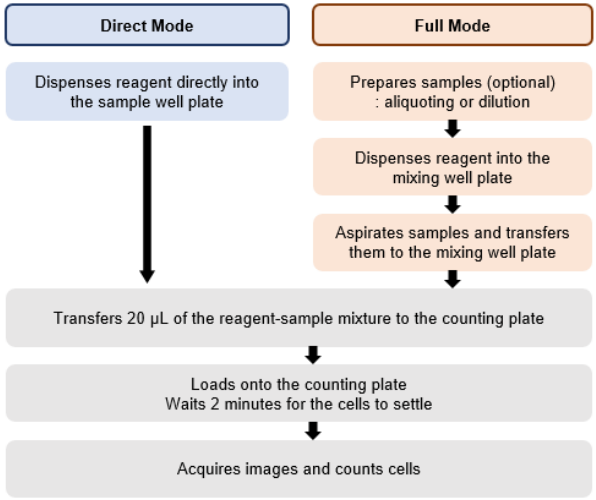
B. **Solution**: Choose one of three available reagents (AO/DAPI, Trypan Blue, and Erythrosin B). Enter the lot number of reagent as shown in the label. The expiration date will be automatically shown based on the lot information. This reagent information will be saved for future projects. Users can modify information by choosing 'Lot Setting' button.

C. **Prep. Mode**: Choose one of the available sample preparation modes (Direct, Full, or None mode).

- Direct: EVE™ HT A26 will expect that each well of sample well plate contains exactly 20µL of samples, and add reagent directly into sample well to perform cell staining.
- Full: EVE™ HT A26 will transfer samples from sample well plate to mixing well plate, and add reagent into the mixing well plate. If aliquoting or dilution steps have been selected, EVE™ HT A26 will perform aliquoting or dilution before performing cell staining. If users want aliquoting or dilution steps, the Full Mode has to be selected.

☞ ***NOTE***

*Direct Mode vs. Full Mode Operation*



## Setup wizard – Step 1

• None: If you already have loaded counting plate(s), you can use None mode to skip the preparation process and proceed directly to imaging. Follow steps 1 through 3 as usual, but no consumables are required. In Step 2, only the samples at the included positions will be imaged and counted.

D. **Mix Cycles**: Choose how many times samples will be mixed before aspirating samples. Available mixing cycles are 3, 5, 10, 15, 20, and 30 cycles.

E. **Sample Volume**: Enter the volume of samples in sample well plate. For Direct Mode, the sample volume is fixed at 20 µL and cannot be manually entered by the user. For Full Mode, it should be at least 30µL and no more than 100µL.

F. **Image Mode**: Available only when AO/DAPI solution is selected; select one of the three imaging modes.

- Fast: Captures 1 image per well
- Normal: Captures 4 images per well (default)
- FFPE: Specialized imaging mode for FFPE(formalin-fixed, paraffin-embedded) samples; uses AO/DAPI reagents but measures only DAPI, and uses the DAPI count as the total cell count.

G. **Real Cell Size**: Available only when AO/DAPI solution is selected. Choose 'On' to take brightfield images along with fluorescence images. Bright field images will not be used for counting cells and provided only for cell size estimation.

H. **Use Aliquot**: Choose 'Yes' to enable the option for aliquoting function which can be set up in the Wizard Step 2-1.

I. **Use Dilution**: Choose 'Yes' to enable the option for dilution function which can be set up in the Wizard Step 2-1.

☛ ***NOTE***

*Aliquot and Dilution modes cannot be enabled simultaneously.*

J. **Save** ( ): Saves the protocol created through this Setup Wizard.

K. **Load** ( ): Loads a previously saved protocol.

L. **Next**: Proceed to the next step.

## Setup wizard – Step 1

**Step 1 - Right panel**


The **right panel** of the Wizard Step 1 displays the currently remaining quantities of consumables in EVE™ HT A26. It is important to verify that the quantities shown in the Wizard match the actual quantities in the instrument. Users can edit quantities in the software manually to match the actual quantities. If a consumable has been replenished, click the Refresh button to reset the count.

A. **Refresh all button**: Resets the remaining stock count for all consumables at once.

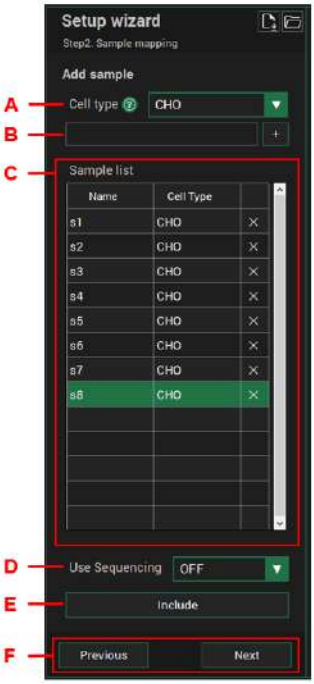
B. **Consumable panel**: Displays a visual representation of available consumables in EVE™ HT A26, along with numbers in the table on the right (box C). Right-click on each consumable panel to access the 'edit window'.

C. **Remaining stock**: Shows exact quantities of available consumables. Click the Refresh button to reset counts.

## Setup wizard – Step 2

In **Step 2**, users can add details about samples and their locations within sample well plate. This information will be saved together with cell counting results and will be used to calculate average and variations of replicates.

### Step 2 - Left panel



## Setup wizard – Step 2

A. **Cell type**: Select one of pre-saved cell types from the dropdown menu.

☛ ***NOTE***

*To register a new cell type, first run a quick measurement in the Quick tab and save it as a new preset. For detailed instructions, refer to the Quick count chapter on page 36.*

B. **Sample Name**: Enter a unique name for a sample. Click the '+' button to add it to the **Sample List**.

C. **Sample List**: Displays sample names and their sample type. To remove a sample, click the 'x' button. How to assign sample to sample well plate will be explained in Step 2 – Right Panel.

D. **Use Sequencing**: If sequential numbering is needed, select 'On' from the dropdown menu for 'Use Sequencing'. When multiple wells are selected and 'Include' is clicked, sample names will be automatically appended with sequential numbers (-01, -02, -03, etc.) on the plate map.

E. **Include**: Assigns the selected sample to the chosen wells on the plate map.

F. **Previous / Next**: Navigate to previous or next step.

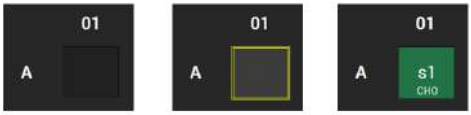
**Step 2 - Right panel**


The **right panel of Step 2** shows a map of sample well plate in a 96-well format. Configure the arrangement of samples to match actual sample layout within the sample well plate. To configure sample well plate map, follow the steps on the next page.

## Setup wizard – Step 2

**[How to Define the Map of Sample Well Plate]**
Repeat the following steps for all samples.
1. Select a sample from the Sample List.
2. Select the target wells in the Plate Map.
3. Click the 'Include' button.
• Tip1: To group samples, right-click on the selected wells and choose 'Include as a Group'.
• Tip2: If aliquoting or dilution function is included, make sure to leave required columns empty.

When a well is assigned to the map of sample well plate, the well will be shown as solid green with sample name (example shown below).




One can use the right button of mouse to include a well or exclude an already included well.


One can add multiple wells together as a group using **'Include as Group'** option. After cell counting is done, the results of wells included as a group will be averaged together. One can also assign wells as 'Group' later in the Data tab after all measurement is completed. Samples assigned to a same group will be displayed in a different color (example shown below).

## Setup wizard – Step 2-1 & 2-2

☛ ***NOTE***

*If you are not using the Aliquot or Dilution functions, skip this step and proceed directly to Step 3.*

EVE™ HT A26 supports options to aliquot or dilute samples before performing cell counting. To use this optional feature, one has to leave enough empty columns in sample well plate because aliquoting and dilution steps will take place within the sample plate.

In **Step 2-1**, you can configure how aliquoting or dilution will be performed. In **Step 2-2**, you can preview the final map of sample well plate after aliquoting or dilution as determined in the Step 2-1.

☛ ***NOTE***

*Aliquot or Dilution functions will be performed in column(s). One can choose entire column to do aliquoting or dilution. Individual well(s) cannot be chosen.*

**Aliquot function**


If **'Use Aliquot' option in Step 1** has been selected, the Setup Wizard will show the optional Step 2-1 (example shown above).

Follow the steps below to configure how aliquoting will be performed.

1. Select a source column (containing samples) from the **'Source'** dropdown menu.
2. Select a target column from the **'Aliquot to'** dropdown menu. Samples in the source column will be aliquoted to the target column.
3. Click the '**Apply**' button.
4. If more than one aliquot is needed, repeat 1–3.

(Continued on the next page)

## Setup wizard – Step 2-1 & 2-2

5. Verify that the sample well plate map on the right reflects the intended sample arrangement.
The aliquot source and target columns are displayed in the same color, indicating that they originate from the same sample.

When configuring the aliquot settings, please note the following.

- Target column(s) do not need to be adjacent to the source column.
- When a well in a column has been assigned to have sample in Step 2, the column containing the well cannot be used as a target column.

After completing the aliquot settings, click the '**Next**' button to proceed to **Step 2-2** screen as shown above.

**Left panel**: Displays information about the aliquot execution.
**Right panel**: Shows the expected arrangement of sample well plate after aliquoting.

The colors assigned in Step 2-1 are displayed as borders around each well, allowing you to identify which column shares the same sample.

☛ ***NOTE***

*Ensure that the number of aliquots matches the prepared sample volume. If the volume is insufficient, accurate counting results cannot be obtained.*
*The required sample volume of the source is calculated as follows:*
*Sample Volume of Source = 30 µL × (Number of Aliquots)*

## Setup wizard – Step 2-1 & 2-2

### Dilution function


If ‘Use Dilution’ option in Step 1 has been selected, the Setup Wizard will show the optional Step 2-1 (example shown above).

Follow the steps below to configure how dilution will be performed.
1. Select a source column (containing samples) from the **‘Source’** dropdown menu.
2. Select a target column from the **'Dilute to'** dropdown menu. Samples in the source column will be transferred to and diluted in the target column.
3. Enter ‘**Dilution Factor**’.
4. Click the ‘**Apply**’ button.
5. If more than one dilution is needed, repeat 1–4.
6. Verify that the sample well plate map on the right reflects the intended sample arrangement.

The dilution source and target columns will be displayed in similar color with progressively lighter shades, reflecting decreasing concentration at each dilution step.

When configuring the dilution settings, please note the following.

- Dilution is performed in column unit.
- Unlike Aliquot, Dilution can only be performed to the **directly adjacent column to the right**. Please note this when preparing the sample well plate.
- When a well in a column has been assigned to have sample in Step 2, the column containing the well cannot be used as a target column.

## Setup wizard – Step 2-1 & 2-2

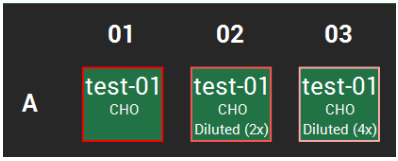
Please note the following regarding sample and diluent preparation.

- The required volumes of sample and diluent are displayed in the dilution setup screen at Step 2-1. If the volumes are insufficient or incorrectly prepared, accurate counting results cannot be obtained.
- Prepare a sufficient amount of diluent in Reservoir D based on the volumes shown on the screen. Depending on the sample type, appropriate diluents such as PBS or culture media may be used.
- The instrument will dispense diluent into each well according to the set dilution factor and proceed with serial dilution automatically.

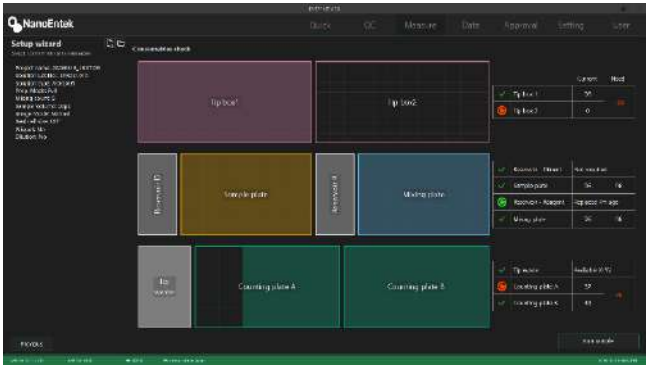

After completing the dilution settings, click the '**Next**' button to proceed to **Step 2-2** screen as shown above.

**Left panel**: Displays information about the dilution execution.
**Right panel**: Shows the expected arrangement of sample well plate after dilution.

The colors assigned in Step 2-1 are displayed as borders around each well, allowing you to identify which column shares the same sample. In Dilution mode, the individual dilution factor (e.g., 2x, 4x) is displayed for each sample on the plate map.

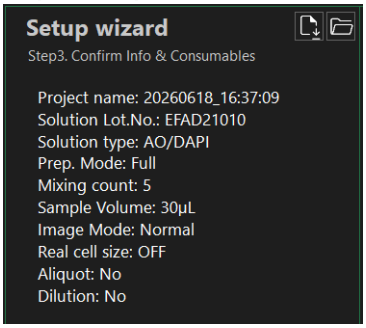


## Setup wizard – Step 3



In **Step 3**, the details of the cell counting project are summarized for a final review. The current remaining quantities of each consumable are displayed alongside the required quantities to run the cell counting project.

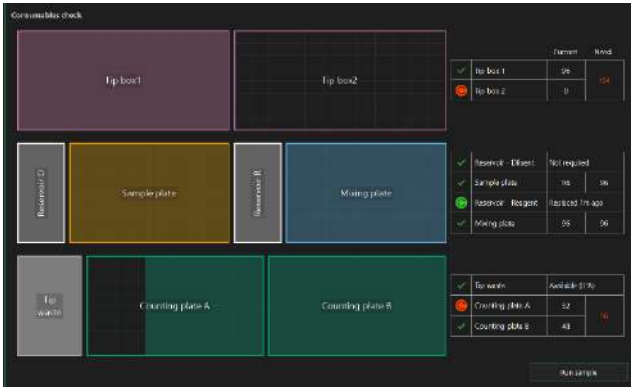
**Step 3 - Left panel**




The settings configured in Steps 1 and 2 are displayed. To modify any settings, press the **Previous** button to return to the previous step.

## Setup wizard – Step 3

**Step 3 - Right panel**




The right panel displays the consumable status, which is similar to Step 1; however, it reflects the experimental design configured in the cell counting project, and displays the **required quantities of each consumable**. Any consumables with insufficient quantity will be highlighted in red.

## Measurement View


Once **'Run Sample'** button is clicked, EVE™ HT A26 will start to run steps as defined in the counting project. Progress will be shown in 2 different stages in 2 tabs: Preparation and Imaging.

**Preparation tab**

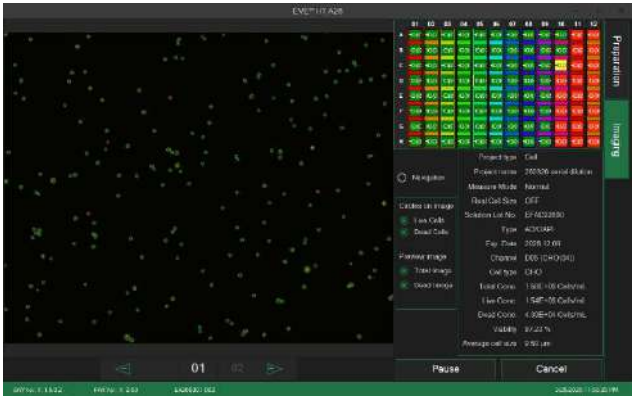
In the Preparation tab, the sample plate map on the left will show which column is being engaged in preparation, and the lists on the right will show the details of each liquid handling step. The current step will be highlighted with light gray.

The status of each well on the sample well plate is indicated by the following colors:

| | |
|---|---|
| **Green** | Loading complete |
| **Yellow** | Sample preparation and loading in progress |
| **Red** | Awaiting preparation |

## Measurement View

**Imaging tab**




Once preparation is complete and the sample is loaded onto the counting plate, the instrument waits for 2 minutes for the sample to settle before imaging begins. Imaging proceeds from top to bottom and left to right.

The status of each well in the sample well plate is indicated by the following colors:

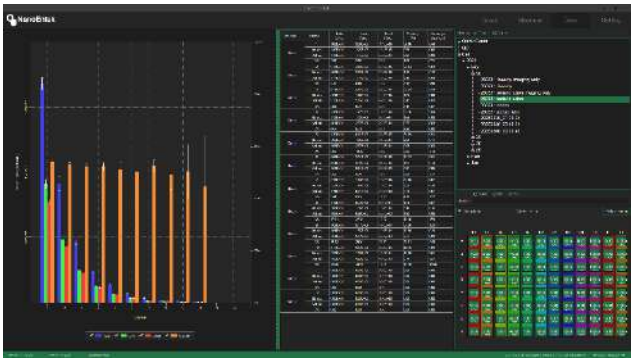
| | |
|---|---|
| **Green** | Imaging and counting completed |
| **Yellow** | Imaging and counting analysis in progress |
| **Red** | Awaiting imaging |

During imaging processes, one can review the images and counting results of other wells which have been imaged. When one of ‘Green’ wells is selected, the captured image of the selected well will appear, and the counting results of the well will appear on the right. When a 'Yellow' well is selected, the well image will be displayed; however, the counting result will appear as '0' and only the well information will be shown. When a 'Red' well is selected, neither the well image nor the counting result will be displayed.

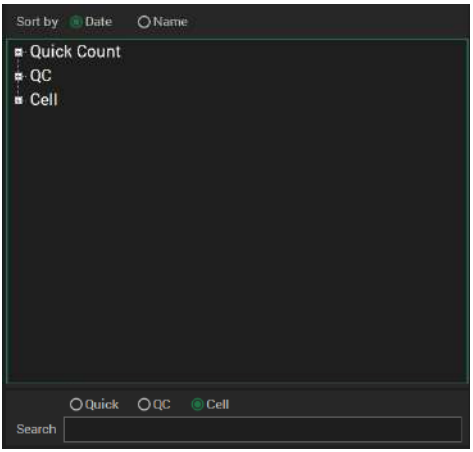
# Data

## Data list & Sample plate map

Clicking the **Data** tab allows you to view the counting results of projects for which imaging and analysis have been completed. You can also edit and save the results from this menu.



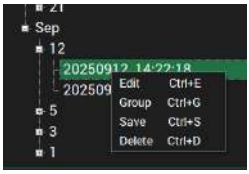
The project list on the right is displayed as shown below.




| | |
|---|---|
| **View type** | It can be viewed by two criteria, date or name. |
| **Quick Count** | Data list taken from the Quick menu. |
| **QC** | Data list taken from the QC mode (Project type). |
| **Cell** | Data list taken from the Cell mode (Project type). |
| **Search** | Search the data in each section. |

## Data list & Sample plate map

Right-click on each project to access sub-menu.




| | |
|---|---|
| **Edit** | Edit the project name.<br>Set the Size gating and dilution factor.<br>Acceptance range is only available in QC data. |
| **Group** | Assign and edit group settings. |
| **Save** | To save project, select **Save**.<br>Select the data type and data path in the pop-up window. |
| **Delete** | To delete project, select **Delete**. |

☞ ***NOTE***

*When the 21 CFR Part 11 option is activated, projects cannot be deleted if they have not been backed up or are in the approval process.*

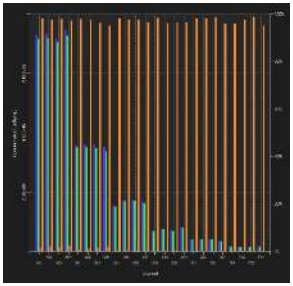
The plate map at the bottom right provides the following functions:

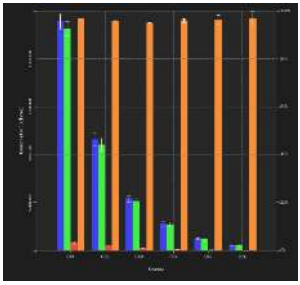

| | |
|---|---|
| **Sample list** | Expands a list showing the name and location of each sample. |
| **Heat map** | Displays the total count values of the samples as a heat map, where a darker shade of green indicates a higher count. |
| **Full plate map** | Opens a popup screen showing an enlarged view of the entire plate map. |

# Data

## Graph and Table

When data is measured with Cell as a project type, results will appear in graph and table form.





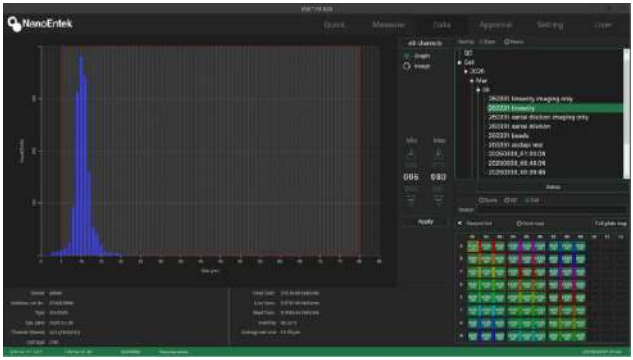
The graph shows total counts (BLUE), live cell counts (GREEN), dead cell counts (RED), and viabilities (ORANGE). You can hide any of these graphs by unchecking boxes below the graph. Grouped data will be displayed as a single graph.

| Channel | Name | Total Conc. | Live Conc. | Dead Conc. | Viability (%) | Average Size (µm) |
|---|---|---|---|---|---|---|
| Group | G01 | 2.99E+05 | 2.91E+05 | 7.68E+04 | 96.79 | 11.36 |
| | Std dev | 7.64E+04 | 7.41E+04 | 8.47E+03 | 0.33 | 0.10 |
| | Std err | 3.82E+04 | 3.70E+04 | 4.24E+03 | 0.16 | 0.05 |
| | CV | 3.19 | 3.20 | 11.02 | 0.34 | 0.90 |
| Group | G02 | 1.16E+06 | 1.11E+06 | 4.90E+04 | 95.75 | 11.37 |
| | Std dev | 6.31E+04 | 6.48E+04 | 6.67E+03 | 0.67 | 0.08 |
| | Std err | 3.15E+04 | 3.25E+04 | 3.33E+03 | 0.33 | 0.04 |
| | CV | 5.45 | 5.85 | 13.60 | 0.70 | 0.67 |
| Group | G03 | 6.60E+05 | 6.14E+05 | 2.61E+04 | 95.17 | 11.30 |
| | Std dev | 3.62E+04 | 3.49E+04 | 1.99E+03 | 0.14 | 0.15 |
| | Std err | 1.82E+04 | 1.74E+04 | 8.46E+02 | 0.07 | 0.08 |
| | CV | 5.74 | 5.79 | 6.49 | 0.15 | 1.33 |
| D05 | D05 | 2.65E+05 | 2.55E+05 | 8.84E+03 | 96.67 | 11.55 |
| D06 | D06 | 2.97E+05 | 2.86E+05 | 1.06E+04 | 96.43 | 11.54 |
| D07 | D07 | 3.04E+05 | 2.92E+05 | 1.24E+04 | 95.93 | 11.42 |
| D08 | D08 | 2.46E+05 | 2.33E+05 | 1.24E+04 | 94.96 | 11.39 |
| E05 | E05 | 1.06E+05 | 1.02E+05 | 5.30E+03 | 95.08 | 11.42 |
| E06 | E06 | 1.24E+05 | 1.20E+05 | 3.53E+03 | 97.14 | 11.87 |
| E07 | E07 | 1.40E+05 | 1.38E+05 | 1.77E+03 | 98.73 | 11.47 |
| E08 | E08 | 1.22E+05 | 1.15E+05 | 7.07E+03 | 94.20 | 11.55 |
| F05 | F05 | 6.01E+04 | 5.83E+04 | 1.77E+03 | 97.06 | 10.94 |
| F06 | F06 | 6.01E+04 | 6.01E+04 | 0.00E+00 | 100.00 | 11.70 |
| F07 | F07 | 4.95E+04 | 4.59E+04 | 3.53E+03 | 92.86 | 11.10 |
| F08 | F08 | 5.83E+04 | 5.65E+04 | 1.77E+03 | 96.97 | 11.46 |

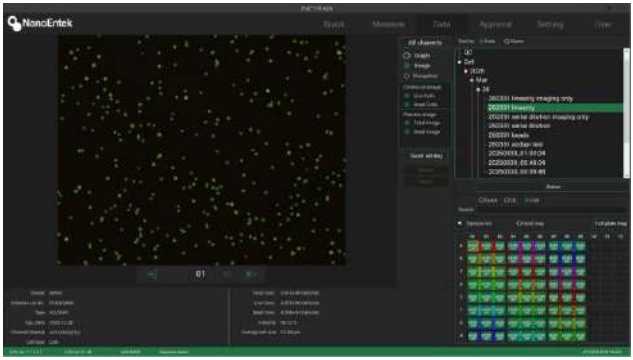
Data for each well (or group) is organized and displayed in a table as shown above. When you click on one of the column names, results will be sorted in ascending or descending order of the selected column (Group data cannot be sorted). For grouped data, the standard deviation, standard error, and CV of each group are automatically calculated and displayed.

## Individual well results

When one well is selected, cell counts (Total, Live and Dead cell concentrations), viability, and average cell size will be shown below cell size histogram. You can change minimum or maximum cell sizes to be included in the results.

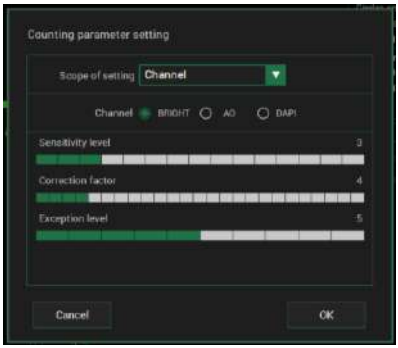


By selecting **'Image'**, you can review raw images. If **'Real cell size'** has been selected when using AO/DAPI solution, brightfield images will also be shown.



## Individual well results

If you click **'Count setting'**, you can change counting parameters in the current data. Refer to page 38 for a detailed explanation of parameters. Parameter values can be applied simultaneously to Channel, Cell type, Group, or Project.

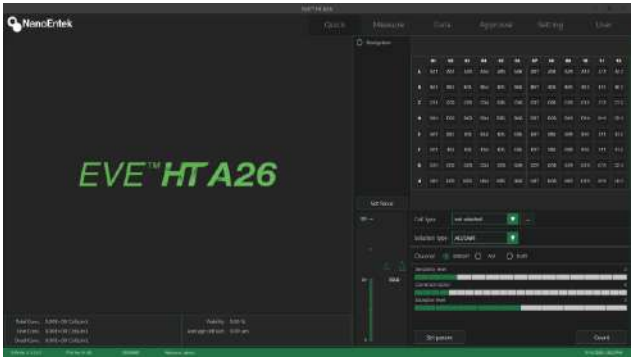



By right-clicking and dragging the image, you can select cells individually as shown below to manually assign those objects in the selected area to either live or dead cells, or debris.

# Quick count

## Quick count

**Quick tab** provides an option to run measurements quickly on a single chamber, without using automated sample preparation. To use ‘Quick count’, samples need to be stained and loaded manually. One can also use this ‘Quick count’ to fine-tune cell counting parameters and focus settings for a new cell type.




1. Stain samples manually by mixing 20 µL of sample with 20 µL of reagent, and transfer 20 µL of the mixture into a cell counting plate. (See figure on the right to find an inlet of a counting chamber).
2. Allow 2 minutes for cells to settle down so that all cells sit on the bottom of the counting chamber.
3. Insert counting plate into the counting plate holder, and close the front door.
4. In EVE™ HT A26 software, click on one of the loaded wells to view the live-feed image.
5. Click the ‘Count’ button to run a Quick count. EVE™ HT A26 will take images and count cells using configured cell counting parameters.
6. To adjust the parameters or focus, click the 'Clear' button and reconfigure the settings. Then click the 'Count' button again to verify the updated results.
7. To save the current focus to a cell type, select the desired cell type and click the 'Set Focus' button.
8. To save the current parameters to a cell type, select the desired cell type and click the 'Set Param' button. The saved cell types and current parameters can be reviewed by clicking the '...' button in the Cell Type menu.

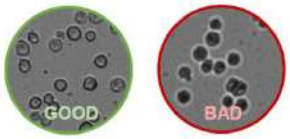
# Quick count

## Quick count

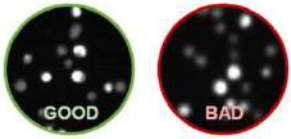

**Fine focusing** is also available while the preview is active. The large arrows adjust the focus in steps of 1, while the small arrows adjust it in steps of 0.1.
Focus adjustment is also possible using the mouse — move the cursor over the preview window and scroll the mouse wheel while holding the Ctrl or Shift key.

☛ ***Note:*** *Focus example*

**AO/DAPI**

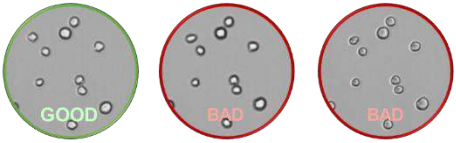



**BRIGHT** : The outline is clear, and the brightness inside the cells is the same as the background.




**AO&DAPI** : clear cell outline

**Trypan blue or Erythrosin B**



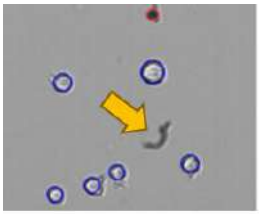

The outline is clear, and the brightness inside of live cells is brighter than the background.

## Counting parameters

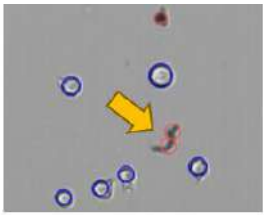
**Counting parameters** are used to deal with wide varieties of cell sizes and shapes. The description of each parameter is as follows.


**1. BRIGHT channel**

**Sensitivity level** : Decreasing this sensitivity parameter will make it easier to prevent debris from being counted. (Caution: If the sensitivity level is too low, then some of the real cells may not be counted.)

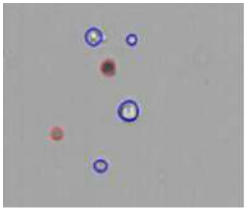


Sensitivity 2

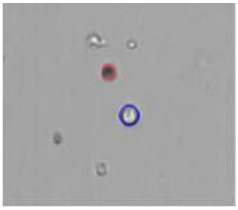


Sensitivity 10

**Correction factor** : Decreasing this correction factor will make it easier to detect cells which are slightly darker than the background. When cells look very transparent, you can decrease this correction factor to pick up vague objects.



Correction factor 4



Correction factor 8

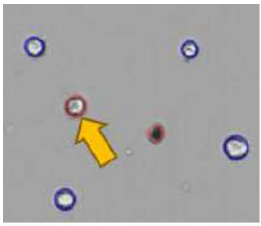
.

# Quick count

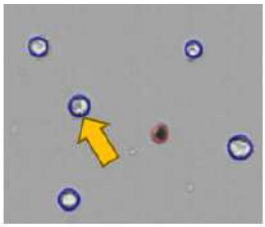
## Counting parameters

**Viability level :** Increasing this viability level will make it easier to detect marginally dark cells as dead cells. Note that increasing this level will increase the dead cell count.

☛***Note:*** *Viability level is available only when cells have been stained with Trypan blue or Erythrosin B.*

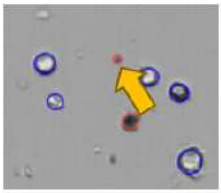


Viability level 2

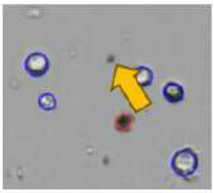


Viability level 5

**Exception level** : Decreasing this exception level will make it easier to detect small objects.



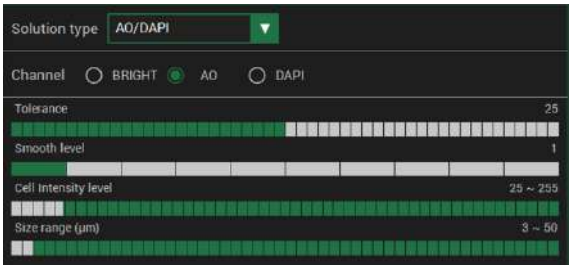
Exception level 5



Exception level 10

# Quick count

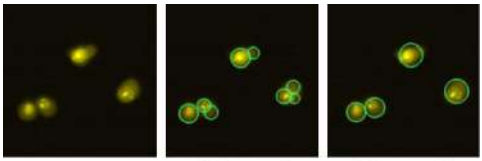
## Counting parameters




**2. AO or DAPI channel**

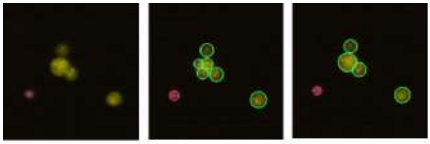
(parameters need to be set for each channel)

**Tolerance**: Tolerance defines how to count dividing cells. Decreasing this parameter will allow counting cells in the middle of cell division or adjacent cells as individual cells. Increasing it will make large cells or aggregated cells count as one cell.



Tolerance 5 Tolerance 25

**Smooth level**: The smooth level controls the smoothness of images. When cells are large and intracellular organelles are distinct enough to be counted as individual cells, you can increase this level to smooth out such distinctive textures.



Smooth level 1 Smooth level 3

**Cell intensity level**: Cell intensity level works similarly to intensity gating. When cells are very dim or too bright, you can change the range of intensity levels of cells to be counted. Here, you can choose both minimum and maximum values.

**Size range**: Size range refers to the range of fluorescent cell sizes. This allows you to determine the size range and exclude small debris from the count.

# Quick count

## Counting parameters

The following table provides examples of recommended parameters for commonly used cell lines. These are recommendations and can be further adjusted by users.

☛ ***Note:*** *Counting PBMCs or small diameter cells with Trypan blue or Erythrosin B is not recommended.*

☛ ***Note:*** *Using AO/DAPI solution is recommended for accurate cell count and viability analysis.*

### AO/DAPI staining solution

| | Cell size | Bright | | |
|---|---|---|---|---|
| | | Sensitivity level | Correction factor | Exception level |
| **PBMC** | 2-80 | 3 | 4 | 5 |
| **CHO** | 5-80 | 3 | 4 | 5 |
| **Jurkat** | 5-80 | 3 | 4 | 5 |
| **U2OS** | 7-80 | 3 | 4 | 5 |
| **HeLa** | 8-80 | 3 | 4 | 5 |
| **HepG2** | 9-80 | 3 | 4 | 5 |

| | AO | | | | DAPI | | | |
|---|---|---|---|---|---|---|---|---|
| | Tolerance | Smooth level | Cell Intensity level | Size range | Tolerance | Smooth level | Cell Intensity level | Size range |
| **PBMC** | 5 | 1 | 25-255 | 2-50 | 5 | 1 | 25-255 | 2-50 |
| **CHO** | 25 | 3 | 25-255 | 3-50 | 25 | 3 | 25-255 | 3-50 |
| **Jurkat** | 5 | 1 | 25-255 | 3-50 | 5 | 1 | 25-255 | 3-50 |
| **U2OS** | 5 | 1 | 25-255 | 3-50 | 5 | 1 | 25-255 | 3-50 |
| **HeLa** | 25 | 1 | 25-255 | 3-50 | 25 | 1 | 25-255 | 3-50 |
| **HepG2** | 25 | 3 | 25-255 | 3-50 | 25 | 3 | 25-255 | 3-50 |

### Trypan blue stain

| | Cell size | Sensitivity level | Correction factor | Viability level | Exception level |
|---|---|---|---|---|---|
| **CHO** | 5-80 | 3 | 4 | 7 | 5 |
| **U2OS** | 7-80 | 3 | 4 | 7 | 5 |
| **HeLa** | 8-80 | 3 | 4 | 7 | 5 |

### Erythrosin B stain

| | Cell size | Sensitivity level | Correction factor | Viability level | Exception level |
|---|---|---|---|---|---|
| **CHO** | 5-80 | 3 | 4 | 5 | 5 |
| **U2OS** | 7-80 | 3 | 4 | 5 | 5 |
| **HeLa** | 8-80 | 3 | 4 | 5 | 5 |

# Quality control

## QC with Beads

EVE™ HT A26 offers two quality control methods (using QC beads and QC plate) to confirm its functions at various steps including sample preparation, image acquisition and image analysis. QC with beads verifies proper operation of EVE™ HT A26 sample preparation and imaging functions. EVE™ HT A26 offers two different QC beads for bright field based assays and fluorescence based assays.

1. Shake a tube containing QC beads vigorously or vortex it briefly for 5 seconds before use.

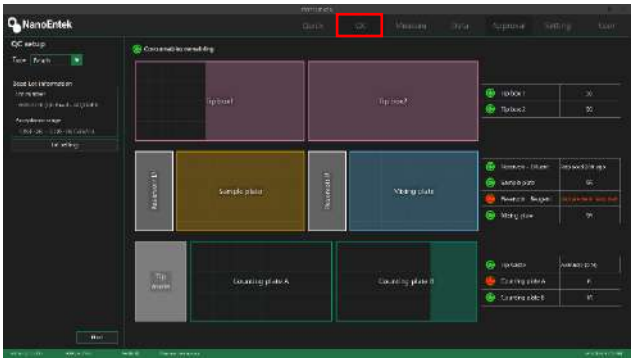
[Shake bottle vigorously]


[Vortexing]

2. Dispense **30 µL of well-mixed beads from the tube into multiple wells in a sample plate**, and place the sample plate in the sample plate holder.
☛ ***Note:*** *It is recommended to use at least one full column (8 wells) to verify the operation of the 8-channel multichannel pipette.*

3. Add a sufficient amount of reagent matching QC beads to a reservoir, and place it in Reservoir R.
**FL test beads**: Use AO/DAPI stain.
**BR test beads**: Use Trypan blue or Erythrosin B stain.

4. In the software, click the QC tab.




5. In the “Type” of the QC Setup menu on the left, confirm that **‘Beads’** is selected. If **‘Plates’** is selected, change it to Beads.

## QC with Beads

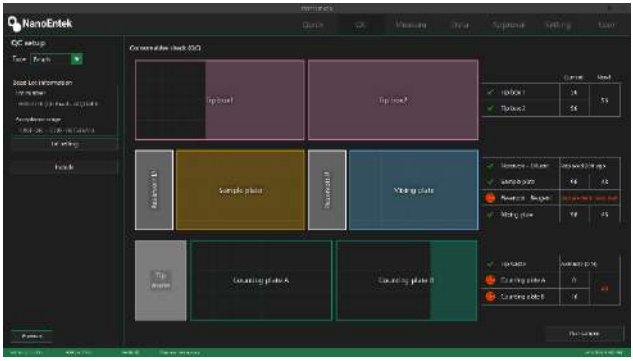
6. Check the bead lot information. If it differs from the beads used, click the '**Lot Setting'** button and select the matching lot from the list or register a new lot based on the information in the label attached to the QC bead tube.

7. Make sure that the remaining quantities of consumables match those in the instrument. If needed, edit remaining quantities to match the current status.

8. Click **Next** to proceed.


9. A sample plate map appears on the right. Select all wells into which beads have been dispensed, and click **'Include'** button or select 'Include' option from sub-menu by right clicking. Make sure that all the selected wells are displayed in green as shown above.

10. Click **Next** to proceed.




11. Check the required quantities of consumables and replenish any consumables if needed.

12. Click **'Run Sample'** to start QC using beads.

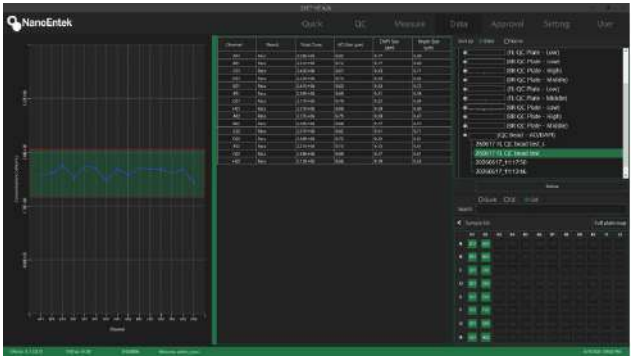
# Quality control

## QC with Beads

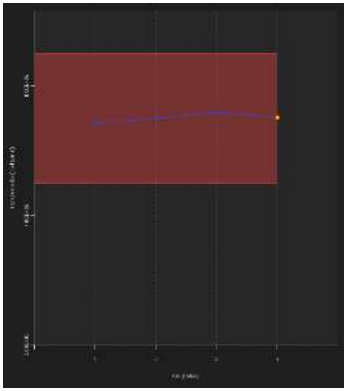
EVE™ HT A26 performs sample preparation and imaging, and the progress can be monitored on the screen as shown in “Measure”.

See page 29–30 for details.

After measurement is completed, select data from the data list and check the dot graph as shown below. One can check total counts and average sizes of beads.




If one clicks on a bead lot number in the data list, one can see previous results of the selected lot of QC beads.

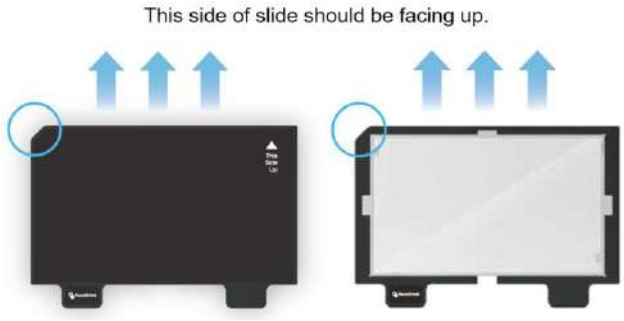


## QC with Plates

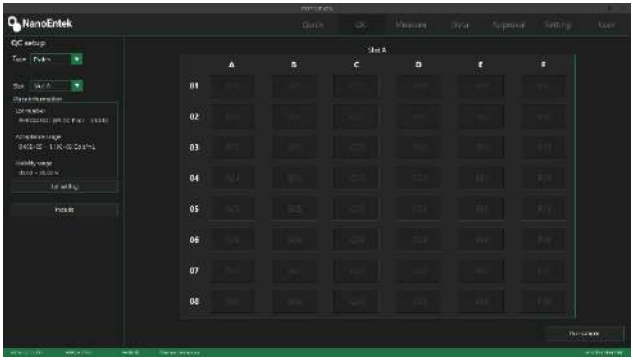
Unlike Bead QC, Plate QC verifies only the proper operation of EVE™ HT A26 imaging function.

1. Take out a QC Plate from a protective case.

2. Insert the QC Plate into the counting plate holder.




3. In the software, click the QC tab.




4. In the “Type” of the QC Setup menu on the left, confirm that **‘Plates’** is selected. If **‘Beads’** is selected, change it to Plates.

5. Select the slot. **Slot A** is on the left side of the counting plate holder, and **slot B** is on the right side. Select the slot where the QC plate is installed.

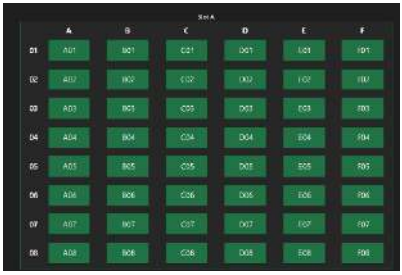
☛ ***Note:*** *Only one plate can be measured at a time. Simultaneous measurement of two plates is not supported.*

## QC with Plates

6. Verify the lot information of the QC plate. If it differs from the plate used, click the **'Lot Setting'** button and select a right lot from the list or register a new lot based on the information in the label attached to the protective case.

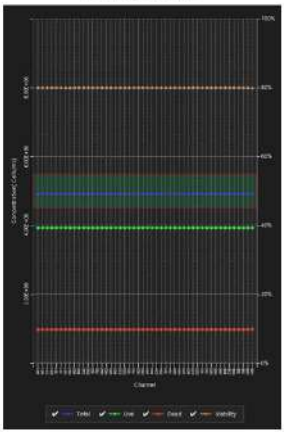
7. Select all 48 channels. For multiple selection, left-click and drag.

8. Click **'Include'** button or select 'Include' option from sub-menu by right clicking. Make sure selected wells are displayed in green as shown.

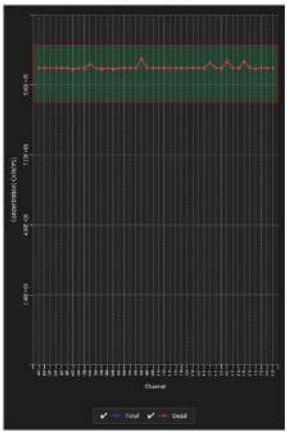


9. Click **'Run sample'** button to start measurement.
After measurement is completed, select data from the data list and check the dot graph as shown below.

BR QC Plate



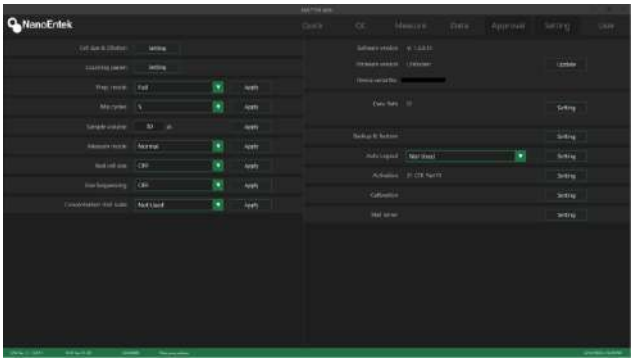
FL QC Plate



If one clicks on a bead lot number in the data list, one can see previous results of the selected lot of the QC plate.

# Setting

## Setting tab



In **Setting**, you can set default values for parameters and dilution factors. These value sets will be used when a cell type is not specified or a new cell type is created.

| | |
|---|---|
| **Cell size & Dilution** | Cell size and dilution factor of sample |
| **Counting parameter** | Parameter values for each BRIGHT, AO, and DAPI channel |
| **Prep. mode** | Default preparation mode (Full, Direct or None) |
| **Mix cycles** | Default value of mixing cycles |
| **Sample volume** | Default value of sample volume |
| **Measure mode** | Default imaging mode for AO/DAPI staining |
| **Real cell size** | Default real cell size setting for AO/DAPI staining |
| **Use Sequencing** | **Default setting for assigning sequential number after the sample name** |
| **Concentration Unit Scale** | Fixes the display of concentration values in exponential format (1.00E+01 to 1.00E+09).<br>**Not applied to QC data* |
| **SW, HW information** | Information about installed software and hardware |
| **Data path** | Path where measurement images and data are saved |
| **21 CFR part 11 activation** | Activate an optional program that complies with 21 CFR part 11 |
| **Calibration** | Calibration of brightness<br>Calibration of the instrument image background level (Only for Trypan blue or Erythrosin B stain) |
| **Mail server** | **Do not change Mail settings.* |

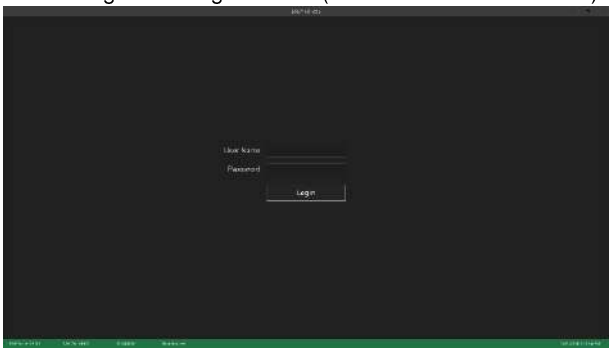
☛***Note**: Mix ratio (sample 20 µL + reagent 20 µL) is already applied, so do not include it in the dilution factor.*

# 21 CFR part 11

The Food and Drug Administration (FDA) of the United States has established regulations on electronic records and electronic signatures (ERES) in Title 21 of the Code of Federal Regulations, specifically 21 CFR Part 11. EVE™ HT A26 offers 21 CFR part 11 compliance program for cGMP facilities as an option. Please contact technical support to add this option.

## Log In

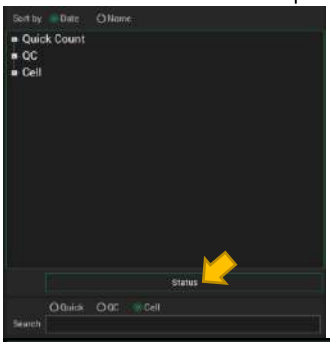
When activating 21 CFR Part 11, the user must log in on the first screen. Log in from login screen. (default ID/PW : admin/0000)

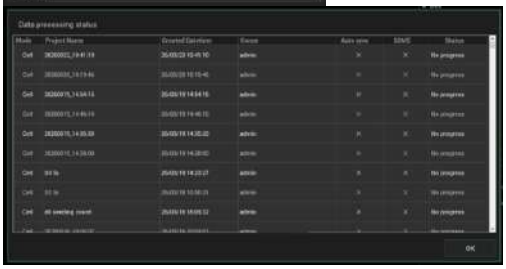



☛ ***Note**: Log out is in the User tab.*

## Data status

Users can check data status upon 21 CFR Part 11 activation.



## Approval

In the approval tab, you can request or approve data.

You can check the report file for the data by double-clicking the data or clicking the Preview button.

Data list is divided into four status (Requestable, Requesting, Approvable, Approved) types depending on the approval status.

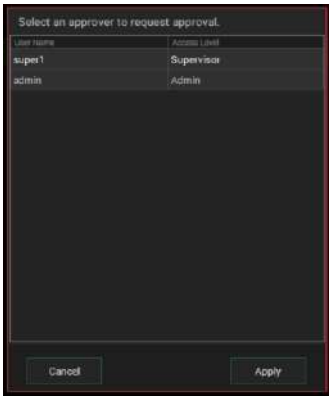
1. **Requestable**

The list shows data that can be requested for approval.

① Select the data to get approval.

② Click the **'Request'** button.

③ Enter the requester's password.




④ Select an approver to request approval.

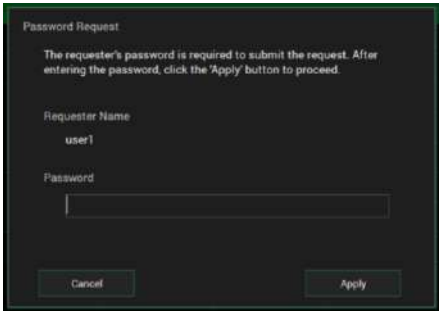
⑤ Click the **'Apply'** button.

## Approval

2. **Requesting**
The requesting data are displayed. It is possible to **'Cancel'** the request for approval. The approver can approve the request in Requesting tab. The approval no need to log in to the approval ID.

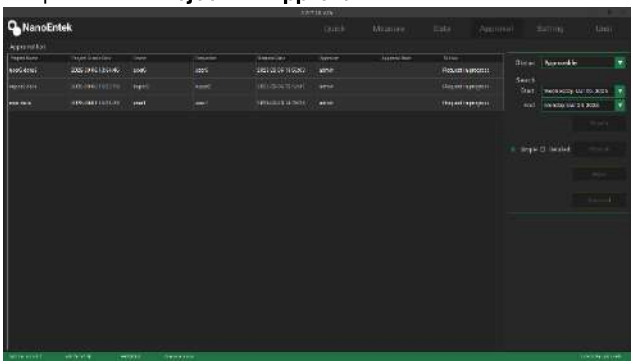
① Select the Data in list.

② Click the **'Approval'**.




③ Enter the approver's password.

3. **Approvable**
The approvable data are displayed on this tab.
It is possible to **'Reject'** or **'Approval'**.




4. **Approved**
The approved data is listed.
It is possible to **'Export'** the Approved data.

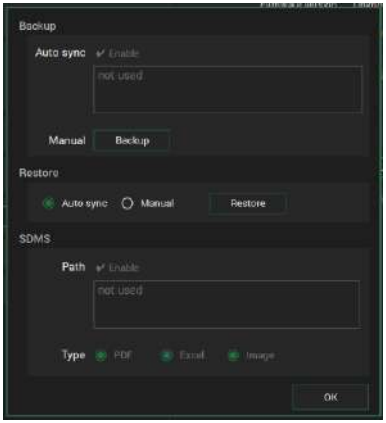
# 21 CFR part 11

## Setting

In the 21 CFR part 11 program, several functions are added to the setting tab.

1. Backup & Restore

① Click the Backup & Restore **'Setting'** button.




② To enable automatic backup, click the Auto sync **'Enable'**, set the backup data path.

③ Click the Manual **'Backup'** button, save the backup data at the current point in time.

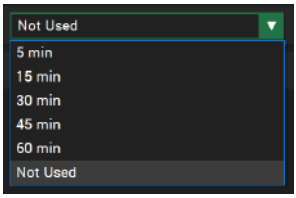
④ The Restore function provides two options. You can back up based on the backup data saved by Auto sync or the backup data saved by Manual.

⑤ Click the SDMS Path **'Enable'**, set the SDMS data path.

⑥ Select the desired type of SDMS data.

2. Auto logout

① Click the '▽' button.



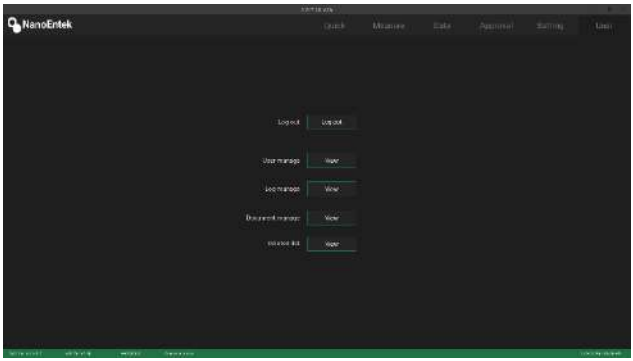

② Select the auto logout limit time.

③ Click the Auto Logout **'Setting'** button.

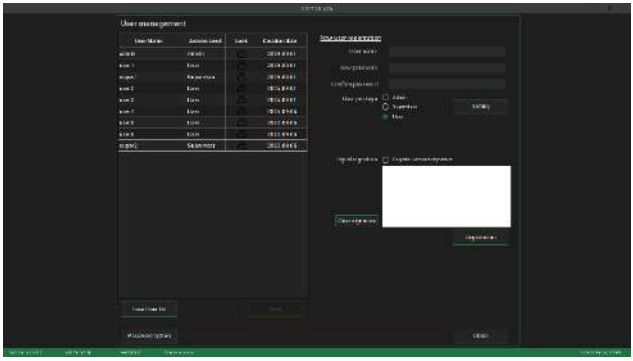
# 21 CFR part 11

## User manage

When the 21 CFR part 11 program is activated, the user tab becomes available.




Click the User manage **'View'** button.



1. New user registration.

① Enter the user ID and PW.

② Click the User privilege **'Setting'**.

③ Set the User privilege in each menu.

④ Set the User and Supervisor permission.

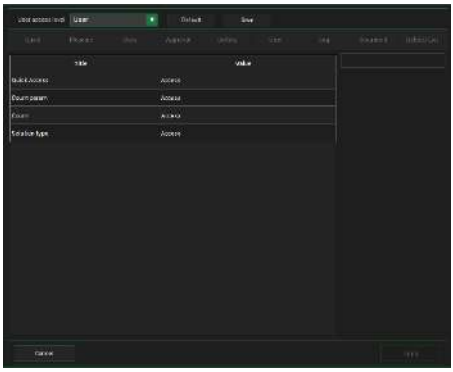
⑤ Click the **'Apply'** button.

⑥ Enter the signature and Click the **'Registration'.**

☛ ***Note**: **'Registration without signature'** is allowed, and the user will be required to register their signature at the time of first login.*

# 21 CFR part 11

## User manage



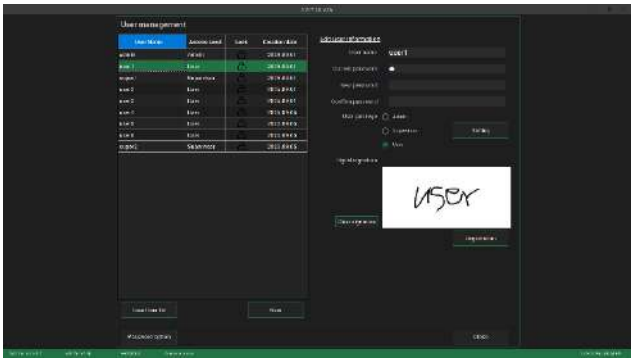
User and Supervisor default permission settings can be changed.
- Set the privilege and click the **'save'** button.
- See the 21 CFR part 11 Supplement for default settings.

2. Edit the user option

① Select the user in user list.

② Do the same process in Creating New user.

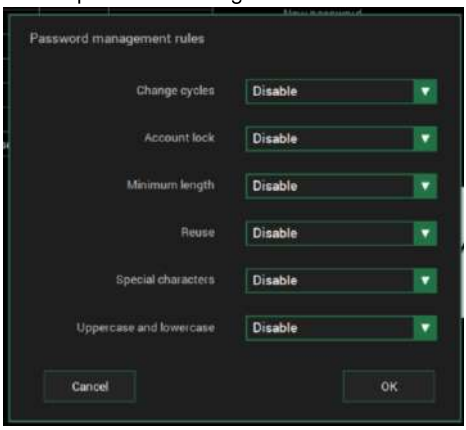
③ Depending on the user's granted privileges, the account can be locked.



## User manage

3. Password option

Set the password management rules.




① Change cycles.
Disable, 30 days, 60 days, 180 days.

② Account lock
Disable, ≥ 3 times, ≥ 5 times, ≥ 10 times, ≥ 15 times.

③ Minimum length
Disable, ≥ 3, ≥ 5, ≥ 10, ≥ 15.

④ Reuse
Disable, ≥ 30 days, ≥ 60 days, ≥ 180 days.

⑤ Special characters
Disable, Enable.

⑥ Uppercase and lowercase
Disable, Enable.

4. Lock in user list

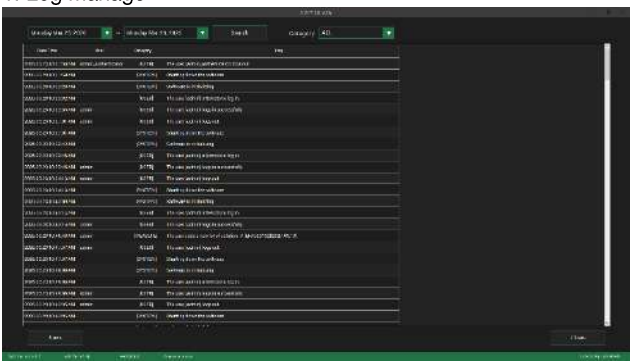
| User Name | Access Level | Lock | Creation date |
|---|---|---|---|
| admin | Admin | | 2026.01.15 |
| supervisor | Supervisor | | 2026.01.21 |
| user | User | | 2026.01.21 |

User ID is locked when login fails. Lock icon turns red. Click the button to unlock user ID and the button changes to grey.

# 21 CFR part 11

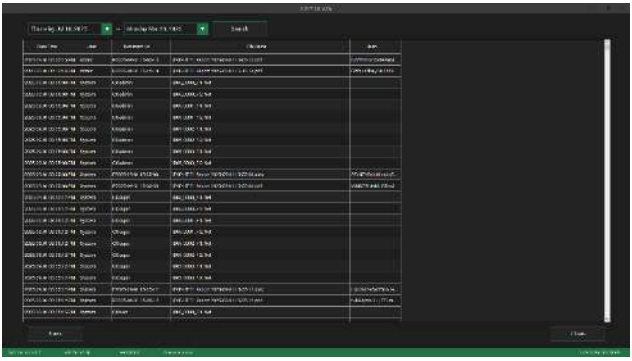
## Log manage, Document manage, Deleted list

1. Log manage



This log manage allows you to view a list of instrument log.

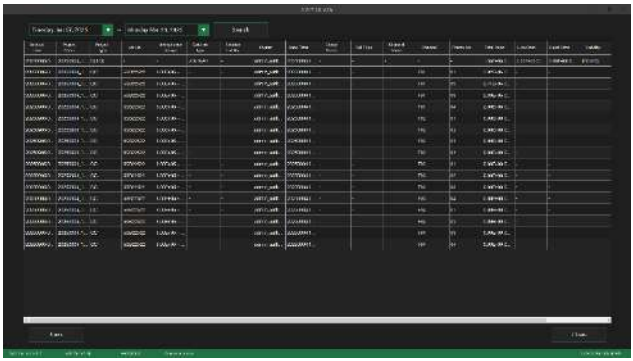
2. Document manage



This document manage allows you to check the documents made in the equipment.

## Log manage, Document manage, Deleted list

3. Deleted list



This deleted list allows you to view a list of deleted data.

☛ ***Note**: EVE™ HT A26 provides a comprehensive solution to comply with the requirements of 21 CFR Part 11. For more information on these features, please refer to the 'Support for 21 CFR Part 11'.*

# Maintenance and Cleaning

Clean the surface of EVE™ HT A26 instrument with a damp cloth. If liquid spills on EVE™ HT A26, turn off the power immediately and wipe dry.

EVE™ HT A26 does not need regular maintenance. To troubleshoot problems with EVE™ HT A26, contact technical support.

⚠ **IMPORTANT! Never disassemble or service EVE™ HT A26 by yourself.**

Unauthorized repairs may damage EVE™ HT A26 or alter its functionality, which will void your warranty. Contact **sales@nanoentek.com** or your local distributor to arrange for service.

⚠ **IMPORTANT! Always wipe surfaces with ethanol-soaked paper towels.**

Do not directly spray ethanol anywhere on EVE™ HT A26.

⚠ **IMPORTANT! Avoid exposing EVE™ HT A26 to UV light.**

UV light may degrade components, including plastic.
Damage from UV exposure is not covered under the manufacturer's warranty.

# Troubleshooting

| | |
|---|---|
| **Hardware** | |
| Pipette does not move to home position after power on | • Restart both EVE™ HT A26 instrument and PC.<br>• If the issue persists, contact the manufacturer. |
| Pipette tips are not securely sealed | • Check the tip boxes are properly installed in the tip box holders.<br>• Make sure to use EVE™ HT A26 compatible pipette tips. |
| **Software / System** | |
| Software does not launch | • Check cable connections and restart both instrument and PC.<br>• Update software to the latest version. |
| Software becomes unresponsive or crashes unexpectedly | • Restart the software.<br>• Reconnect cables.<br>• If the issue persists, contact the manufacturer. |
| **Optical measurement** | |
| Fluorescence signal is weak or nonexistent. | • Run calibration beads to verify hardware performance.<br>• Check reagent expiration date and storage conditions.<br>• Make sure to add enough volume of reagent to reservoir. |
| Fluorescence signal is very bright. | • Run quality check to verify hardware performance.<br>• Check reagent expiration date and storage conditions.<br>• Make sure that cell samples have sufficient volume in sample well plate. |
| Inaccurate results in Bright field counting reagents | • Bright field-based staining methods (Trypan Blue or Erythrosin B) are known to produce inaccurate cell counting results when there are debris or red blood cells in samples. Consider using fluorescence-based staining method using AO/DAPI solution. |
| Incorrect focus | • Save the correct focus for each cell type in Quick tab. Refer to page 37. |

# Troubleshooting

| Inaccurate result | |
|---|---|
| Cell counts lower than expected | • Make sure that cells are not clumped and well dispersed.<br>• Make sure that there are sufficient volumes of cell samples in sample well plate. The required sample volume varies depending on the selected mode — refer to page 11 for details. |
| Cell counts higher than expected | • Inspect presence of debris or contamination.<br>• If needed, adjust cell counting parameters and/or cell size gating to exclude incorrectly counted objects. |
| Cell counts above or below measurable ranges | • EVE™ HT A26 is designed to measure samples from 1 × $10^4$ cells/mL to 2 × $10^7$ cells/mL.<br>• If samples are above or below these ranges, make sure to either dilute or concentrate samples before running measurements. |
| Dilution factor | • The 'Dilution factor' in sample information (as set in Setup Wizard Step 1) refers only to the dilution manually done by users before putting samples in sample well plate.<br>• Dilution functions will update 'Dilution factor' automatically.<br>• Dilution due to adding reagents will be automatically applied to final results. |
| Dusts or bubbles | • Avoid making bubbles during sample preparation.<br>• If needed, adjust cell counting parameters and/or cell size gating to exclude incorrectly counted objects.<br>• Remove dusts or bubbles using the image edit function. Refer to page 35. |
| Too big or too many clumpy cells | • Increase the number of sample mixing cycles to help disaggregate clumped cells.<br>• It is highly recommended to start measurements within 30 minutes after preparing sample well plate to avoid cell aggregation. |
| **Problems in sharing data** | |
| E-mail | • Check the internet connection. |
| USB storage | • Check the storage path. |

# Warranty

NanoEntek provides (1) year warranty service for defects of material and workmanship.

If any defects occur in EVE™ HT A26, NanoEntek provides repair services for the defective parts at its discretion.

The following defects, however, are specifically excluded:

1. Defects caused by improper operation.
2. Repair or modification done by anyone other than NanoEntek or an authorized agent.
3. Damage caused by substituting alternative parts.
4. Use of fittings or spare parts supplied by anyone other than NanoEntek.
5. Damage caused by accident or misuse.
6. Damage caused by disaster.
7. Corrosion caused by improper solvent or sample.

For your protection, EVE™ HT A26 units being returned must be insured against possible damage or loss. NanoEntek cannot be responsible for damage incurred during shipment of a defective instrument. It is recommended that you save the original packing material in which the instrument was shipped. This warranty is limited to the replacement of defective products.

For any inquiry or request for repair service, please contact **sales@nanoentek.com** or your local distributor.

# Safety precautions

Review and follow the safety instructions below:

- If water or other material enters the instrument, the adaptor, or power inlet, disconnect the power cord and contact a service person. For operating environment, refer to Product Specifications.
- Do not touch the main plug or power cord with wet hands.
- Always ensure that the power supply input voltage matches the voltage available at your location.
- This instrument is air-cooled and its surfaces may become hot during operation. When installing, leave a space of more than 10 cm (4 inches) around the instrument and do not place any objects between the instrument and walls.
- Do not install an instrument on a slant or a place prone to vibrations, which induces the risk of malfunction or damage of the instrument.
- Never insert any objects into the air vents of the instrument as this can result in electric shock, personal injury, and equipment damage.
- Plug the power cord firmly into the wall outlet and AC adapter.
- To avoid potential shock hazard, make sure that the power cord is properly grounded.
- Be sure to position the instrument such that it is easy to disconnect.
- Turn off an instrument before unplugging the power cord and/or moving the instrument.
- If an instrument is dropped or broken, disconnect the power cord and contact a service person. The warranty will be void in case of disassembly.
- Use only authorized accessories (adaptor, power cord, and USB drive).

⚠ ***WARNING***

***Class A equipment is intended for use in an industrial environment. In the documentation for the user, a statement shall be included drawing attention to the fact that there may be potential difficulties in ensuring electromagnetic compatibility in other environments, due to conducted as well as radiated disturbances.***

# Mesures de sécurité

Examiner et suivre les instructions en matière de sécurité ci-dessous:

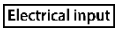
- Si de l'eau ou d'autres matières entrent dans l'instrument, l'adaptateur, ou l'entrée de la prise, débrancher le cordon d'alimentation et contacter un technicien de service. Pour l'environnement d'exploitation, se reporter aux Spécifications du Produit.
- Ne pas toucher la prise principale ou le cordon d'alimentation avec les mains mouillées.
- S'assurer toujours que la tension d'alimentation correspond à la tension disponible à votre localisation.
- Cet instrument est refroidi à l'air et ses surfaces peuvent devenir chaudes pendant le fonctionnement. Lors de l'installation, laisser un espace de plus de 10 cm (4 pouces) autour de l'instrument et ne placer aucun objet entre l'instrument et les murs.
- Ne pas installer d'instrument sur une pente ou un endroit sujet aux vibrations, qui entraînent un risque de défaillance ou de détérioration de l'instrument.
- Ne jamais insérer d'objets dans les évents d'air de l'instrument, car cela peut causer des chocs électriques, des blessures corporelles et des dommages de l'instrument.
- Mettre le cordon d'alimentation fermement dans la prise murale et l'adaptateur courant alternatif.
- Pour éviter tout risque de choc, s'assurer que le cordon d'alimentation est correctement mis à la terre.
- S'ssurer de positionner l'instrument de telle sorte qu'il soit facile à débrancher.
- Éteigner l'instrument avant de débrancher le cordon d'alimentation et/ou de le déplacer.
- En cas de chute ou de rupture d'un instrument, débrancher le cordon d'alimentation et contacter un technicien de service. La garantie sera annulée en cas de démontage.
- Utiliser uniquement les accessoires autorisés (adaptateur, cordon d'alimentation et clé USB).

## ⚠ *AVERTISSEMENT*

***L'équipement de classe A est destiné à être utilisé dans un environnement industriel. Dans la documentation pour l'utilisateur, une déclaration doit être incluse pour attirer l'attention sur le fait qu'il peut y avoir des difficultés à assurer la compatibilité électromagnétique dans d'autres environnements, en raison de perturbations aussi bien conduisées que radiées.***

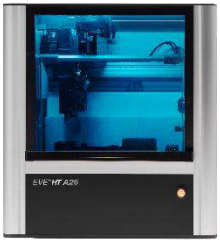
# Safety Symbols

The following symbols are found on the medical device and this document. Always use the instrument in the safest possible manner.

| Symbol | Meaning |
|---|---|
|  | Caution & Warning |
|  | Protective earth (Ground) |
|  | Power On/Off |
|  | The moving parts symbol indicates areas of the medical device in which moving parts can cause injuries.<br>Do not operate the medical device with the door open. |
|  | This device and consumables conforms to the EC Declaration of Conformity. |
|  | This equipment has been tested and found to comply with the limits for a Class A digital medical device, pursuant to Part 15 of the FCC Rules.<br><br>These limits are designed to provide reasonable protection against harmful interference when the equipment is operated in a commercial environment. This equipment generates, uses, and can radiate radio frequency energy and, if not installed and used in accordance with the instruction manual, may cause harmful interference to radio communications. Operation of this equipment in a residential area is likely to cause harmful interference in which case the user will be required to correct the interference at his own expense. |
|  | USB Connection |
|  | This product conforms to UL 61010-1, CAN/CSA C22.2 No.61010-1 “Safety Requirements for Electrical Equipment for Measurement, Control, and Laboratory Use, Part I: General Requirements.” This instrument bearing the TÜV symbol are certified by TÜV Product Services to be in conformance with the applicable safety standard for the US and Canada. |
|  | Catalogue number/Reference number |
|  | Serial number |
|  | Manufacturer |
|  | Electrical input |
|  | Consult Instructions for Use<br>An electronic instructions for us (eLFU) indicator (website address) may accompany the symbol when used to indicate an instruction to consult an eIFU. |

| | |
|---|---|
|  | Disposal of your old appliance<br>1. When this crossed-out wheeled bin symbol is attached to a product it means the product is covered by the European Directive 2012/19/EU.<br>2. All electrical and electronic products should be disposed of separately from the municipal waste stream via designated collection facilities appointed by the government or the local authorities.<br>3. The correct disposal of your old appliance will help prevent potential negative consequences for the environment and human health.<br>4. For more detailed information about disposal of your old appliance, please contact local distributor, waste disposal service or call the number listed in the manual. |
|  | LED |
|  | Physician.<br>Keep dry<br>Keep away from rain |
|  | Fragile, handle with care |
|  | This way up |
|  | General symbol for recover/recyclable |
|  | Team lift |
| US Corporation | US Corporation |
| European Corporation | European Corporation |
| EC REP | Authorized representative in the European community |
| UK Representative | Authorized representative in United Kingdom |
| CH REP | Authorized representative in Switzerland |
| BRH | Authorized representative in Brazil |

# Product specifications



| EVE™ HT A26 | |
|---|---|
| **Analysis time** | 96 samples in 10 minutes |
| **Sample volume** | 20 µL per count (Direct mode)<br>≥ 30 µL per count (Full mode) |
| **Measuring range** | Detectable range: $1 \times 10^{4}$–$2 \times 10^{7}$ cells/mL<br>Optimal range: $1 \times 10^{5}$–$1 \times 10^{7}$ cells/mL |
| **Cell size range** | Detectable size : 1–85 µm<br>Optimal size: 5–80 µm |
| **Channel** | Dual fluorescence (AO & DAPI), Bright field |
| **Staining solution** | AO/DAPI mixed solution<br>Trypan blue solution<br>Erythrosin B solution |
| **Capacity** | 96 samples per run |
| **Pipette head** | 8-channel pipette head |
| **Operation system** | Windows 11 LTSC |
| **Power** | 100–240VAC, 50/60Hz, 110W |
| **Dimensions** | 700 × 690 × 856 mm (W×D×H) |
| **Weight** | 147 kg |
| **21 CFR Part 11 Option** | Available (Optional) |

# Ordering information

| Cat. No. | Description | Contents |
|---|---|---|
| **EVE-HT A26** | Fully Automated Cell Counting System | Main device<br>Desktop PC<br>Monitor |
| **EVA26-020** | EVE HT A26 Counting kit | 960 tests / kit<br>Counting plate (48 channels × 20 plates)<br>Sample & mixing well plate (96 wells × 20 plates)<br>Reservoir (5 pcs × 4 packs) |
| **EVA26-020S** | EVE HT A26 Starter kit | Counting kit (1kit)<br>Disposable pipette tips (5 trays x 3 packs)<br>AO/DAPI staining solution (20 mL x 2 bottles) |
| **EVAD-960** | AO/DAPI Staining Solution | 20 mL × 2 bottles<br>- Expires 2 months after opening |
| **EVAD-961** | AO/DAPI Staining Solution | 50 mL × 2 bottles<br>- Expires 2 months after opening |
| **EVTB-960** | Trypan blue Stain | 20 mL × 2 bottles<br>- Expires 6 months after opening |
| **EVTB-961** | Trypan blue Stain | 50 mL × 2 bottles<br>- Expires 6 months after opening |
| **EVEB-960** | Erythrosin B stain | 20 mL × 2 bottles<br>- Expires 6 months after opening |
| **EVEB-961** | Erythrosin B stain | 50 mL × 2 bottles<br>- Expires 6 months after opening |
| **EVPT-960** | EVE HT A26 Disposable pipette tips | 96 tips x 5 trays x 3 packs |

# Ordering information

| Cat. No. | Description | Contents |
|---|---|---|
| **EHGQ-001** | EVE HT FL&A26 QC Plate Fluorescence (optional) | Low level, 1pc |
| **EHGQ-002** | EVE HT FL&A26 QC Plate Fluorescence (optional) | Middle level, 1pc |
| **EHGQ-003** | EVE HT FL&A26 QC Plate Fluorescence (optional) | High level, 1pc |
| **EFB-001** | EVE HT series Test Beads Fluorescence (optional) | 1 x 1 mL / Pack |
| **EHBQ-001** | EVE HT FL&A26 QC Plate Bright (optional) | Low level, 1pc |
| **EHBQ-002** | EVE HT FL&A26 QC Plate Bright (optional) | Middle level, 1pc |
| **EHBQ-003** | EVE HT FL&A26 QC Plate Bright (optional) | High level, 1pc |
| **EHB-001** | EVE HT series Test Beads Bright (optional) | 1 x 1.5 mL / Pack |
| **EVE HT A26 21 CFR Part 11** | EVE HT A26 21 CFR Part 11 software (optional) | 21 CFR Part 11 software |

# Technical support

Visit our Website at **www.nanoentek.com** for:

- Technical resources, including manuals, FAQs, etc.
- Technical support contact information
- Additional product information and special offers

For more information or technical assistance, please call or email.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# EVE™ HT A26

NESMU-EHA26-001EN (V.0.0)


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