

## FlowCAM – Fluid Imaging

(10x) VS Series/PV-100/8100, (20x) VS Series/8100, (2x) VS Series, (4x) VS Series/PV-40/8100, Nano (40x)

### INTRODUCTION

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The Lumetics LINK™ software platform scans network locations for new measurement data files, copies data directly to a centralized database, and provides a powerful user interface for rapid multi-measurement multi-technique data aggregation, visualization, analysis, and reporting. LINK employs a client/server-based architecture where the LINK server hardware is provided by the end user and resides on the end user's network. The LINK client is a portable web-based application that may be placed on any computer with network connectivity to the LINK server. For successful import, the LINK webserver requires read access to the folders where user data resides.

FlowCAM rapidly counts and images particles in a fluid stream in order to characterize the particles using a variety of measurements.

### DETAILS

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LINK requires the CTX, LST, and TXT files for VisualSpreadsheet (ViSP) 4 and earlier. LINK may directly connect to FlowCAM database and import data from ViSP 5. Particle images (TFF) files and Notes/Comments TXT are optional. The FlowCAM system generates the required files automatically.

The following raw curve data may be imported, in addition to all available instrument/analysis settings and parameters calculated by the instrument software:

- Particle count or concentration vs. morphology

The following are advanced morphological parameters calculated upon import:

- Geodesic Aspect Ratio
- Geodesic Length
- Geodesic Thickness

Particle count/concentration for each supported morphological parameter occurs at a pre-defined bin spacing and measurement range as per the table in the Users Manual located in the software help menu (section 4.14). Default measurement ranges will dynamically widen based as required, based on the datasets imported to LINK.

- Particle Volume % for the number of particles in each 0.25um bin is calculated automatically and imported to LINK. (Particle Volume % = volume of particles in a given size range bin divided by the volume of particles in all bins, assuming particles are spherical in volume).

LINK allows direct database connection to ViSP 5 version 5.2.25. This connection is established when importing FlowCAM data, selectin ViSP 5 from the drop down and adding the FlowCAM database as a connection. The username and password for the FlowCAM database can be accessed from within the ViSP 5 software via the *File -> Show Read Only Login* menu option and requires a read-only license to be purchased from Fluid Imaging before using. For easy measurement search, the keyword field will query the 'name' & 'notes' fields in ViSP 5, and the date range field allows for custom date search ranges. Runs from ViSP 5 that have deleted particles or are flagged as "deleted" will not be imported.

The second requirement for successful ViSP 5 support involves activation of read only tables that LINK requires access to. The tables we expect to see from the ViSP software are:

- context\_history
- image\_frame



- particle
- particle\_image
- particle\_property
- run
- run\_summary

If none of these tables are visible, please follow the below instructions:

- Navigate to the ViSP 5 app to File -> "Update Read Only Tables"
- The software will prompt for the Password to the database

Proceed to add the missing tables

### Import Data

To start an import job, please select the instrument, the data location (typically a network share that the LINK Server can access), an Import Method, and a Project to import the data into.

Select Instrument:

Select Database Connection:

Search Keywords:

Date Range:

Select Import Method:

Select LINK Project:

Overwrite Measurements (Metadata Fields will be preserved)

Note: LINK calculates particle concentration dynamically within LINK using the particle counts and the volume analyzed. To obtain the volume analyzed, the ml is stripped from the "Fluid Volume Imaged" field value and used the number for LINK calculations. If the run\_summary file reports both Count and Concentration values, these are used for Volume Analyzed and Sample Analyzed field calculations. If This method is used, the system LINKdb field 'LNK\_Volume\_Analyzed\_Calculated' will note a value of "yes" instead of "no". If using ViSP v4.x, for calculated concentrations to line up with Visual Spreadsheet, select the Context option from the Toolbar, select Reports, and select the "Export list summary when run terminates" option.

The screenshot shows the VisualSpreadsheet interface with four graphs of Frequency vs. Diameter (ESD). The graphs show a distribution peaking around 19.10 and tapering off towards 37.20. A 'Context' dialog box is open, showing options for Exports and Classification. The 'Export list summary when run terminates' option is checked. The 'Classification' section has 'Generate classification when run terminates' and 'Export classification summary when run terminates' options, both unchecked. A 'Classification Template' field is visible with a 'Find...' button. The bottom status bar shows: Count 25837 of 25837, Efficiency 23.3%, Start Time 2014-01-29 12:48:52, Droplet mg/L NA, Particles/ml 204060, PPI NA, Sampling Time 3 min 34 sec, Solids mg/L NA.

Import Method options can be defined and calculated each measurement during import if it is first created/defined and then requested within an Import Method. More information and instructions can be found in the Users Manual located in the software help menu (section 4.13). The following is the default FlowCAM-specific import method settings:

- Default Sub-Populations: **None**
- Custom Particle Parameters: **None**
- Container Fill Volume: **None**
- Number of Thumbnails to Crop: **250**
- Ignore Thumbnails less than: **5  $\mu\text{m}$**
- Import Images in order of Largest-to-Smallest: **Yes**
- Dilution Factor: **1 (no dilution)**

### **Subpopulations**

In addition to the total particle population data set, sub-populations based morphological parameter filters may be generated at the point of measurement import. An unlimited number of sub-populations may be specified for each measurement. For each sub-population, the particle count/concentration vs. morphological parameter will be available, as well as representative particle thumbnail images. The sub-populations do not count towards the measurement limit per project. Additional Sub-Populations will affect import speeds.

When creating as sub-population, select a Morphological Parameter from the pre-defined list and specify the filter criteria. Select the + to add the filter term to the Live Expression View. The Live Expression View can be edited directly. Brackets, mathematical operators/values, logical ANDs or ORs are valid operators. Syntax must be exact, for successful application.

### **Custom Particle Parameters**

Select from the available user-defined Custom Particle Parameters, those that shall be calculated for each individual particle within each measurement. This selection applies to the total particle population and all defined sub-populations.

### **Container Fill Volume**

Specify the Container Fill Volume, permitting LINK to calculate and report Particles per Container (in addition to Particle Count, and Particle Concentration). This selection applies to the total particle population and all defined sub-populations.

### **Stuck/Repeating Particles**

Ignore particles images which remain, or move slowly, in successive image frames. This selection applies to the total particle population and all defined sub-populations.

### **Thumbnail Images**

Specify the maximum number of particle thumbnail images to be imported to the LINKdb for each measurement population. Apply a size threshold (the default is 5 microns). Choose whether particles should be first sorted from largest to smallest (based on ECD) or imported in their order of appearance during the analysis. These selections apply to the total particle population and all defined sub-populations.

### **Automated Metadata Extraction**

Specify criteria for automated metadata extraction from measurement file fields (e.g. Comments, File Name, Import Path etc...) utilizing specified delimiters. Both metadata name and value may be extracted or only the value only.

### **Dilution Factor**

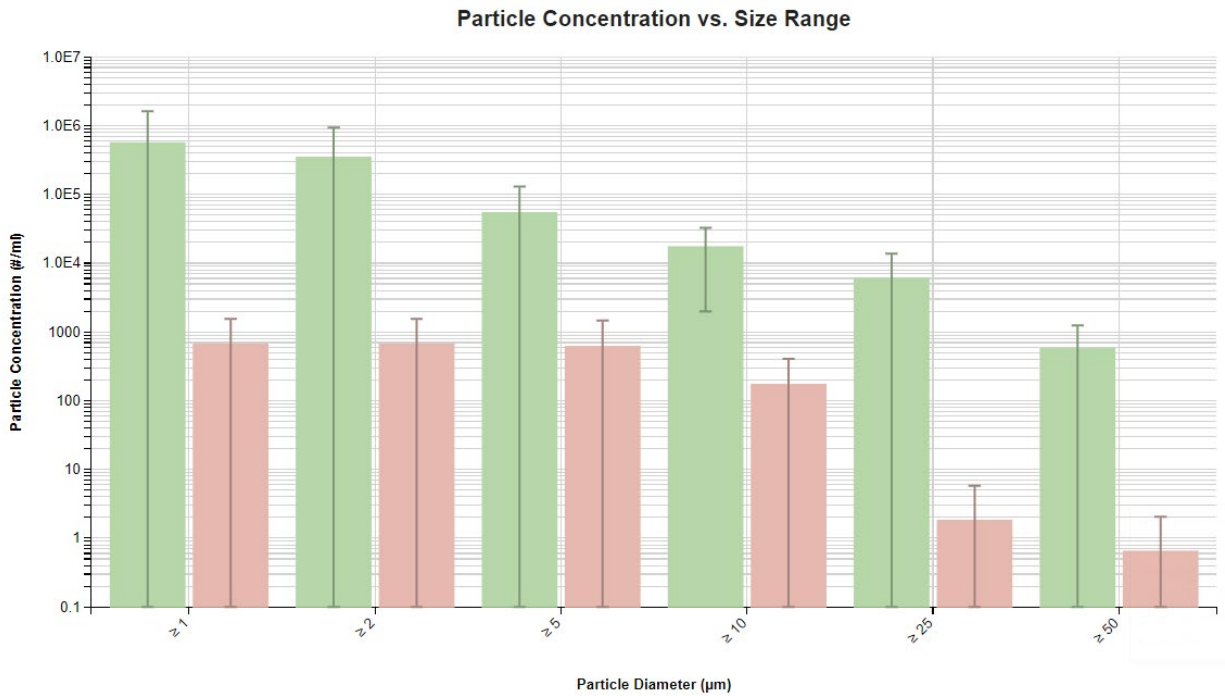
Specify a dilution factor for automated particle count/concentration data adjustment. This selection applies to the total particle population and all defined sub-populations. A LNK\_Dilution\_Factor may also be applied after import, utilizing the Add/Edit LINKdb Fields feature.



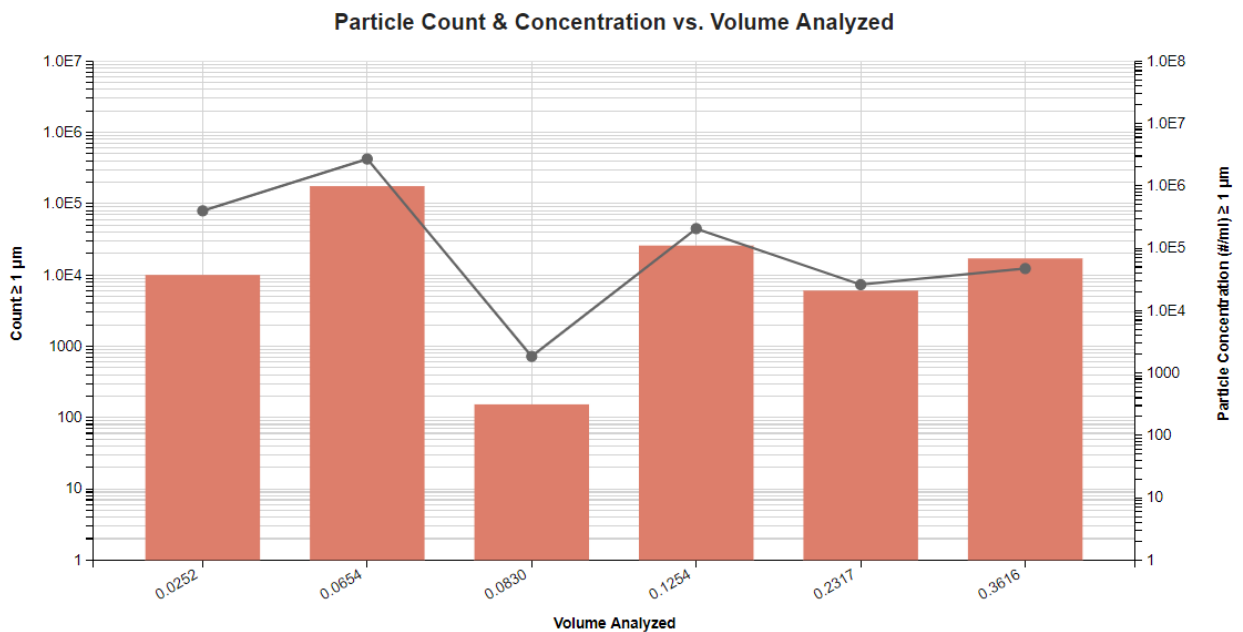
## EXAMPLES

Included below are example dashboards from FlowCAM measurement files:

### 1. Histogram plotting binned raw data for Particle Concentration vs. Particle Diameter



### 2. Column chart plotting Particle Count $\geq 1\mu\text{m}$ & Concentration $\geq 1\mu\text{m}$ vs. Volume





## DASHBOARD DOWNLOADS

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Included below is a link to downloadable dashboards for FlowCAM measurement files:

<http://lumetics.com/dashboards/FlowCAM/FlowCAM.zip>

## CONTACT LUMETICS

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For direct assistance, please contact Lumetics LINK™ Support:

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