

NanoSight – Malvern Panalytical

INTRODUCTION

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.

NanoSight instruments utilize Nanoparticle Tracking Analysis (NTA) to characterize nanoparticles in solution. They provide high resolution nanoparticle size, count-based concentration, and aggregation measurements while a fluorescence mode provides specific results for suitably labelled particles. With real time monitoring, subtle changes in the characteristics of particle populations are provided with all these analyses confirmed by visual validation.

DETAILS

LINK requires the Batch/Experiment Summary.CSV data file. The *.CAP used in older versions of NTA software (might contain sample name and analysis date) is optional. The Particle Count/Intensity CSV data file is optional. The AVI video file is not utilized. The NTA software generates all required files automatically. The NTA software generates all required files automatically however it is advised to export files as Individual vs. Summary, to avoid potential issues related to inconsistent bin spacing.

Particle Concentration data MUST be present in the CSV file for successful Import. If the same measurement exists in the Summary CSV and ExperimentSummary CSV files, LINK will ignore the redundant data. Each measurement in an ExperimentSummary CSV file is treated as a unique measurement. Each will be assigned the same Sample Name but will have a different File Name based on the Capture timestamp.

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

- Automated Metadata Extraction: None
- Dilution Factor: 1 (no dilution)

The NanoSight CSV data file example is as follows:



- 4	A	В	С	D	Е
1	[Software Info]				
2	Build	NTA 2.3	build 0017		
3	[Sample Information]				
4	Operator				
5	Sample Description				
6	Diluent				
7	Concentration				
8	Pre-treatmeant				
э	Comments				
10	[Results Summary]				
11	Total Frames	977			
12	Processed Frames	977			
13	Valid Tracks	251			
14	Overall Drift (nm/s)	166.09			
15	X-Drift (nm/s)	-50.33			
16	Y-Urik (nm/s)	-158.29			
17	Calibration (nm/pixel)	185			
18	Size Data - Concentration Weighted				
19	Mode (nm)	288			
20	Mean (nm)	530.7			
21	Standard Deviation (nm)	346.59			
22	Size data - Model Fit				
23	Mean (nm)	U			
24	Standard Deviation (nm)	U			
25	User Line 1	0			
26	User Line 2	0			
27	Average # centres/frame	25.76			
28	Average # particles/frame	17.58			
23	Equivalent particle concentration (E8 particles/mi)	2.09			
30	Selected average # particlesmame	0			
31	Selected equivalent particle concentration (E8 particles/mi)	U			
32	[Settings]	0-			
33	Diableses	011			
04	Chin	1			
26	Diur Cine	Auto I			
30	Detection Threshold Tune	Multi			
20	Detection Threshold Type	21			
29	Min Track Length	Auto			
40	Min Expected Size (nm)	Auto			
40	[Measurement Conditions]	Adio			
40	Camera Tune	<cm05< td=""><td></td><td></td><td></td></cm05<>			
43	Camera Sputter (ms)	18.38			
44	Camera Gain	135			
45	Frame rate (fns)	2173			
46	Camera bit-conversion limits	2145	8840		
47	Temperature (°C)	23			
48	Viscositu (cP)	0.931			
4.9	[Warnings]				
50	Low Completed Tracks	No			
51	High Noise	1frames			
52	Auto max jump	Success	ful		
53	Drift off	No			
54	Settings Changed Whilst Processing	No			
55	Vibration detected	Vibration	detected	<~10% er	ror
56	Additional Errors	None			
57	[Graph Data]				
58	Particle Size (nm)	Concent	ration (E6	particles	/ ml)
59	0	0			,
60	1	0			
61	2	0			
62	3	0			
63	4	0			

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.

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 is an example dashboard from NanoSight measurement files:

1. Line chart plotting raw data curves for Particle Parameters

Particle Diameter vs. Concentration



Particle Diameter (µm)

2. Tabular Summary examples

Measurement	Summary	Table –	Instrument Settings
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LINK Record ID #	InstrumentName	Sub-Population	Sample Name	Blur Size	Weighting	Version	Total frames analysed - AVG	Total Frames - AVG
58	NanoSight	Total Population	Sample 1	Auto	Number	NTA 3.2 Dev Bui	749	1500
59	NanoSight	Total Population	Sample 2	Auto	Number	NTA 3.2 Dev Bui	749	900
60	NanoSight	Total Population	Sample 3	Auto	Number	NTA 3.2 Dev Bui	749	1000
66	NanoSight	Total Population	Sample 4	Auto	Number	NTA 2.3 build 0	749	1499

Measurement Summary Table – Measurement Results

LINK Record ID #	Sample Name	Vibration detected	Viscosity (cP) - AVG	Volume (%) (Particle Diam. (μm) <=1) - AVG	Particle Conc. (#/ml) (Particle Diam. (µm) <1) - AVG
58	Sample 1	No	0.88	100.00	1268142878
59	Sample 2	No	0.88	100.00	1437700967
60	Sample 3	No	0.88	100.00	1439394116
66	Sample 4	No vibration detected	0.87	100.00	196717900



NANOSIGHT DASHBOARDS

LINK contains an extensive built-in dashboard library from LINK version 2.4.0.210401 and later. This function contains specific pre-created dashboards for all instruments and application groups.

CONTACT LUMETICS

For direct assistance, please contact Lumetics LINK[™] Support: **E-mail:** <u>support@lumetics.com</u> **Phone:** 1.613.417.1839 **Website:** <u>http://lumetics.com/</u>

