

Uncle – Unchained Labs

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.

Uncle is a one-stop protein stability platform that uses fluorescence, SLS and DLS detection to enable 12 different applications. Multiple measurements such as thermal melting, aggregation, and sizing can be performed with the same samples in just one experiment.

DETAILS

LINK requires at least one DLS/DSC tabular summary xlsx file with a sheet (**sheet1**) that is not empty. For DLS assays, optional XLSX curve data for Autocorrelation, Mass, and Intensity will be imported. For DSC assays, optional XLSX curve data for Fluorescence Peak Height, Fluorescence Peak Area will be imported. For dual assays, optional XLSX curve data for Z-Avg vs. Temperature and Derived Intensity vs. Temperature will be imported.

Helpful Notes:

- Each folder needs to have at least one of the two:
 - DSC-type table (containing a header column that includes "Tm")
 - DLS-type table (a table file that has not been tagged as DSC)
- LINK will create a single measurement per row in the table file, and if both a 'DSC' and 'DLS' table are detected, the table results will be combined.
- All data related to the same experiment/plate/samples should be in their own folder, along with corresponding table files (either the DLS-type, DSC-type, or one of each).
- Autocorrelation raw data file, melting curve raw data file, and Mass/Intensity raw data file must include "mass" or "intensity" in the file root name as either a prefix or suffix to the "uni-", and the root file name must remain intact.
Example: DLS_**Intensity**_Viral Toolbox redo 2_10_21.uni-2021-02-11T17-48-41
DLS_Viral Toolbox redo 2_10_21.uni-2021-02-11T17-48-41_**Mass**
- If a replicate run is present, then LINK will assign a Replicate Number accordingly. Replicate is defined as the same well position noted twice in the table file, and if applicable, two curves present in a corresponding raw data file.

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

- Autocorrelation Function vs. Delay Time
- Intensity % vs. Particle Diameter
- Mass % vs. Particle Diameter
- BCM/nm vs. Temperature
- SLS 266nm/Count vs. Temperature
- Z-Average vs. Temperature
- Derived Intensity vs. Temperature
- SLS 473nm/Count vs. Temperature
- Fluorescence – Area/nm vs. Temperature
- Fluorescence – Peak Height/Count vs. Temperature

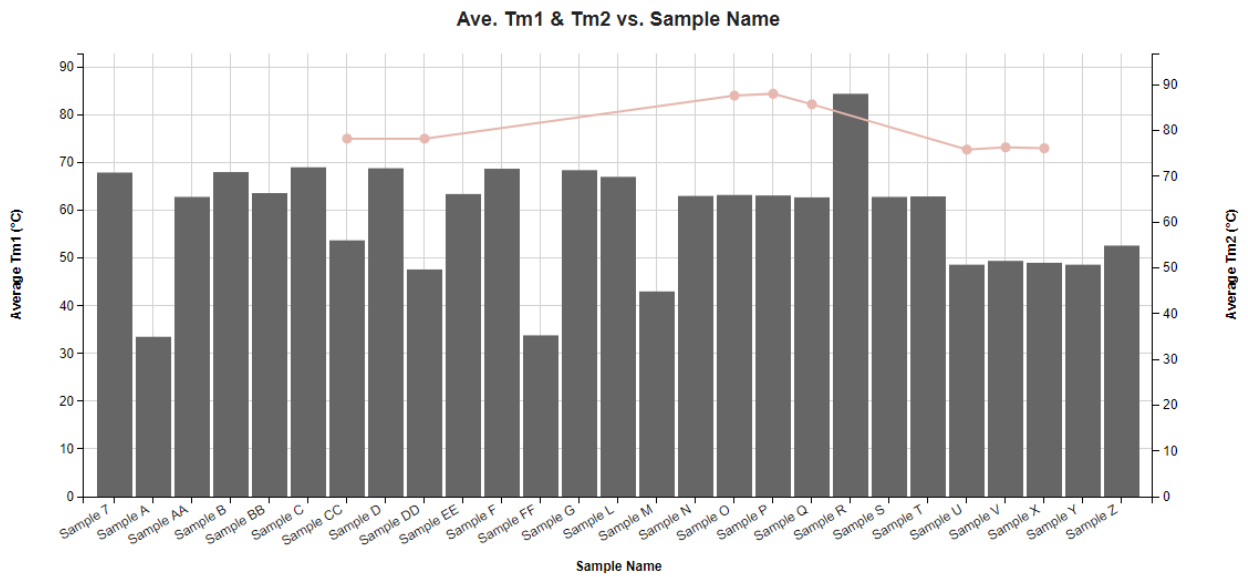
The Uncle XLSX data file example is as follows:

A	B	C	D	E	F	G	H	I	J	K	L	M	N	O	P	Q	R	S	T	U	V	W	X	Y	Z					
1	Well	Sample	Formulation	T (°C)	Z-Ave	Dia	D50	D10	Intensity	Pk 1 Mode	Dia	Pk 1 est. M.W.	Pk 1 Polydispersity	Pk 1 Mass	Pk 2 Mode	Dia	Pk 2 est. M.W.	Pk 2 Polydispersity	Pk 2 Mass	Pk 3 Mo	Pk 3 est	Pk 3 Poly	Pk 3 M	Date	Filter	Viscosity	RI	Derived Intensity	Min Pk Area	Min Rh
2	A1	Sample 1	Poly IgG, 250 mM Glycine	24.98187637	7.49	5.56	5.663	51317	5.59	5.12	22.58%	99.88%	53.02	-	-	Out of Range	178.81%	0.14%	-	-	-	-	-	B22 & kD	0.907	1.3334	21507.42	5.00	0.50	
3	B1	Sample 2	Poly IgG, 250 mM Glycine	24.98187637	71.90	5.13	0.069	112,635	9.61	139.38	57.50%	99.96%	127.69	-	-	Out of Range	94.59%	-	-	-	-	-	-	B22 & kD	0.907	1.3334	212548.4	5.00	0.50	
4	C1	Sample 3	Poly IgG, 250 mM Glycine	24.99149323	54.57	5.88	1.590	500,353	9.60	139.47	54.58%	99.87%	106.69	-	-	Out of Range	93.03%	0.13%	-	-	-	-	-	B22 & kD	0.907	1.3334	500230.7	5.00	0.50	
5	D1	Sample 4	Poly IgG, 250 mM Glycine	24.97225952	7.81	4.54	5.337	285,614	7.73	78.04	56.97%	100.00%	139.32	-	-	Out of Range	13.44%	-	-	-	-	-	-	B22 & kD	0.908	1.3334	285499.7	5.00	0.50	
6	E1	Sample 5	Poly IgG, 250 mM Glycine	24.98187637	7.61	5.16	5.459	545,759	7.86	94.72	51.80%	100.00%	-	-	-	-	-	-	-	-	-	-	-	B22 & kD	0.907	1.3334	245658.9	5.00	0.50	
7	F1	Sample 6	Poly IgG, 250 mM Glycine	24.99149323	6.67	4.30	0.415	130,546	6.46	64.42	51.06%	100.00%	-	-	-	-	-	-	-	-	-	-	-	B22 & kD	0.907	1.3334	330411.4	5.00	0.50	
8	G1	Sample 7	Poly IgG, 250 mM Glycine	24.99149323	6.07	3.39	0.313	280,466	5.57	43.78	51.58%	100.00%	-	-	-	-	-	-	-	-	-	-	-	B22 & kD	0.907	1.3334	448438.9	5.00	0.50	
9	H1	Sample 8	Poly IgG, 250 mM Glycine	24.97225952	5.47	3.82	0.260	245,893	5.04	56.04	29.67%	100.00%	-	-	-	-	-	-	-	-	-	-	-	B22 & kD	0.908	1.3334	393159.9	5.00	0.50	
9	I1	Sample 9	Poly IgG, 250 mM Glycine	24.98187637	6.62	5.12	0.210	514,024	4.46	20.21	51.24%	100.00%	534.47	-	-	-	-	-	-	-	-	-	-	B22 & kD	0.907	1.3334	502095	5.00	0.50	
11	J1	Sample 10	Poly IgG, 250 mM Glycine	24.99149323	6.10	1.93	0.221	504,538	5.96	16.67	13.92%	100.00%	233.90	-	-	Out of Range	13.01%	-	-	-	-	-	-	B22 & kD	0.907	1.3334	670601.6	5.00	0.50	
12	K1	Sample 11	Poly IgG, 250 mM Glycine	24.99149323	5.62	1.80	0.248	506,976	5.54	11.33	19.51%	100.00%	148.30	-	-	Out of Range	17.60%	-	-	-	-	-	-	B22 & kD	0.907	1.3334	671970.2	5.00	0.50	
13	L1	Sample 12	Poly IgG, 250 mM Glycine	24.98187637	5.43	4.42	0.171	505,733	5.30	5.33	26.17%	100.00%	182.80	-	-	Out of Range	26.28%	-	-	-	-	-	-	B22 & kD	0.907	1.3334	587251.7	5.00	0.50	
14	M1	Sample 13	Poly IgG, 250 mM Glycine	24.98187637	5.00	1.35	0.204	505,146	5.99	7.69	15.66%	100.00%	117.74	-	-	Out of Range	10.90%	-	-	-	-	-	-	B22 & kD	0.907	1.3334	671940.4	5.00	0.50	
15	N1	Sample 14	Poly IgG, 250 mM Glycine	25.00111008	2.96	0.57	0.037	527,617	5.62	5.24	54.18%	100.00%	184.73	-	-	Out of Range	51.15%	-	-	-	-	-	-	B22 & kD	0.907	1.3334	721422.3	5.00	0.50	
16	O1	Sample 15	Poly IgG, 250 mM Glycine	24.98187637	0.06	0.04	0.537	21,483	68.25	961.36	256.33%	100.00%	-	-	-	-	-	-	-	-	-	-	-	B22 & kD	0.907	1.3334	21474.25	5.00	0.50	
17	P1	Sample 16	Poly IgG, 250 mM Glycine	24.97225952	591.04	541.97	0.552	51,091	1,888.52	-	Out of Range	501.10%	100.00%	-	-	-	-	-	-	-	-	-	-	B22 & kD	0.908	1.3334	13078.34	5.00	0.50	

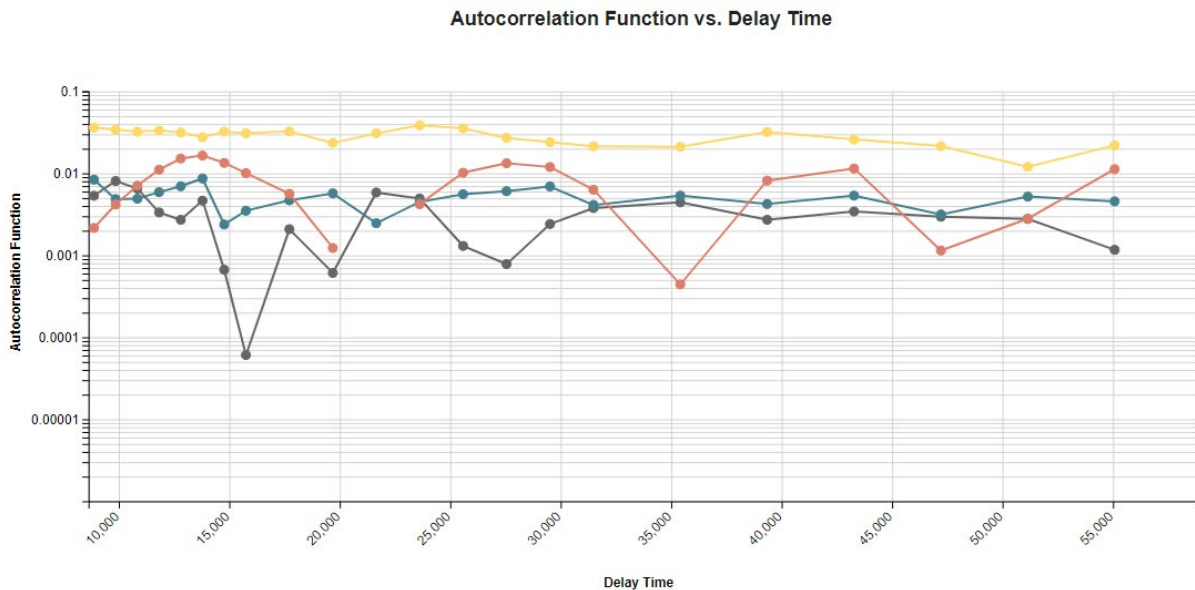
EXAMPLES

Included below is a sample dashboard from Uncle measurement files:

1. Column Chart plotting measurement results for Average Tm1 & Tm2 vs. Sample Name



2. Line Chart plotting raw data measurement results for Autocorrelation Function vs. Delay Time



3. Tabular Summary examples

Measurement Summary Table – Measurement Results

Sample Name	Well	Tm1 (°C) - AVG	Average Tm1 (°C) - AVG	%CV Tm1 - AVG	%CV Tagg473 - AVG	%CV Tagg266 - AVG
Sample 7	J1	67.90	67.90			
Sample A	A1	33.50	33.50		26.13	25.49
Sample AA	Multiple (3 Values)	62.80	62.80	0.48	0.26	0.13
Sample B	Multiple (2 Values)	68.00	68.00	1.18		10.98
Sample BB	Multiple (3 Values)	63.60	63.60	1.42	0.13	0.40
Sample C	Multiple (2 Values)	69.00	69.00			30.00
Sample CC	Multiple (3 Values)	53.70	53.70	15.83	0.68	1.10
Sample D	F1	68.80	68.80			
Sample DD	Multiple (2 Values)	47.55	47.60	0.63	0.14	0.28
Sample EE	Multiple (2 Values)	63.45	63.40	0.32	0.69	0.97
Sample F	G1	68.70	68.70			
Sample FF	Multiple (2 Values)		33.80		19.68	28.41

Measurement Summary Table – Instrument Settings

Sample Name	LINK Record ID #	InstrumentName	File Name	Well Row - AVG	Well Column - AVG	Well
Sample 7	106	UNcle	UNCLE Report	10.00	1.00	J1
Sample A	97	UNcle	UNCLE Report	1.00	1.00	A1
Sample AA		UNcle	UNCLE Report	3.00	3.00	Multiple (3 Values)
Sample B		UNcle	UNCLE Report	2.50	1.00	Multiple (2 Values)
Sample BB		UNcle	UNCLE Report	6.00	3.00	Multiple (3 Values)
Sample C		UNcle	UNCLE Report	4.50	1.00	Multiple (2 Values)
Sample CC		UNcle	UNCLE Report	9.00	3.00	Multiple (3 Values)
Sample D	102	UNcle	UNCLE Report	6.00	1.00	F1
Sample DD		UNcle	UNCLE Report	11.50	3.00	Multiple (2 Values)
Sample EE		UNcle	UNCLE Report	13.50	3.00	Multiple (2 Values)
Sample F	103	UNcle	UNCLE Report	7.00	1.00	G1
Sample FF		UNcle	UNCLE Report	15.50	3.00	Multiple (2 Values)
Sample G	104	UNcle	UNCLE Report	8.00	1.00	H1
Sample L	110	UNcle	UNCLE Report	14.00	1.00	N1

UNCLE 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

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