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Explore the future

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Particle Characterization

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Particle Classroom Series VI: Method Development

November 12, 2019

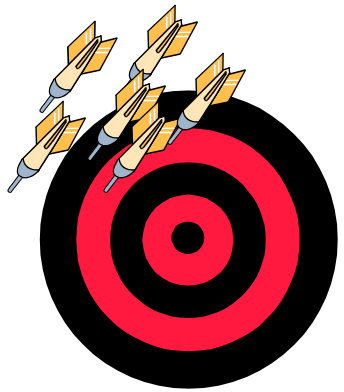
Overview

- **Goal: Reproducible method that tracks product performance**
- Choose measurement approach (dry vs. suspension)
- Lock down RI
- Vary measurement settings that can influence result
  - Dry: measurement duration, concentration, air pressure
  - Wet: sampler selection, dispersion, duration, concentration, energy (mixing + ultrasound)
- Test method (reproducibility)
  - Meet ISO, USP or internal guidelines
  - Check COV at d10, d50, d90

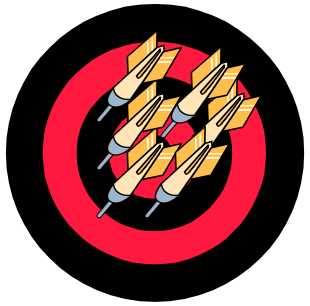
Goals

- Reproducible method that tracks product performance
- You might have other goals
  - Accuracy: tricky subject, is it the “real” particle size?
  - Repeatability: liquid suspension re-circulating in sampler
  - Reproducibility: prepare, measure, empty, repeat
  - Resolution: optimize to find second populations
  - Match historic data (sieves), but quicker, easier technique
- Use structured approach for any decision/choice that may influence result
- Have data to support selections made
- Document process so colleagues understand your choices

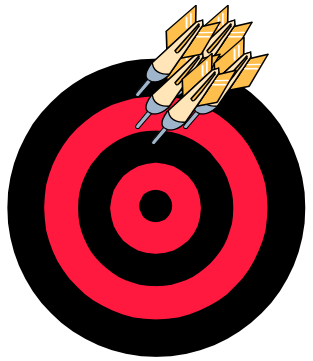
Accuracy vs. Precision



LOW ACCURACY
LOW PRECISION



HIGH ACCURACY
LOW PRECISION



LOW ACCURACY
HIGH PRECISION

HIGH ACCURACY
HIGH PRECISION

(A) Low accuracy, low precision measurements form a diffuse, off-center cluster
(B) Low accuracy, high precision measurements form a tight off-center cluster
(C) High accuracy, low precision measurements form a cluster that is evenly distributed but distant from the center of the target
(D) High Accuracy, high precision measurements are clustered in the center of the target.

Accuracy

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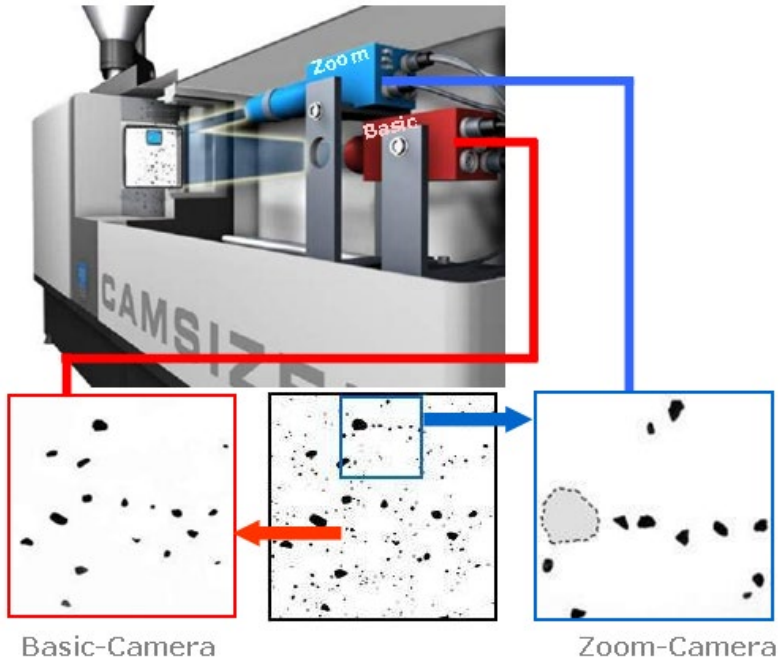
Is it the "real particle size"?

- Comparison to referee technique
- Microscope (image analysis) is referee technique for particle characterization
- Two kinds of image analysis:
  - Dynamic image analysis; particles flowing
  - Static image analysis; particles sit on slide on automated stage

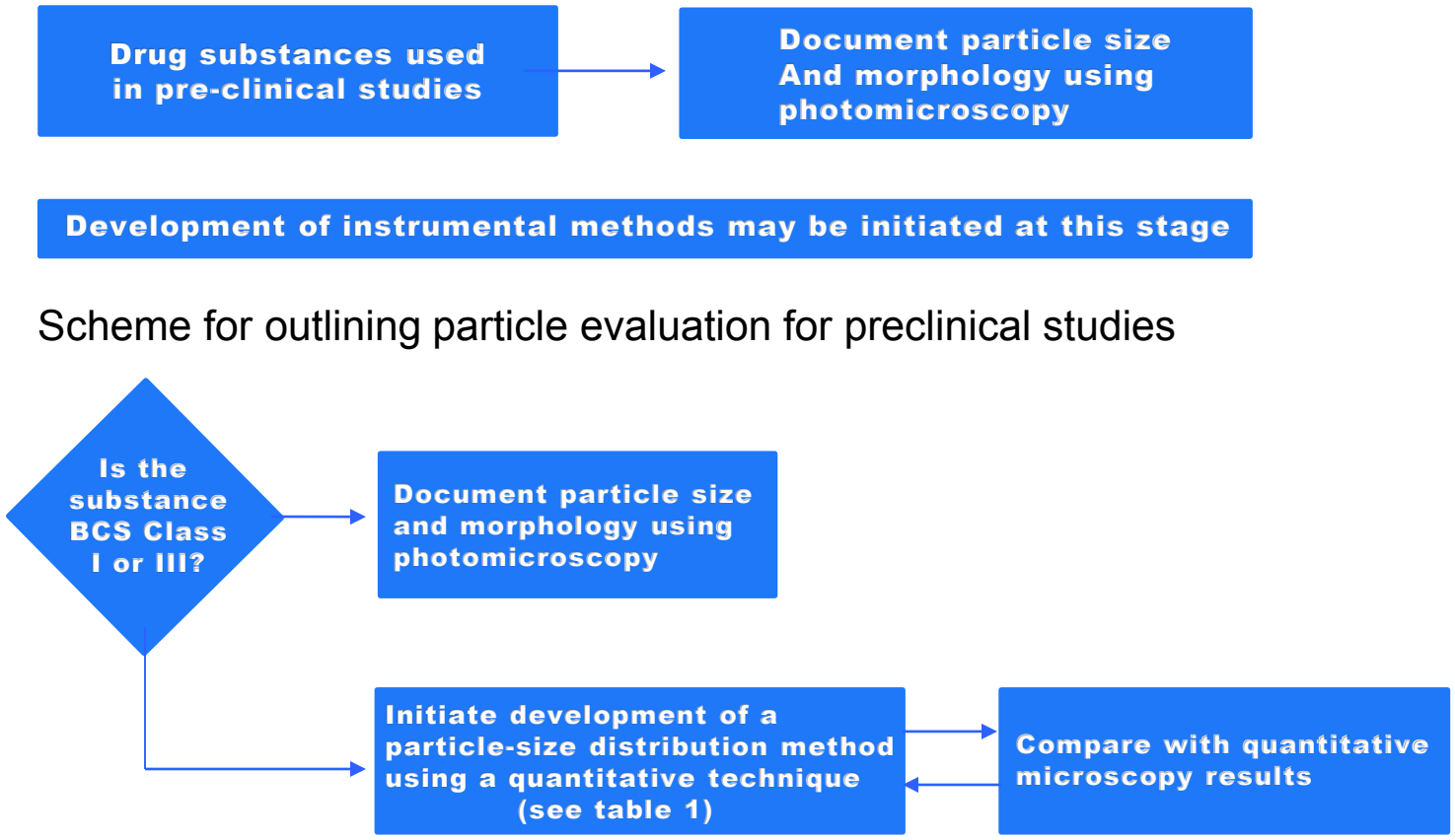
Image Analysis

Dynamic:
particles flow past camera



Static:
particles fixed on slide,
stage moves slide

Guidelines



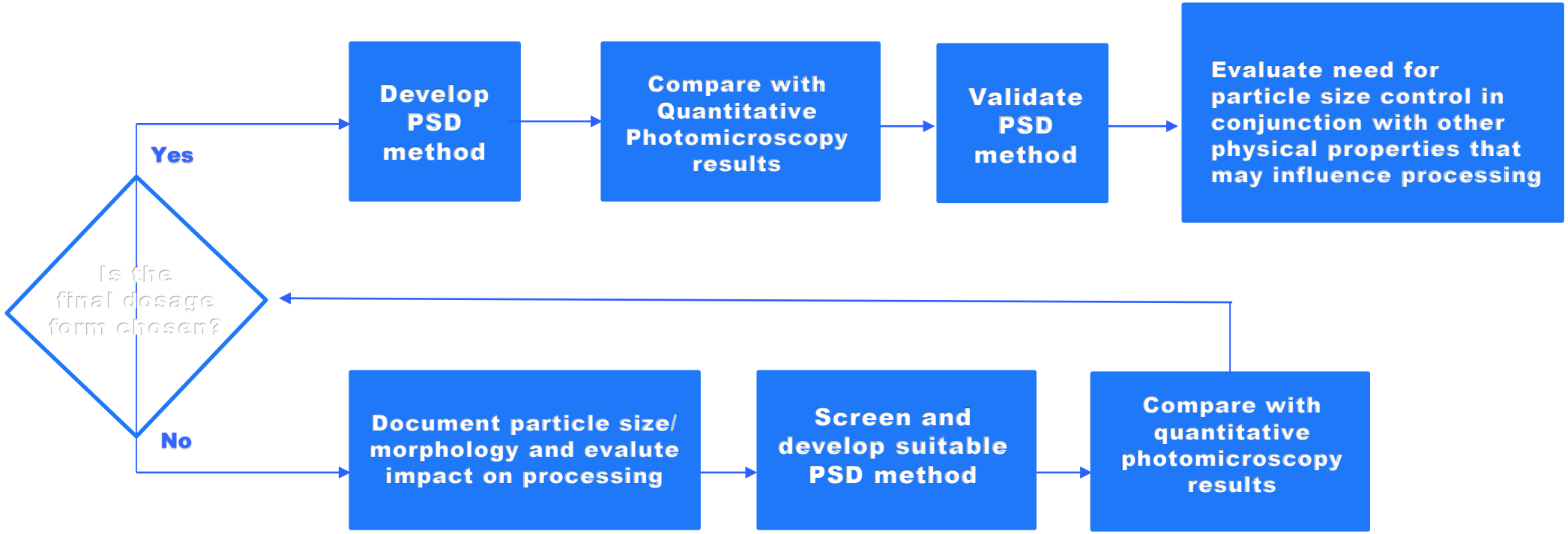
Scheme for outlining particle evaluation for preclinical studies

Decision tree outlining particle evaluation for Phase I clinical studies

If the drug is poorly soluble, you need to pay attention to particle analysis.

Compare with quantitative microscopy.

Guidelines Phase III



Compare Validate Document

Types of Precision

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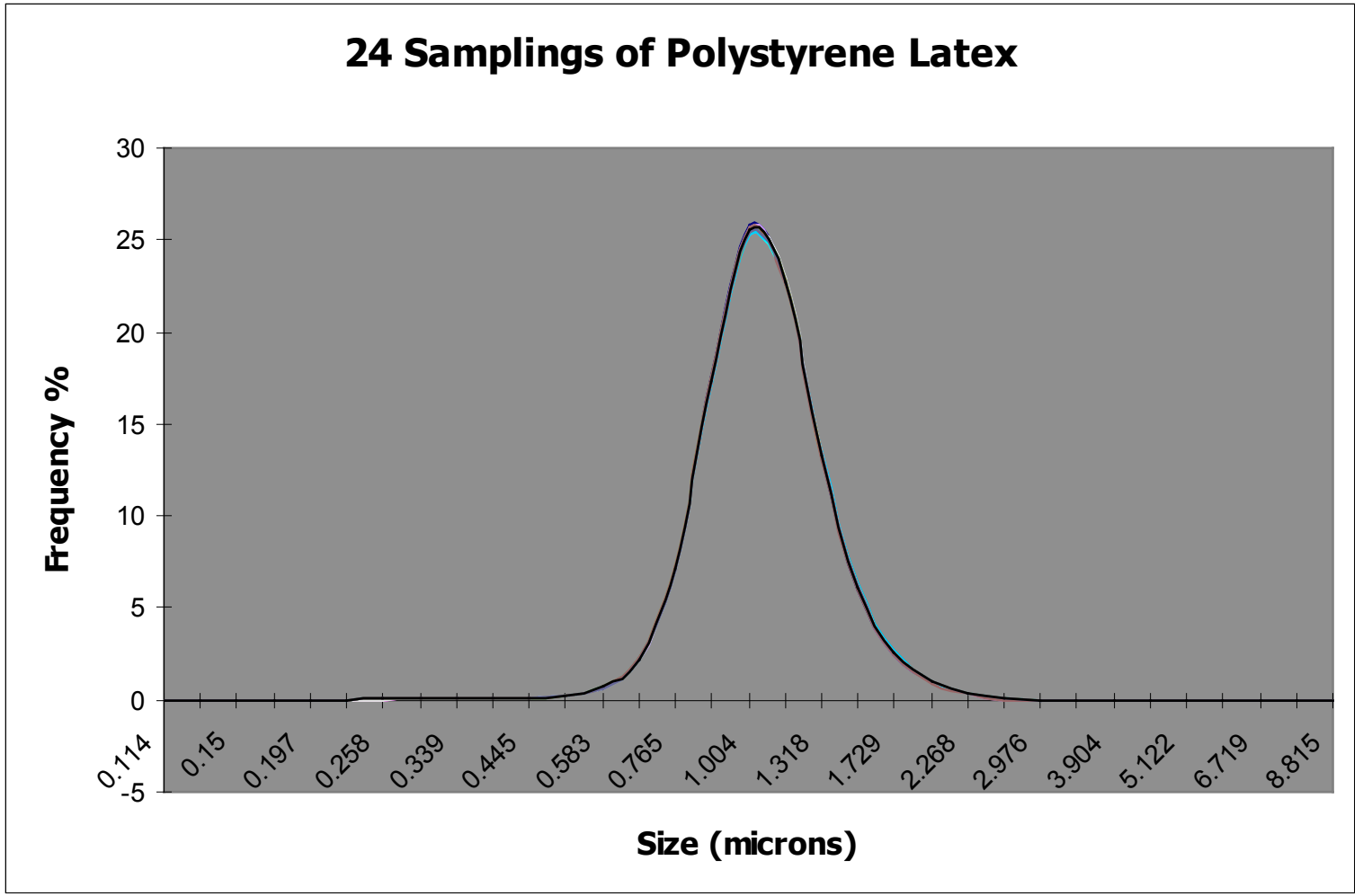
Repeatability

- Prepare sample, add to wet sampler, re-circulate, measure same multiple times (suspensions only)
- Provides limited information (mostly a test of analyzer performance)

Reproducibility

- Prepare sample, measure, drain, repeat (suspensions + dry)
- Distinguishes great methods

Reproducibility



Yes, we emptied instrument between each analysis.

Reproducibility

Reproducibility: prepare, measure, empty, repeat

What would be good reproducibility?

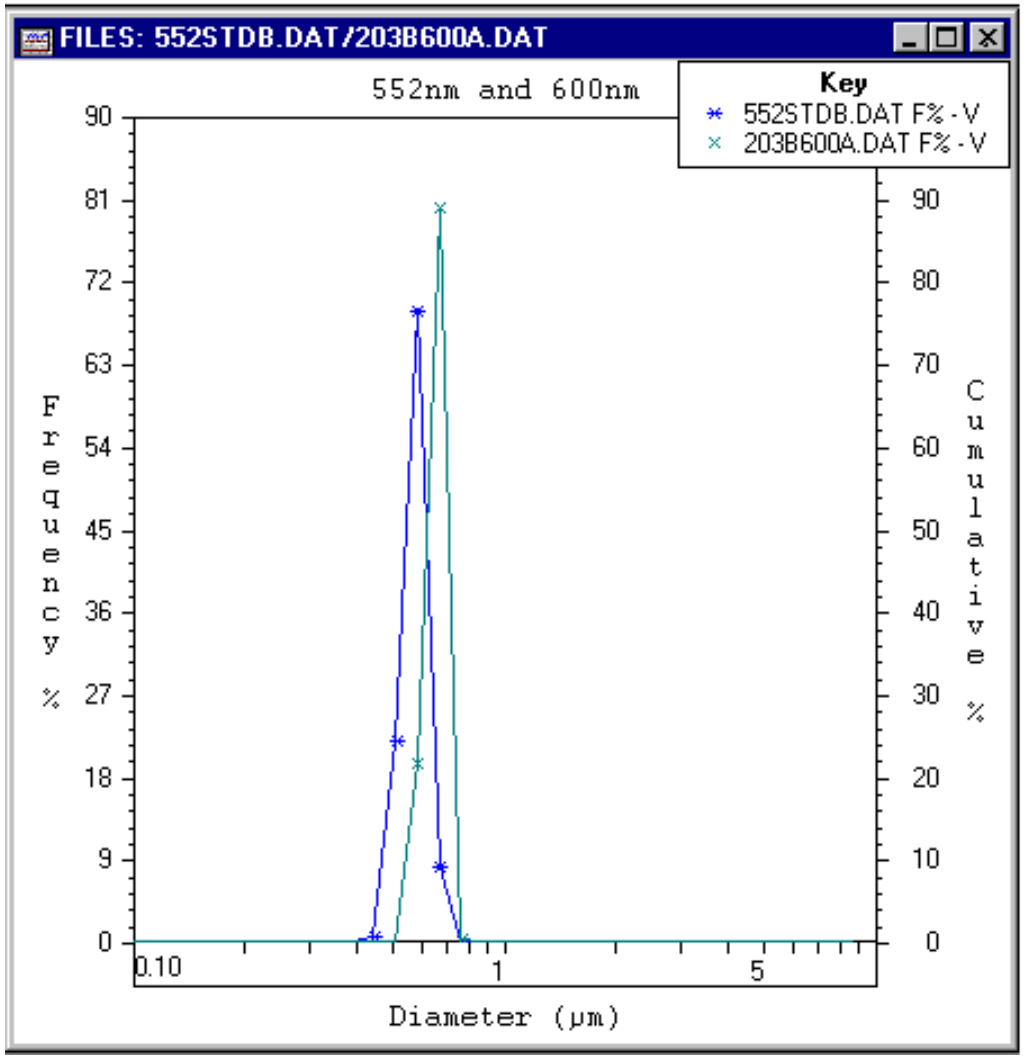
- Look at accepted standards
- Measure 3 times, calculated COV at d_{10}, d_{50}, d_{90}
  - COV (RSD) = st dev/mean * 100
- ISO13320
  - COV < 3% at median d_{50}
  - COV < 5% at d_{10} & d_{90}
- USP<429>
  - COV < 10% at median d_{50}
  - COV < 15% at d_{10} & d_{90}

Note: double all limits when d_{10}, d_{50}, or d_{90} < 10 microns

Resolution

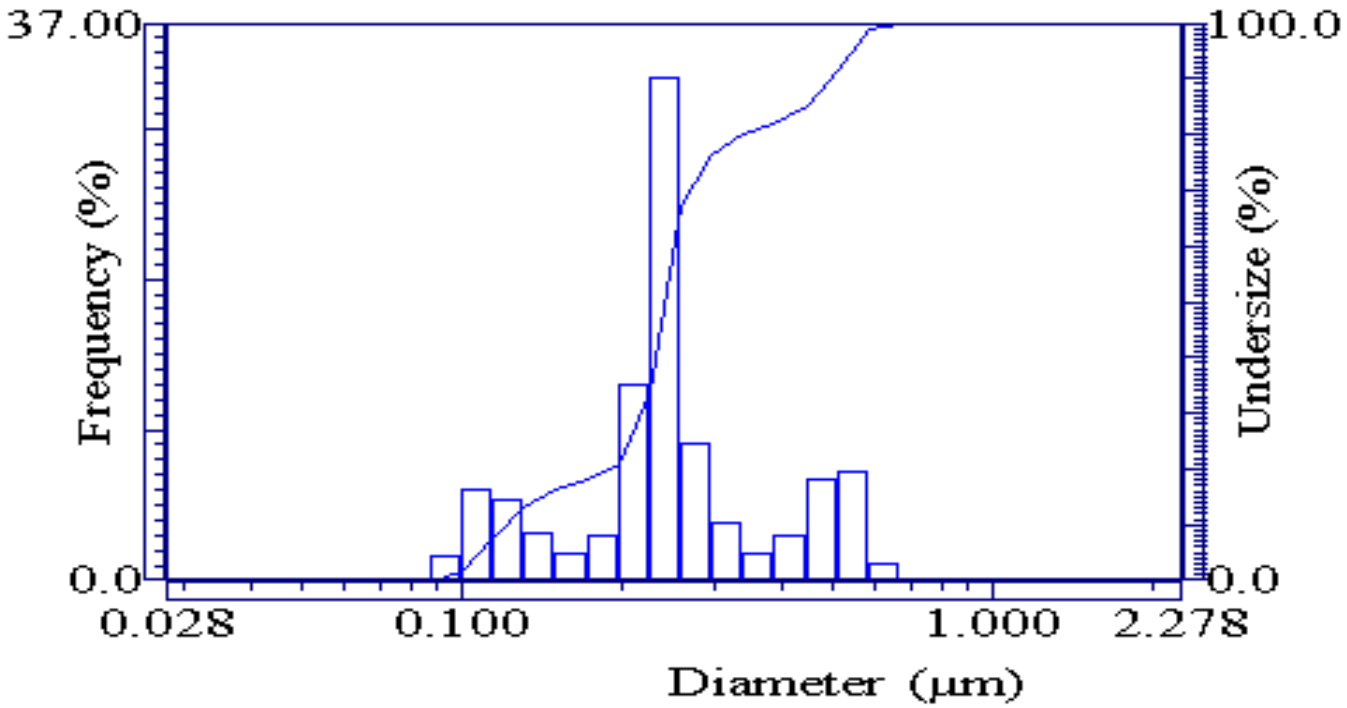
- Ability to measure small differences in particle size.
- Small differences between successive samples (different production lots) are most important. This is tied to reproducibility.
- Detection limit of small amount of material outside of main size distribution.
- Best defined by user's real-world requirements.

High Resolution?



- Resolve size difference between two materials of similar size
- 552nm and 600nm PSL
- Measured separately: high resolution
- Measure mixed together: peaks would blend and you cannot discern two populations.

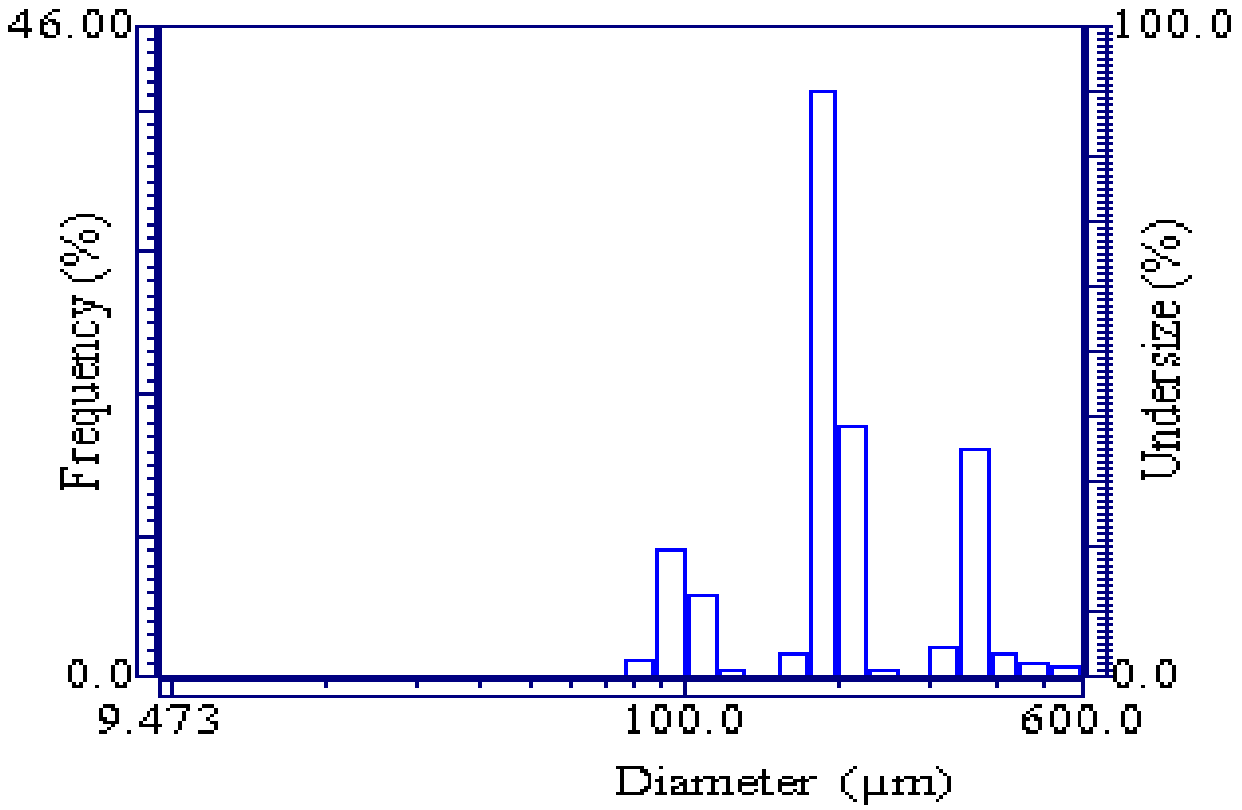
Separate peaks



83nm,
204 nm,
503nm
PSL

Resolution of multiple modes in a single sample
Next peak at least 2x of previous size

Separate Peaks

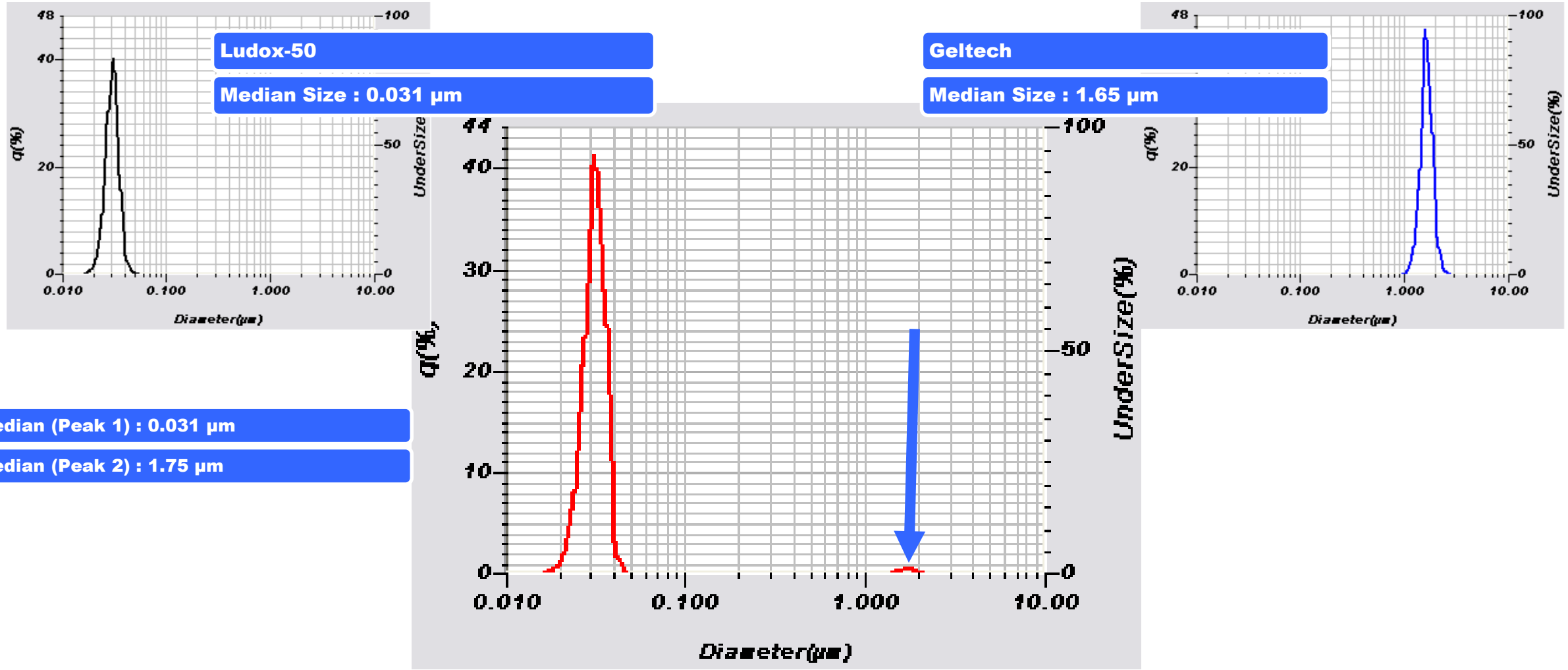


100µm,
200µm,
400µm
glass beads

Next peak 2x of previous size

Resolution is independent of where you are on size scale

Identifying Trace Impurities



0.05wt% Geltech 1.5 in LUDOX

Wet Method Development

Workflow

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First determine RI

Choose solvent (water, surfactants, hexane, etc.)

Sampler selection: sample volume

Pump & stirrer settings

Concentration

Measurement duration

Does the sample need ultrasound?

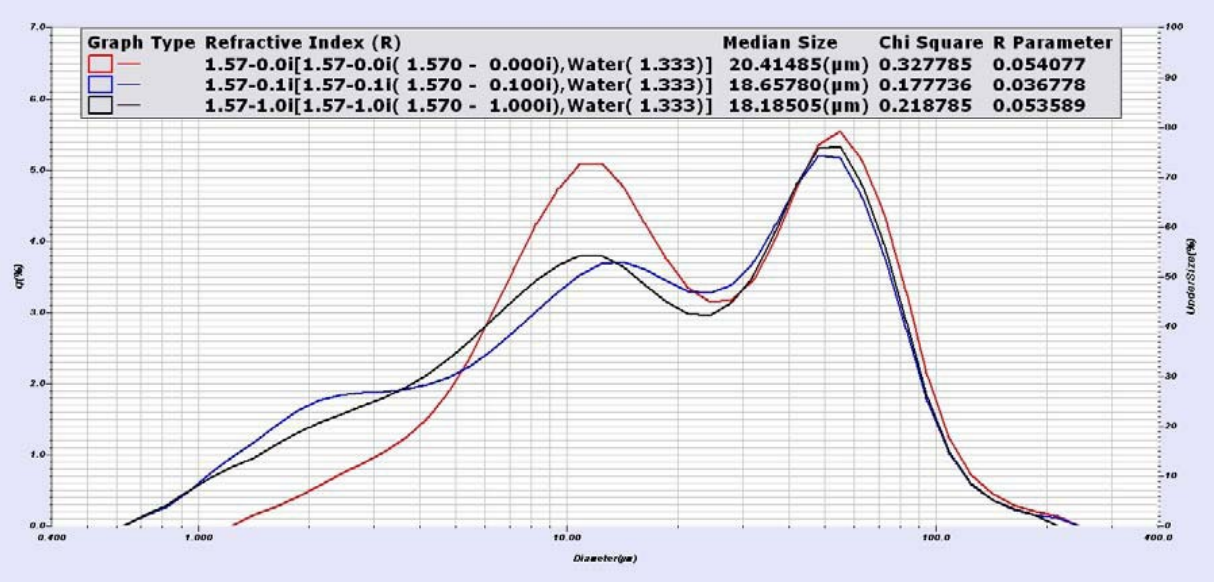
- Document size-time plot
- Disperse sample, but don’t break particles
- Check for reproducibility

Refractive Index

Real component via literature or web search, Becke line, etc.

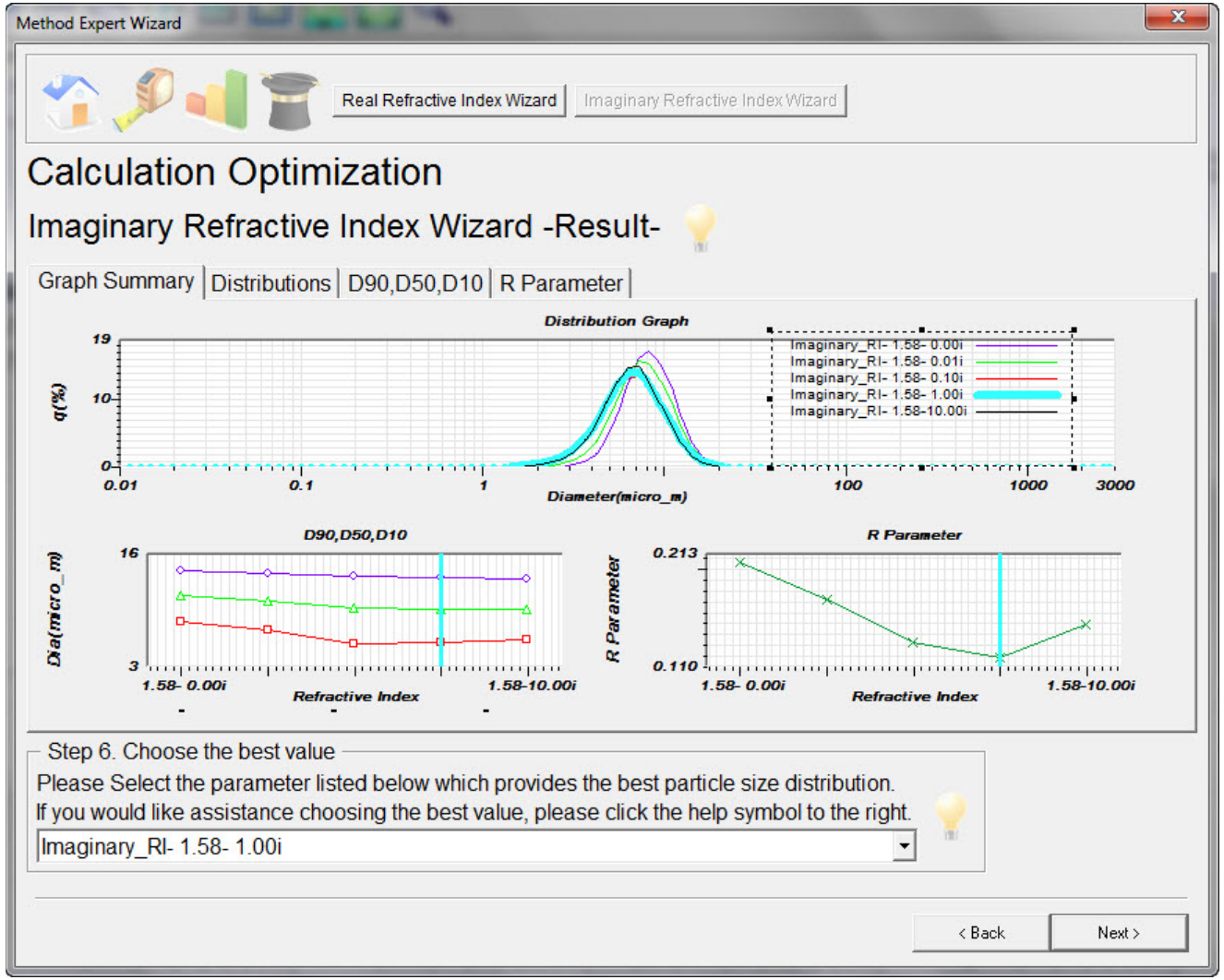
Measure sample, vary imaginary component to see if/how results change

Recalculate using different imaginary components, choose value that minimizes R parameter error calculation



[More in the refractive index webinar at : https://www.horiba.com/en_en/products/by-segment/scientific/particle-characterization/particle-analysis-webinar-series/](https://www.horiba.com/en_en/products/by-segment/scientific/particle-characterization/particle-analysis-webinar-series/)

LA-960 Method Expert

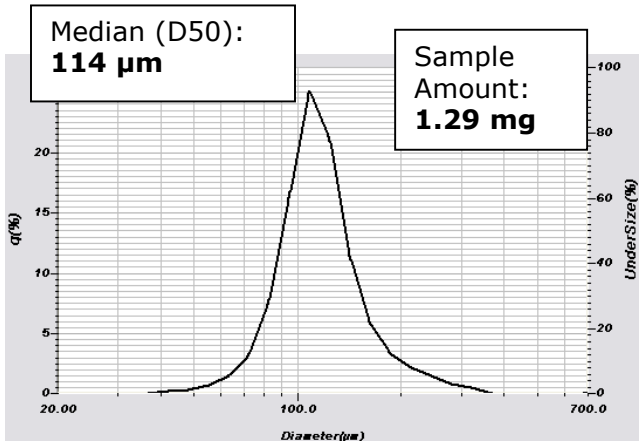


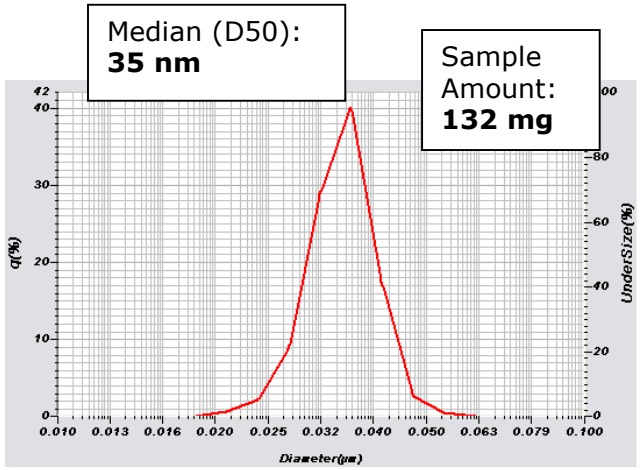
Sample Handling

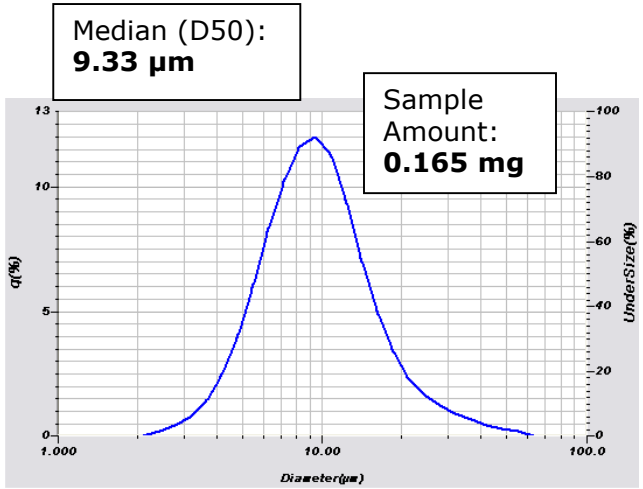
Larger, broad distributions require larger sample volume

Lower volume samplers for precious materials or solvents

LA-960 Sample Handlers	Dispersing Volume (mL)
Aqua/SolvoFlow	**180 - 330**
MiniFlow	**35 - 50**
Fraction Cell	**15**
Small Volume Fraction Cell	**10**



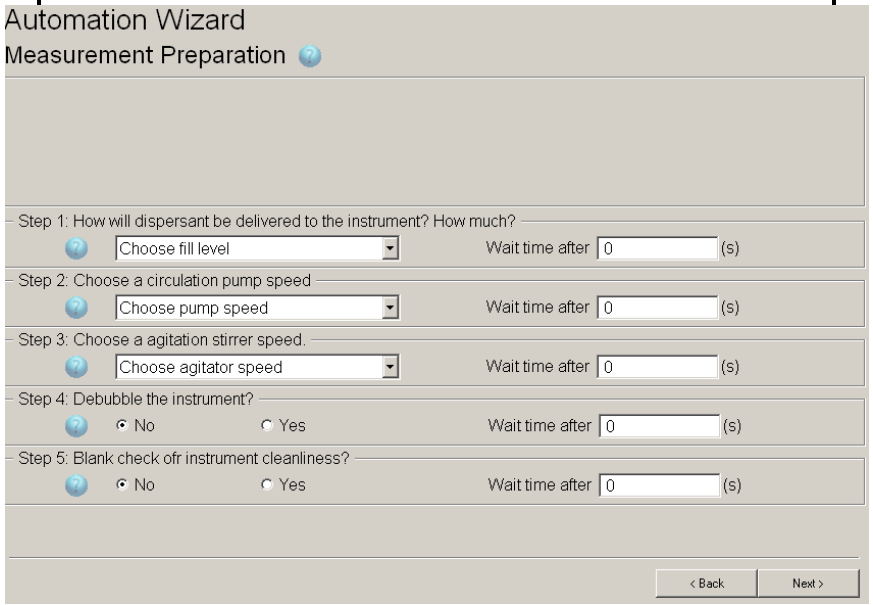




Pump and Stir

- Must be high enough to suspend & circulate heavy particles-if too low speed, larger particles won't be measured and apparent size decreases with time.
- Not so high that bubbles are introduced
- Adding energy – can disperse loose agglomerates
- Measure at several settings & select optimum (repeatability)
- Can be automated in software

Exp #	Agitation	Circulation	$D_{mean\ (nm)}$	D_{10} (nm)	D_{90} (nm)
1	1	1	187.03	137.5	245.7
2	1	3	184.23	135.9	242.1
3	3	1	187.28	137.8	245.8
4	3	3	184.61	136.1	242.5
5	1	1	185.32	136.3	243.7
6	1	3	184.04	135.8	241.8
7	3	1	184.13	135.8	241.9
8	3	3	184.98	136.4	242.9



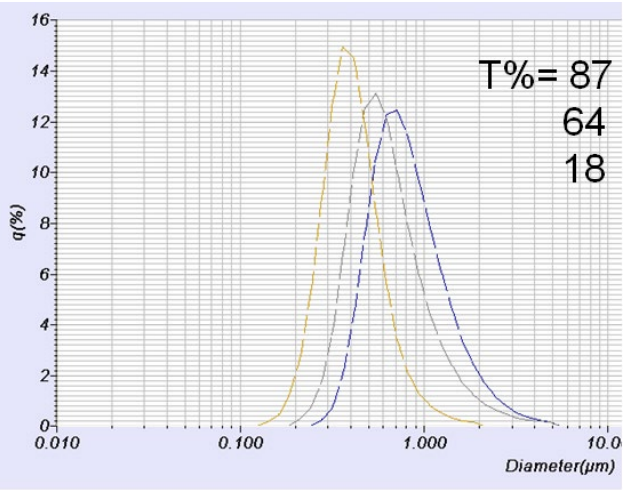
Concentration

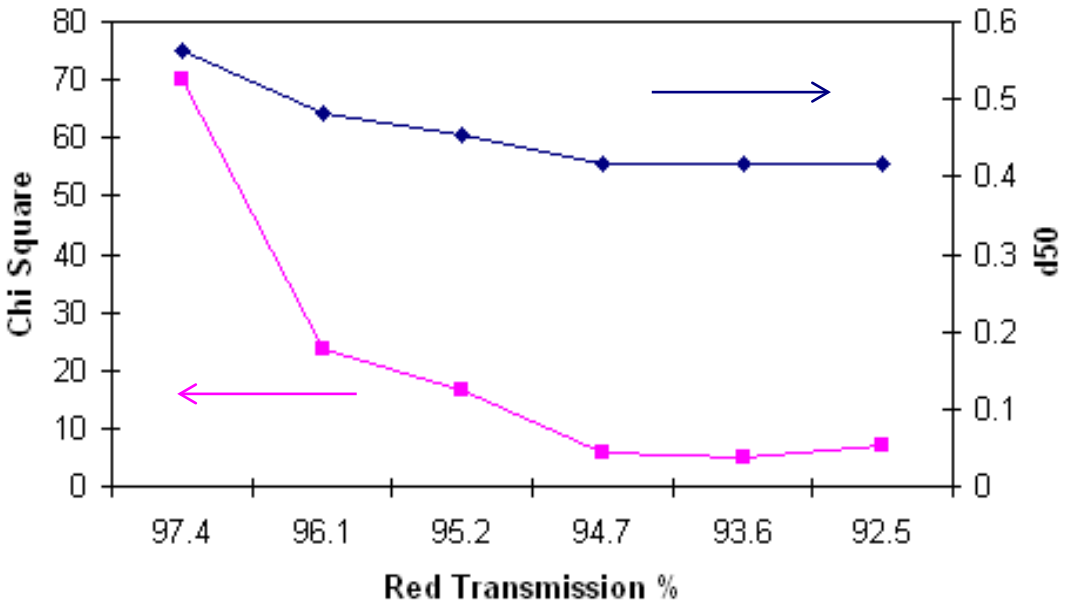
High enough for good S/N ratio

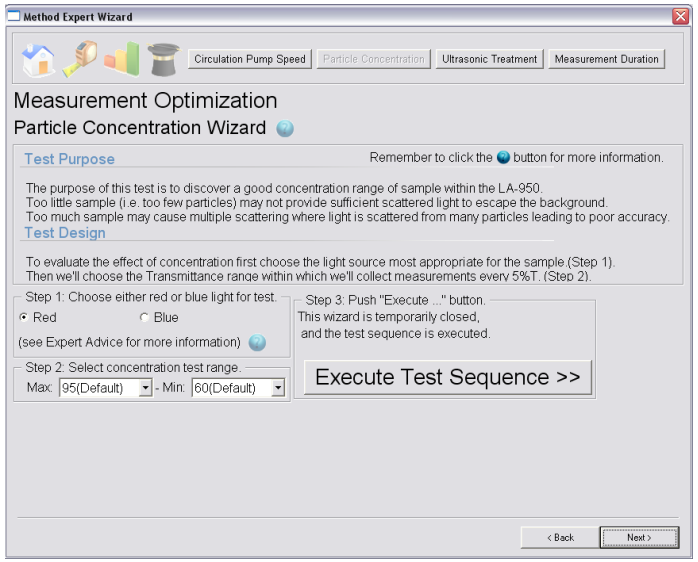
Low enough to avoid multiple scattering

Typically 95 – 80 %T

Measure at different T%, look at Chi Square calculation

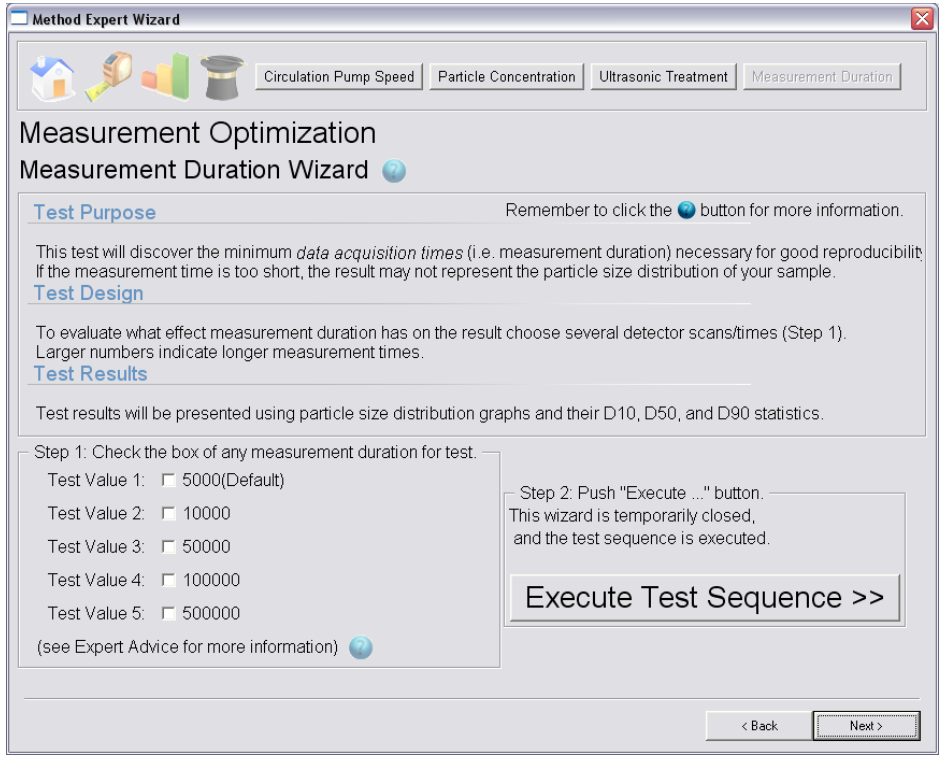






Duration

- Long enough for reproducibility
- Typically 5 sec, up to several minutes
- Longer time for large, broad distributions
- Can be automated in software
- Could be used for robustness testing during method validation



Ultrasonic Dispersion

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Adding energy to break up agglomerates – disperse to primary particles, without breaking particles

Similar to changing air pressure on dry powder feeder

Typically set to 100% energy, vary time (sec) on

Investigate tails of distribution

- High end to see if agglomerates removed
- Small end to see if new, smaller particles appear (breakage)

Test reproducibility, consider robustness

Note:

- Do not use on emulsions
- Can cause thermal mixing trouble w/solvents - wait
- Use external probe if t> 2-5 minutes

More in the sampling and dispersion webinar at :
https://www.horiba.com/en_en/products/by-segment/scientific/particle-characterization/particle-analysis-webinar-series/

Automation

LA-960 Method Expert will systematically vary:

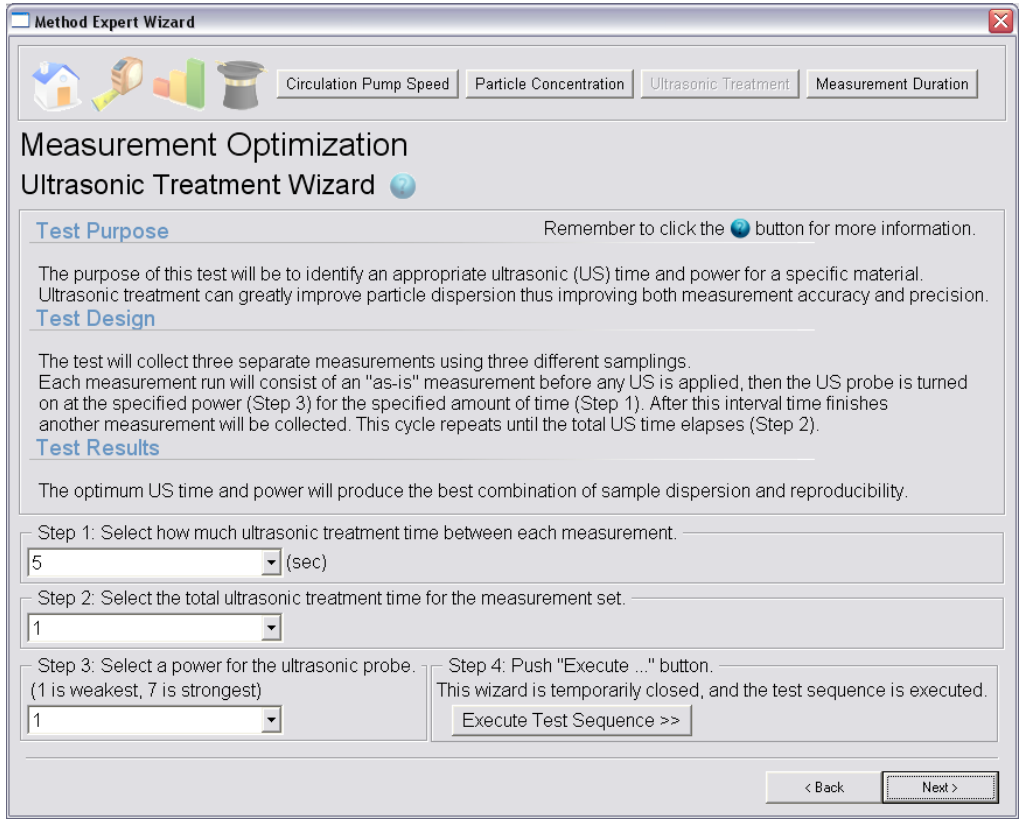
Level (power)

Time on

Iterations

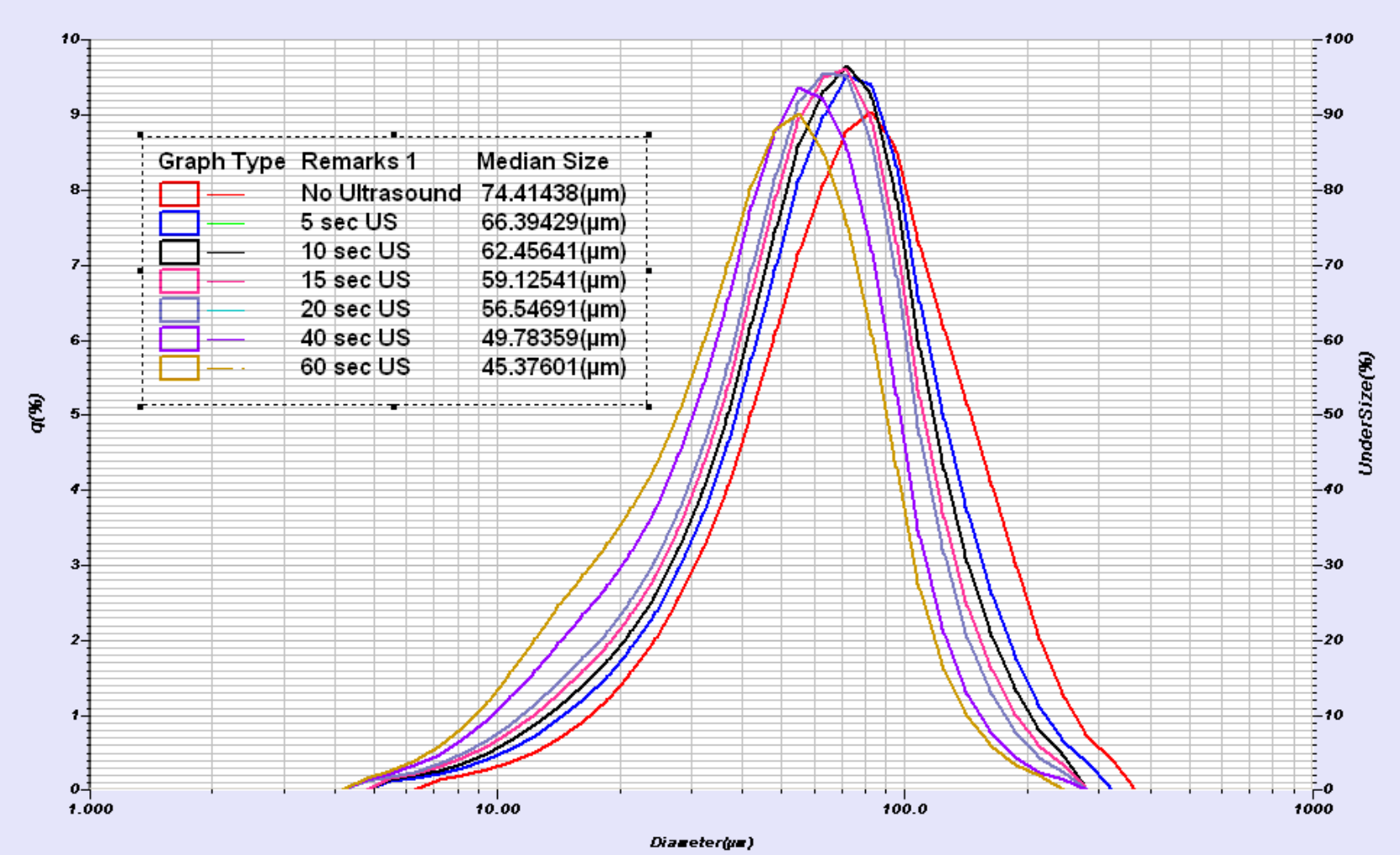
Delay

Generate result graphs

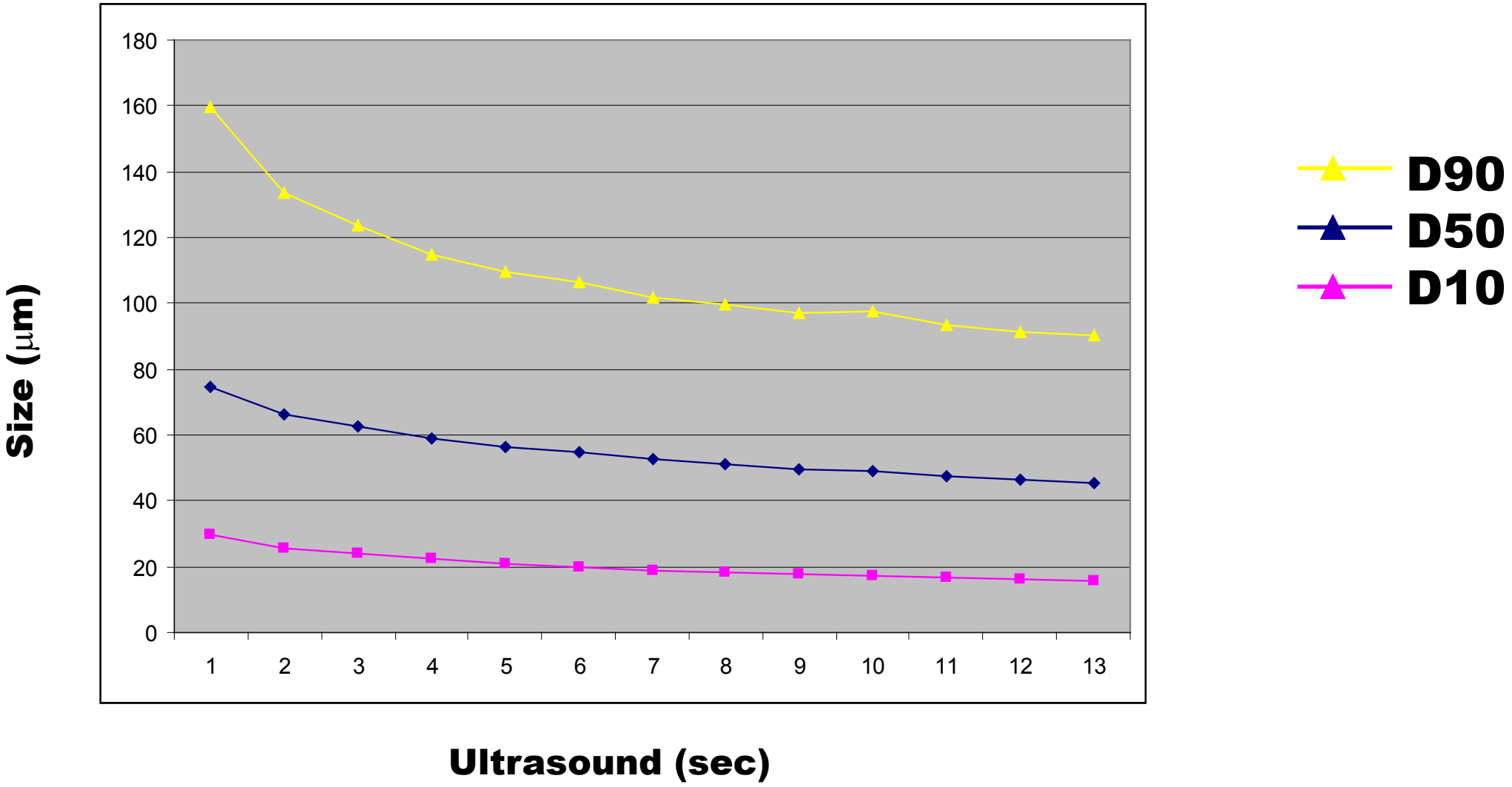


Examples

Ultrasound

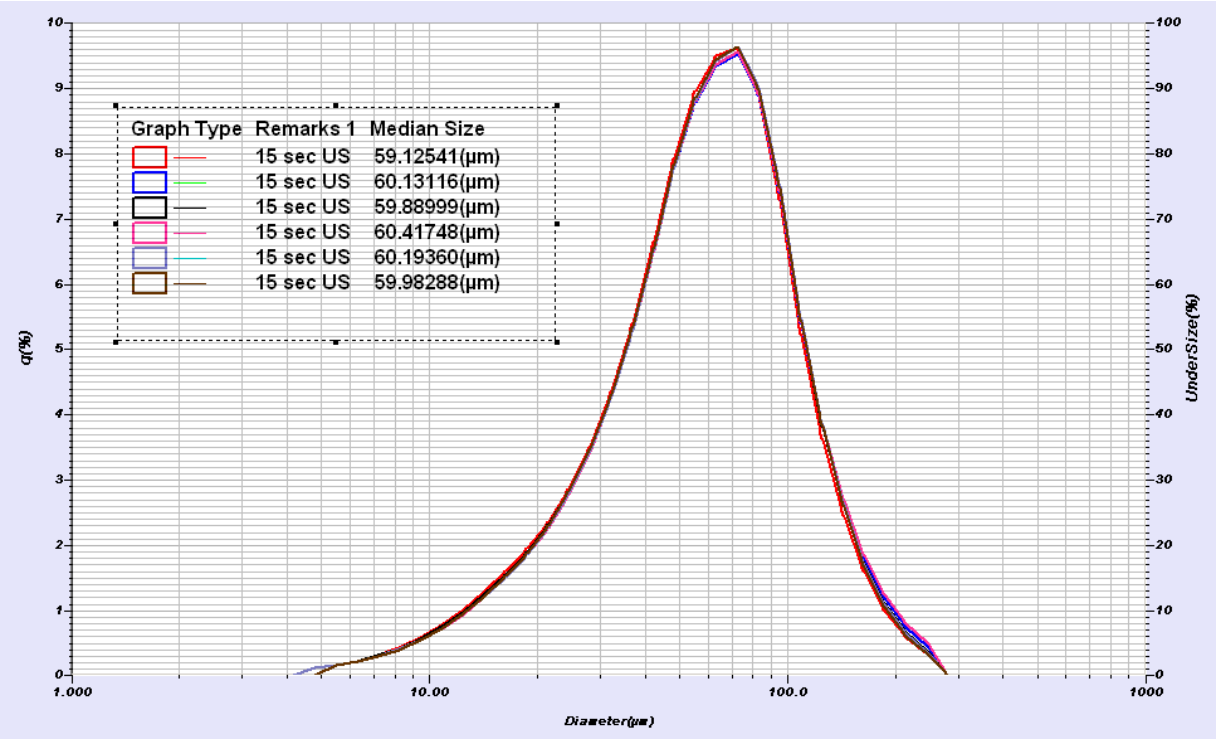


Plot summary data



Reproducibility

- **ISO13320**
  - **COV < 3% at median d_{50}**
  - **COV < 5% at d_{10} & d_{90}**



- **USP<429>**
  - **COV < 10% at median d_{50}**
  - **COV < 15% at d_{10} & d_{90}**

Data Name	Sample Name	D50	D10	D90	Comments
200707161438232.NGB	Avicel PH-101	59.125	22.221	114.93	15 sec US
200707161500243.NGB	Avicel PH-101	60.131	22.662	119.116	15 sec US
200707161507246.NGB	Avicel PH-101	59.89	22.392	117.452	15 sec US
200707161516249.NGB	Avicel PH-101	60.417	22.955	119.974	15 sec US
200707161523252.NGB	Avicel PH-101	60.194	22.765	117.289	15 sec US
200707161531255.NGB	Avicel PH-101	59.983	22.829	116.586	15 sec US
Mean		59.95667	22.63733	117.5578	
Standard Deviation		0.446191	0.278596	1.799955	
COV (st dev/mean)*100		0.74419	1.230691	1.531123	

Reproducibility

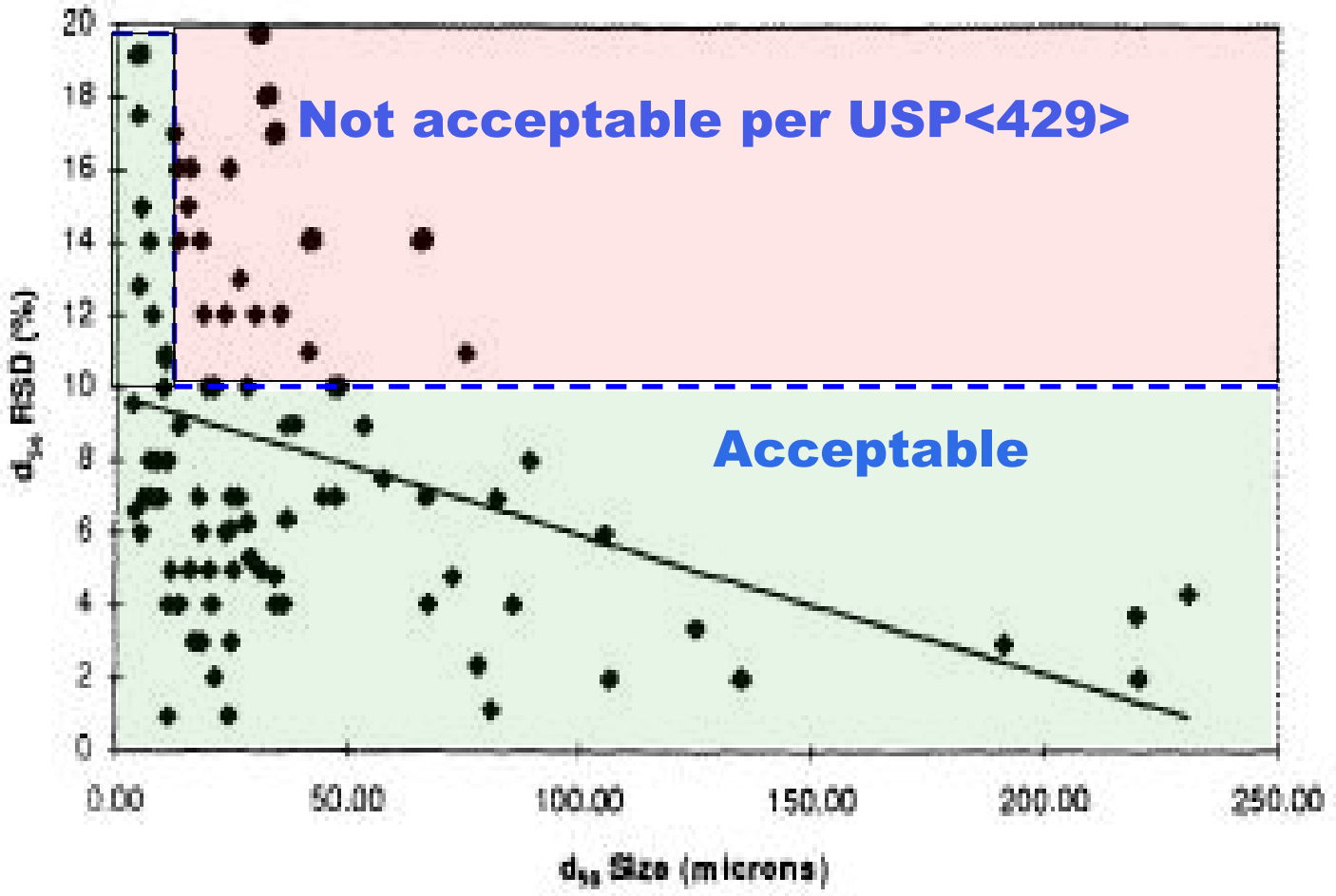
58 methods

Image analysis for morphology

Laser diffraction for PSD

If RSD for d50 < 20%, then acceptable for QC environment

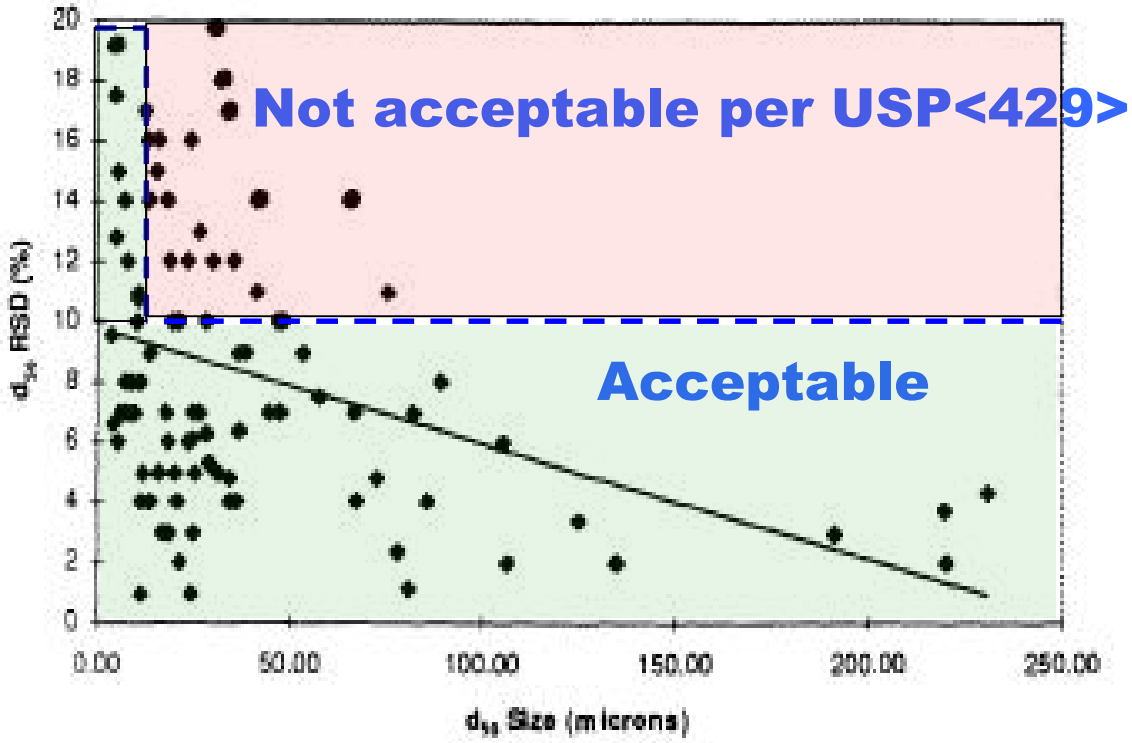
Note: RSD increases with decreasing size

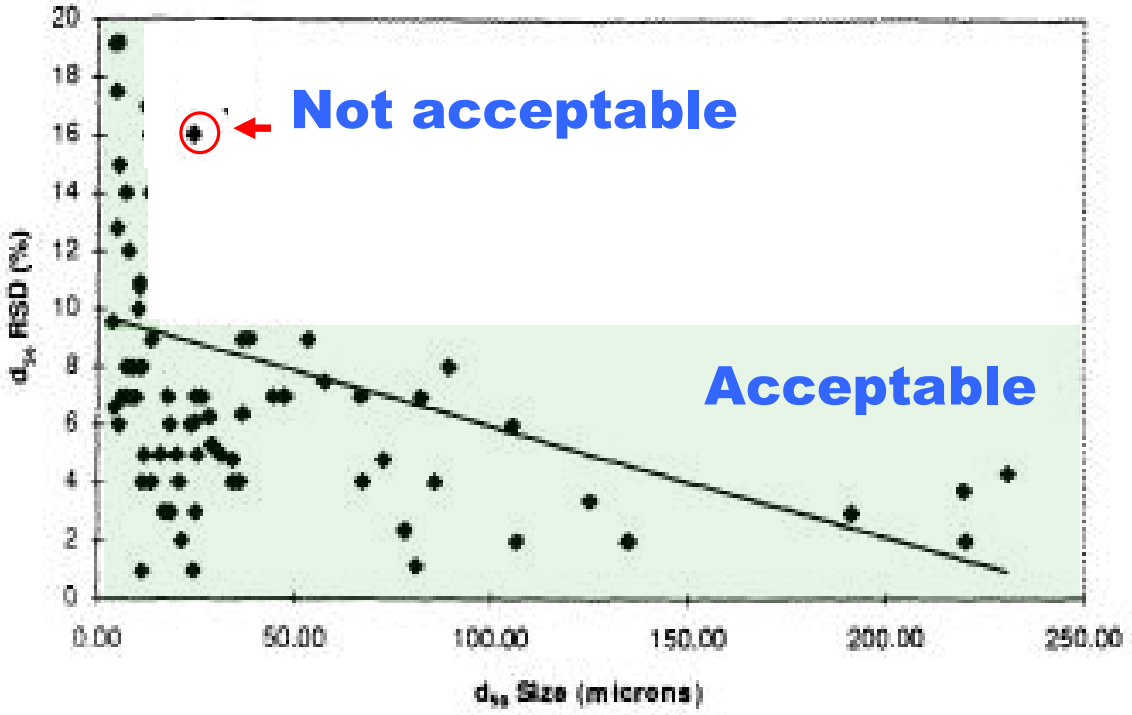


*Barber, Keuter, and Kravig, A Logical Stepwise Approach to Laser Diffraction Particle Size Distribution Analysis Methods Development and Validation Pharmaceutical Development and Technology, 3(2), 153-161 (1998)

Sampler Selection

Remove points from not acceptable region using Fraction Cell





Systematic analysis gives good hint about fraction cell use and importance of pumping for larger particles.

Dry Method Development

Workflow

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First get sampling right & determine RI

Measure at 3 different pressures (low, medium, high)

Determine optimum pressure based on good dispersion while not breaking particles

Can also compare dry vs. wet measurements

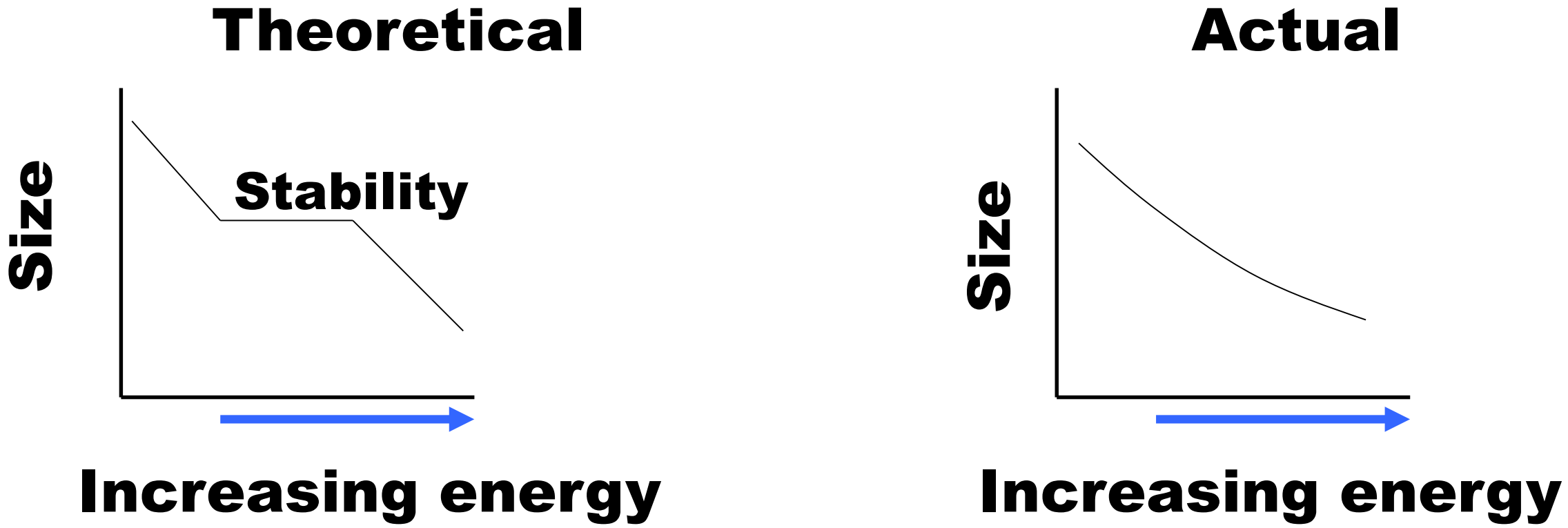
Adjust other settings to optimize sample concentration & duration

Ideally measure all of powder placed into the sampler

- Segregation can occur on vibrating tray
- Constant mass flow rate important for stable concentration during measurement

Once settings chosen, test reproducibility

Dispersion vs Breakage

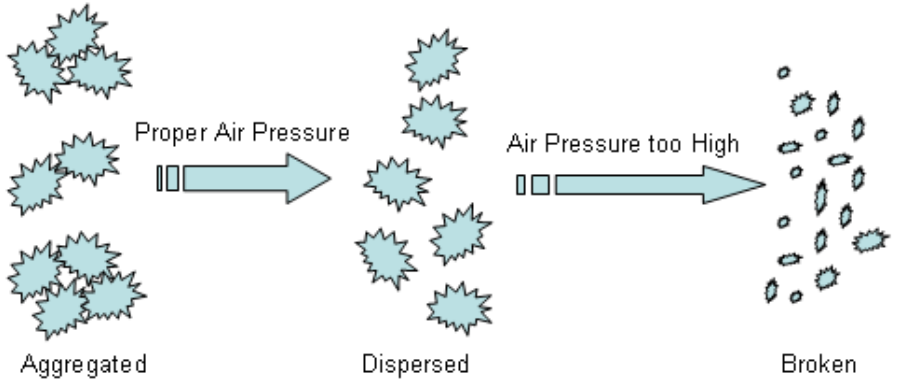


Higher air pressure or longer ultrasound duration

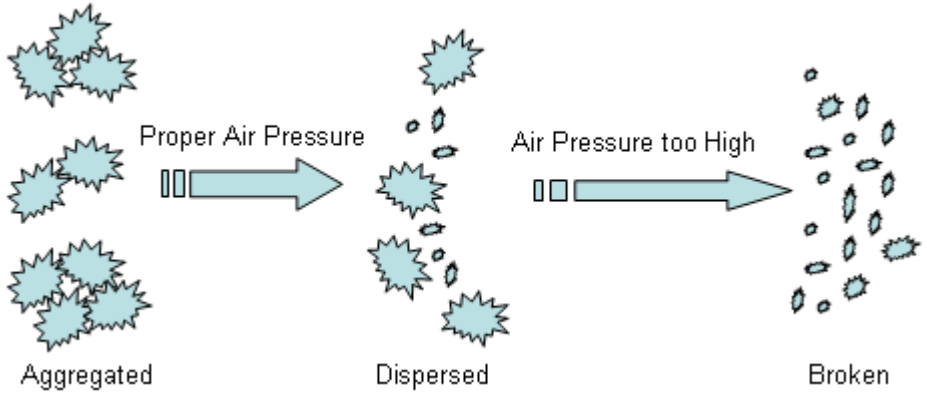
Dispersion vs Breakage

- **Dispersion and milling can be parallel rather than sequential processes**

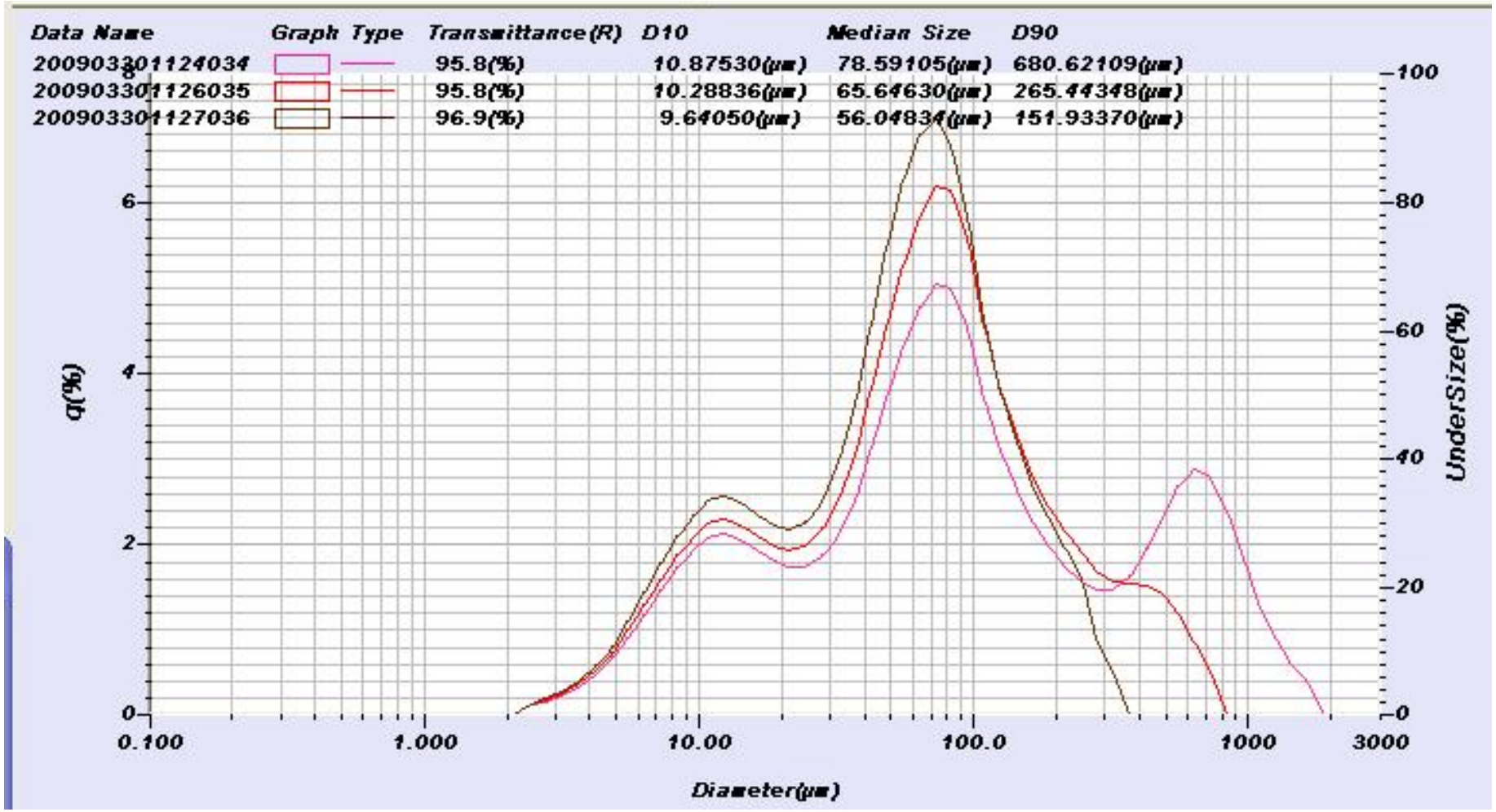
Theoretical



Actual

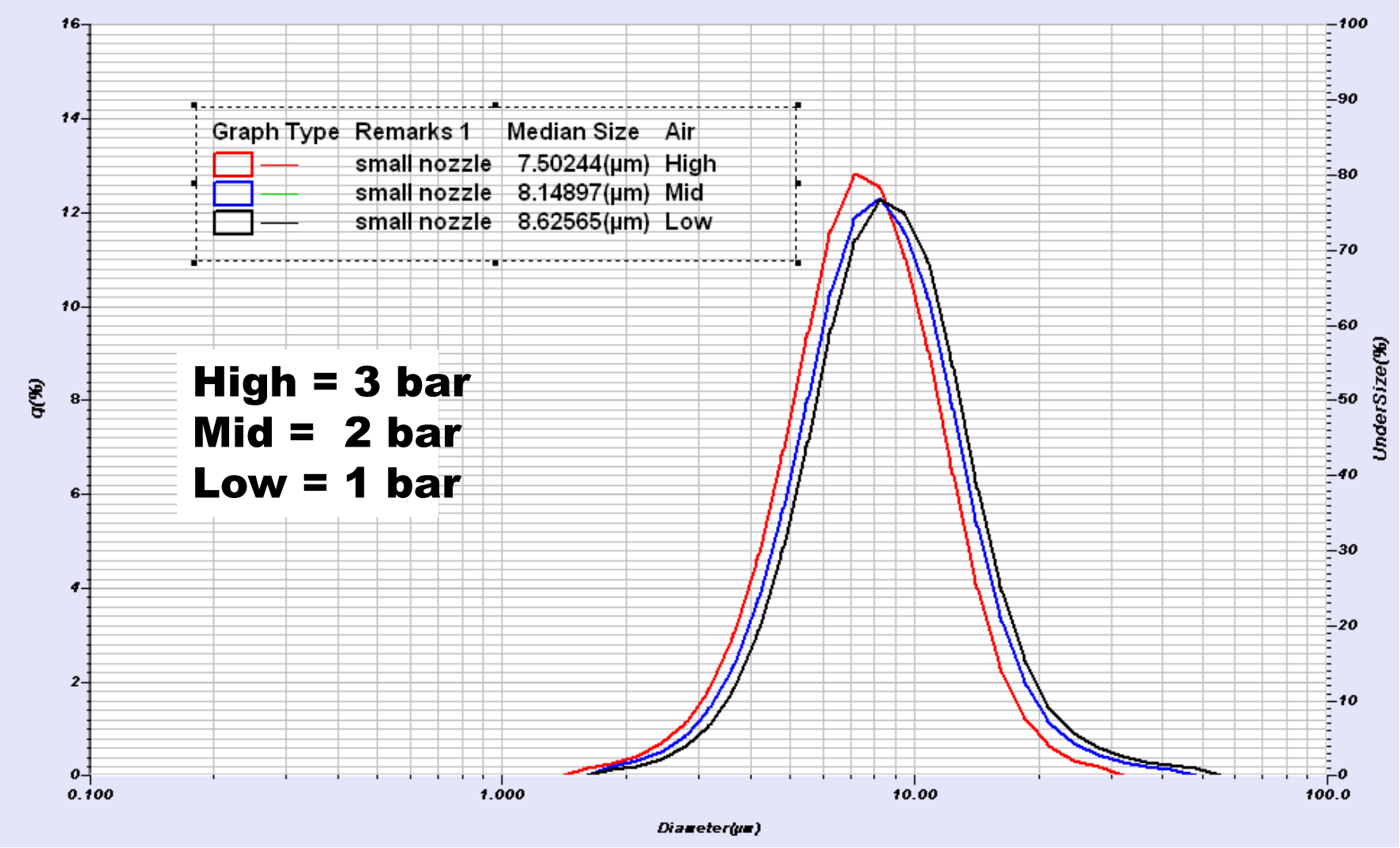


Pressure Titration

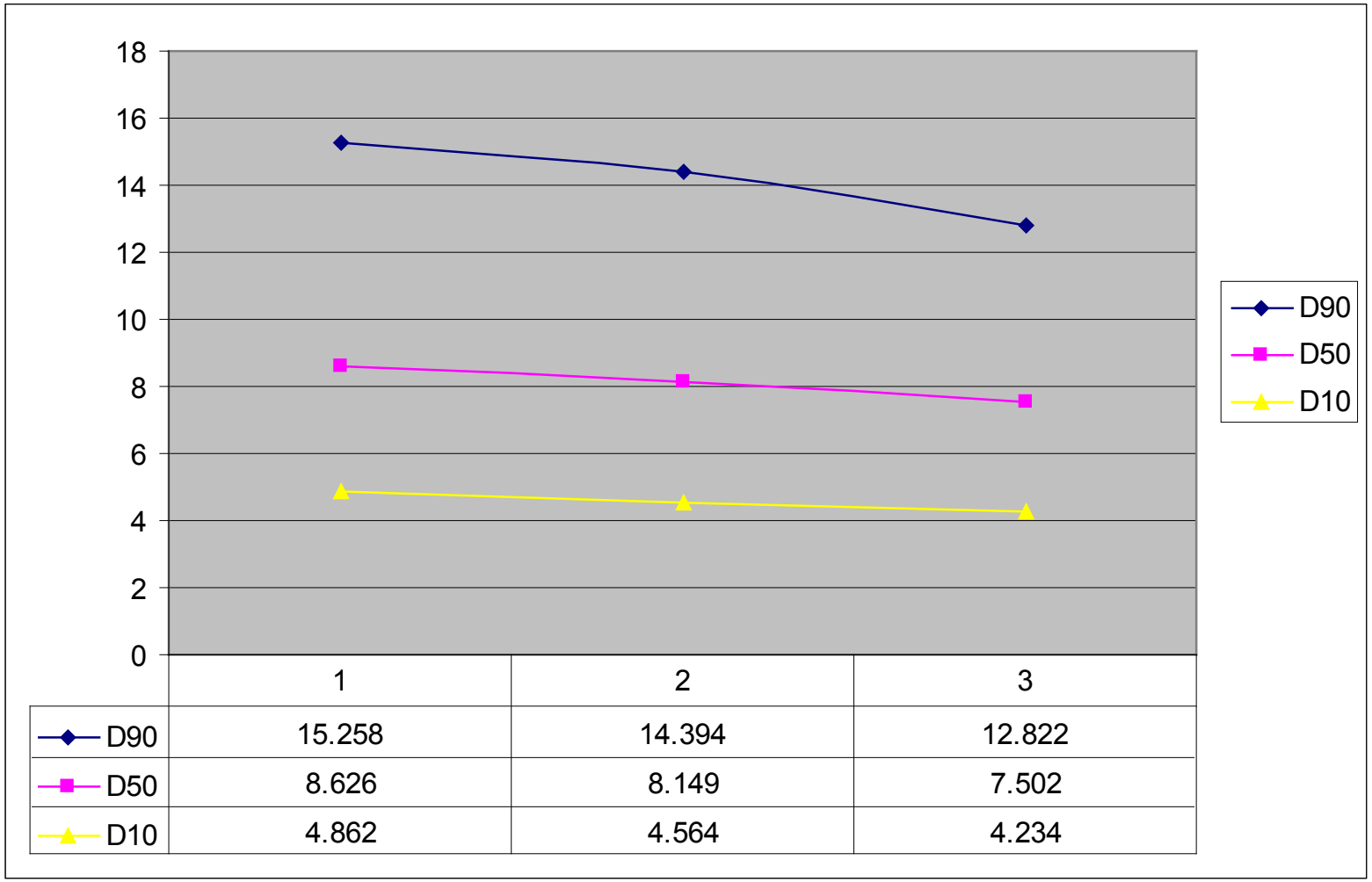


Examples

Mg Stearate

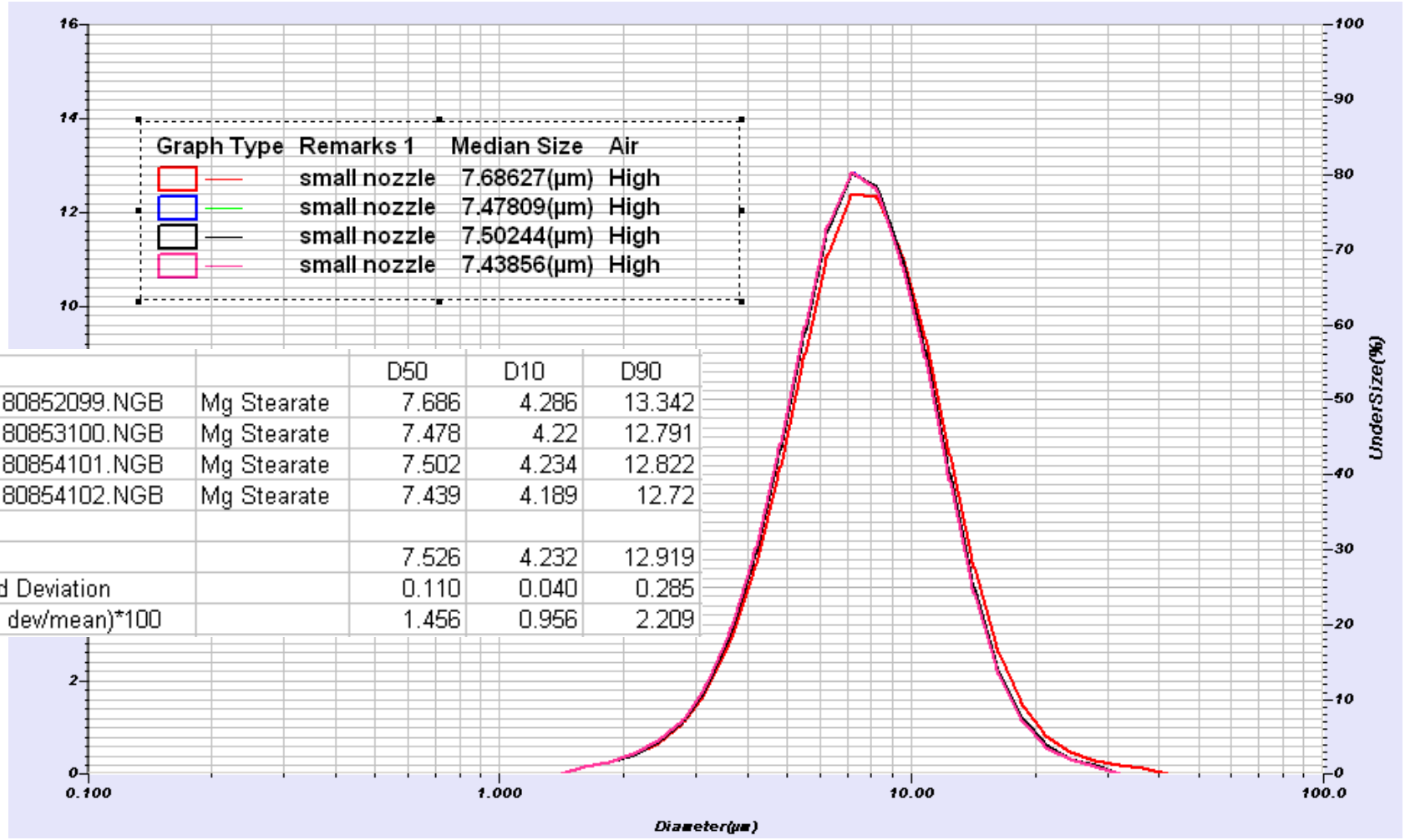


Mg Stearate



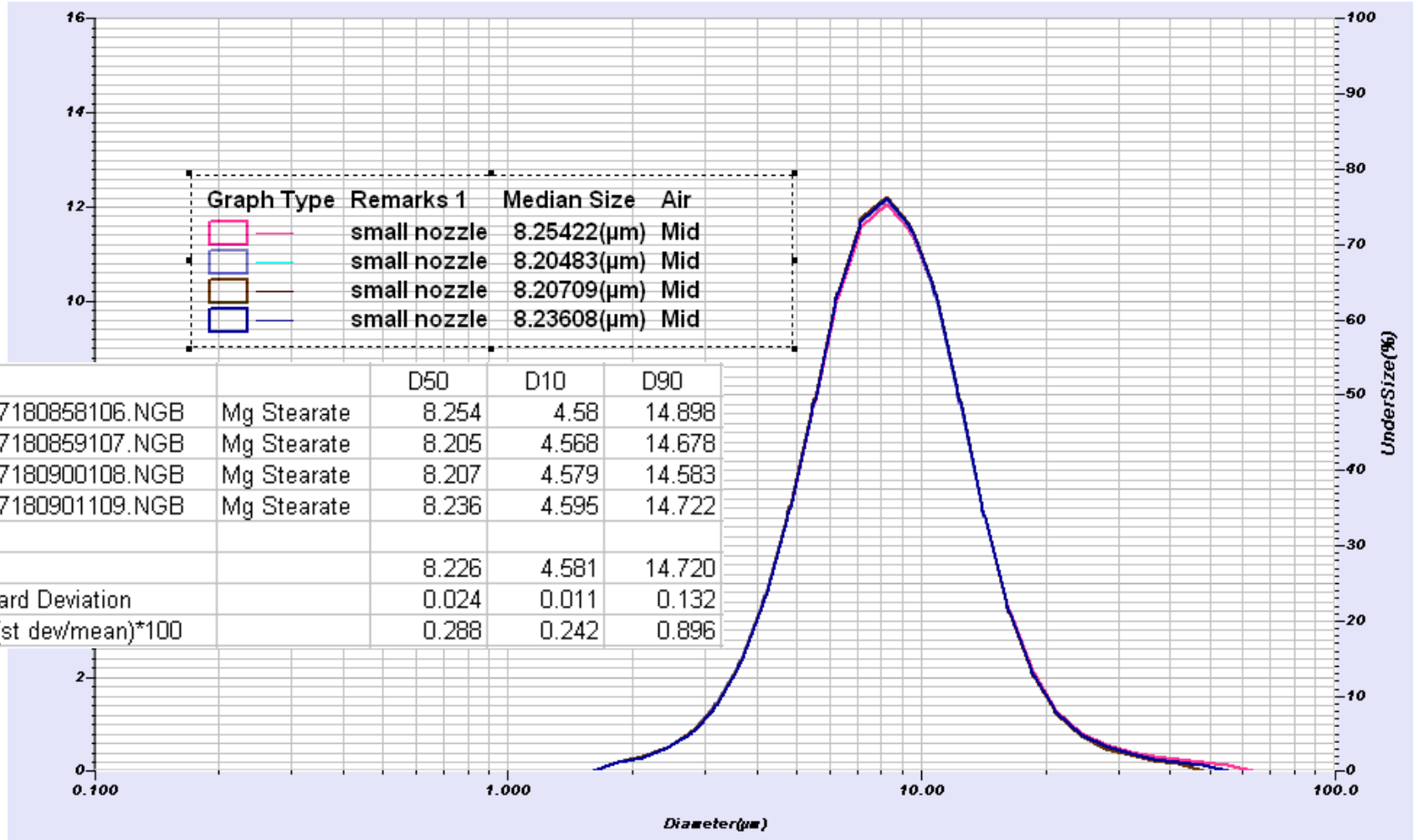
	1	2	3
D90	15.258	14.394	12.822
D50	8.626	8.149	7.502
D10	4.862	4.564	4.234

Reproducibility at 3 Bar



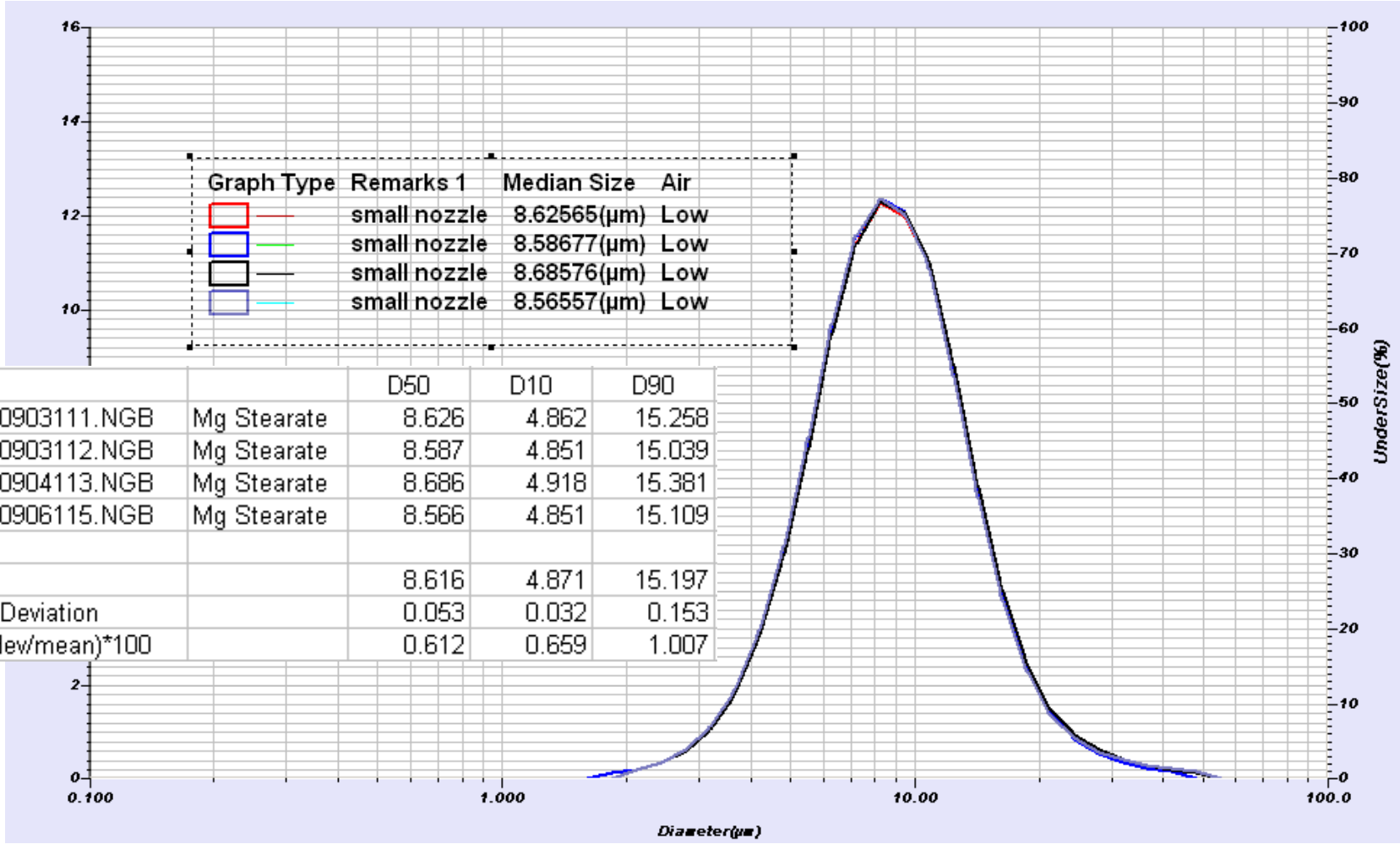
		D50	D10	D90
200707180852099.NGB	Mg Stearate	7.686	4.286	13.342
200707180853100.NGB	Mg Stearate	7.478	4.22	12.791
200707180854101.NGB	Mg Stearate	7.502	4.234	12.822
200707180854102.NGB	Mg Stearate	7.439	4.189	12.72
Mean		7.526	4.232	12.919
Standard Deviation		0.110	0.040	0.285
COV (st dev/mean)*100		1.456	0.956	2.209

Reproducibility at 2 Bar



		D50	D10	D90
200707180858106.NGB	Mg Stearate	8.254	4.58	14.898
200707180859107.NGB	Mg Stearate	8.205	4.568	14.678
200707180900108.NGB	Mg Stearate	8.207	4.579	14.583
200707180901109.NGB	Mg Stearate	8.236	4.595	14.722
Mean		8.226	4.581	14.720
Standard Deviation		0.024	0.011	0.132
COV (st dev/mean)*100		0.288	0.242	0.896

Reproducibility at 1 Bar



		D50	D10	D90
200707180903111.NGB	Mg Stearate	8.626	4.862	15.258
200707180903112.NGB	Mg Stearate	8.587	4.851	15.039
200707180904113.NGB	Mg Stearate	8.686	4.918	15.381
200707180906115.NGB	Mg Stearate	8.566	4.851	15.109
Mean		8.616	4.871	15.197
Standard Deviation		0.053	0.032	0.153
COV (st dev/mean)*100		0.612	0.659	1.007

Summarize Reproducibility

Pressure	D10 COV	D50 COV	D90 COV
1 Bar	0.659	0.612	1.007
2 Bar	0.242	0.288	0.896
3 Bar	0.956	1.456	2.209

This is reproducibility; Sampling is important!

Must have representative sample

Summary

HORIBA Scientific

Must have representative sample

Powders: select air pressure

Suspensions: wet, disperse

Check accuracy w/microscope or DLS

Investigate system settings: concentration, agitation, ultrasound

Design for maximum precision

Follow guidelines in standards

Thank you

Thank you

감사합니다

Cảm ơn

ありがとうございました

Dziękuję

धन्यवाद

Grazie

Merci

谢谢

நன்ற

ขอบคุณครับ

Obrigado

Σας ευχαριστούμε

شُكْرا

Tack ska ni ha

Большое спасибо

Danke

Gracias