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Supplementary Materials

Initial Stage Flocculation of Positively Charged Colloidal Particles in the Presence of Ultrafine Bubbles

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Microscopic analysis of particle dimer separation distances

This section provides the raw data supporting the analysis of inter-particle distances discussed in Section 3.2 of the main text. The center-to-center distances of AL dimers were measured from optical micrographs to quantify the effect of UFBs adsorption on particle separation. Table S1 lists all individual measurements for both the AL control (no UFBs) and the AL-UFBs system, along with the calculated mean and standard deviation for each dataset. The increased separation in the presence of UFBs provides direct visual evidence for the formation of an interfacial UFB layer.

AL with UFBs			AL Control		
Sample Type	**Dimer ID**	**Distance (μm)**	**Sample Type**	**Dimer ID**	**Distance (μm)**
AL with UFBs	1	1.37	AL Control	1	1.14
AL with UFBs	2	1.34	AL Control	2	1.14
AL with UFBs	3	1.39	AL Control	3	1.11
AL with UFBs	4	1.38	AL Control	4	1.10
AL with UFBs	5	1.36	AL Control	5	1.14
AL with UFBs	6	1.36	AL Control	6	1.12
AL with UFBs	7	1.35	AL Control	7	1.12
AL with UFBs	8	1.40	AL Control	8	1.12
AL with UFBs	9	1.38	AL Control	9	1.12
AL with UFBs	10	1.37	AL Control	10	1.11
Statistic	**n = 10**	**1.37**	**Statistic**	**n = 10**	**1.122**

Table S1. Individual center-to-center distance measurements for AL dimers in the presence and absence of UFBs.

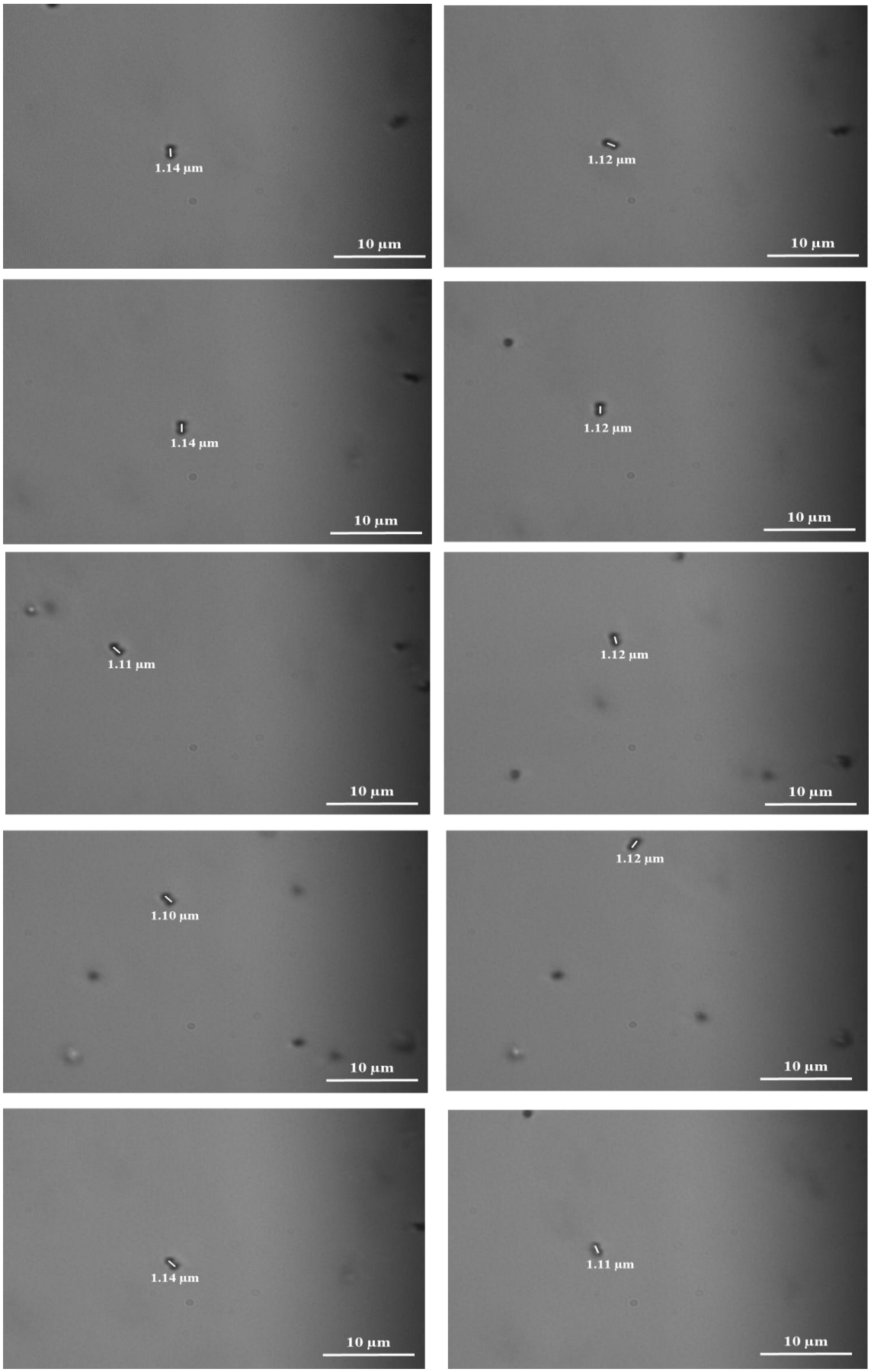


Figure S1: Microscopic images of AL dimers in the absence of UFBs (control) at pH 6.0 ± 0.3 and NaCl of 0.1 mM.

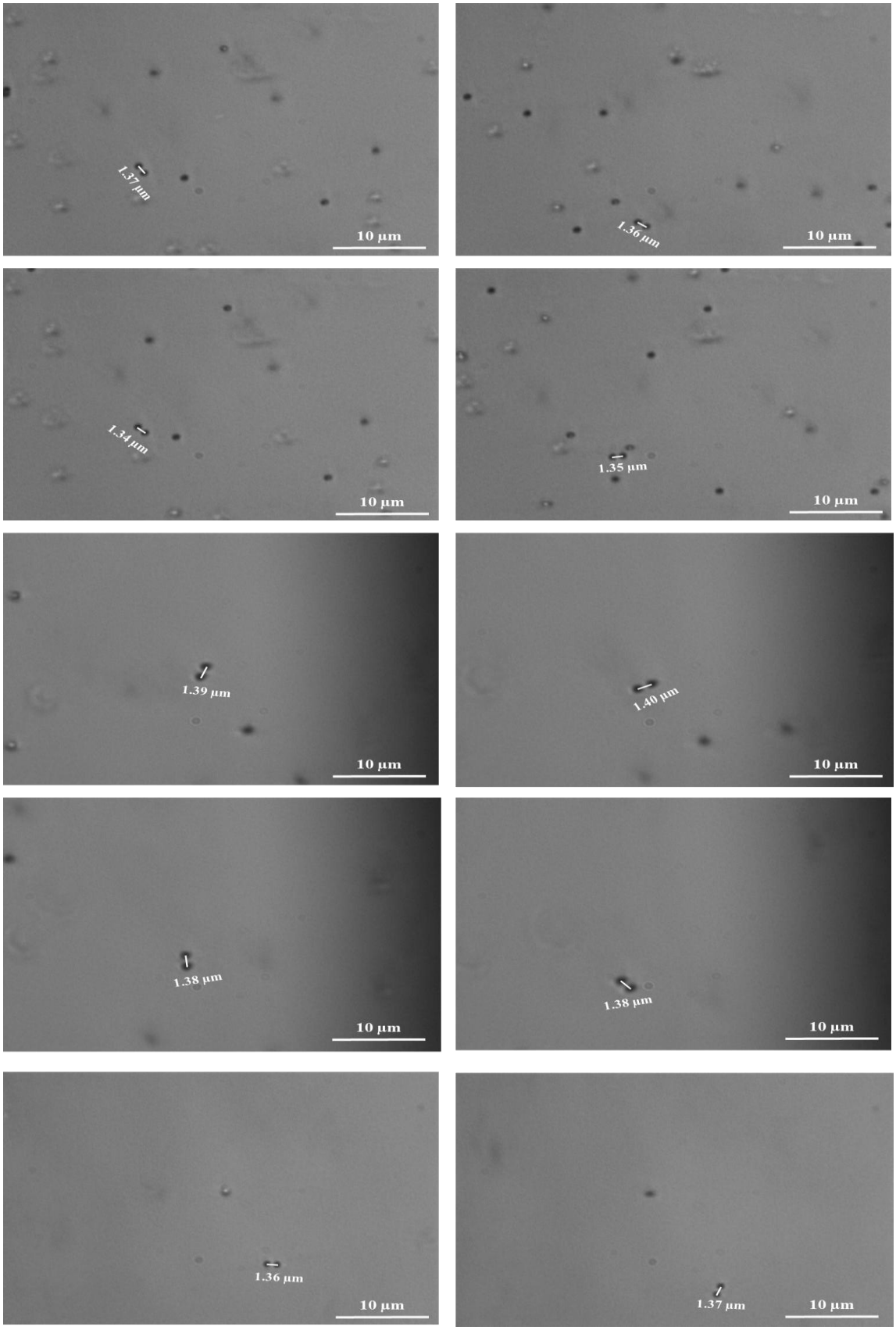


Figure S2: Microscopic images of AL dimers in the presence of UFBs at pH 6.0 ± 0.3 and NaCl of 0.1 mM.

Nanoparticle tracking analysis (NTA) data

Additional typical size distributions of air UFBs generated in H_20 measured by NanoSight using the NTA method 10 minutes after generation of UFBs are given below.

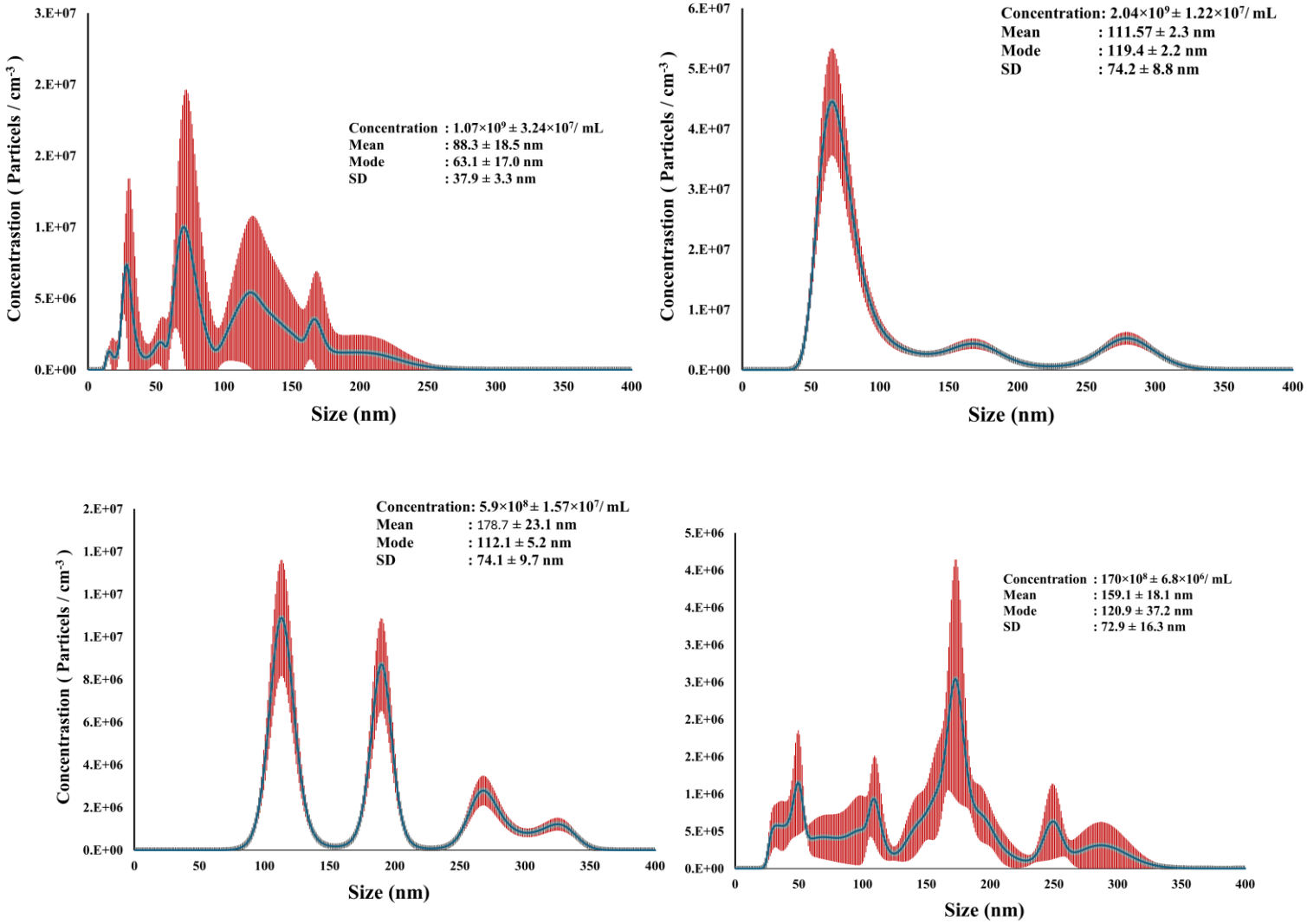


Figure S3: Additional representative NTA measurement results for air UFBs generated in H_20. All measurements were conducted 20 minutes after bubble generation. The red bars show standard error.

Characterization of amidine latex (AL) as model colloid

SEM measurement

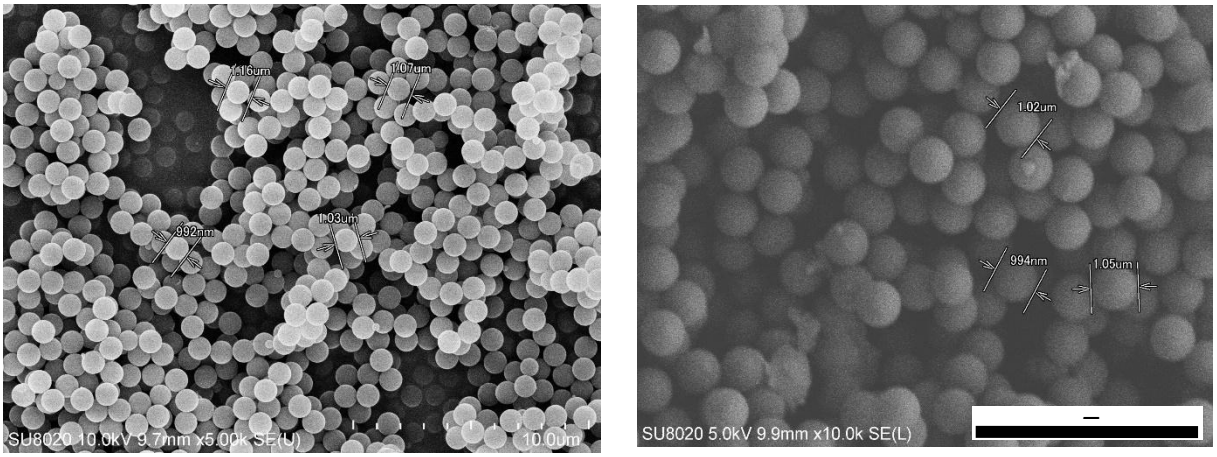


Figure S4: Scanning electron microscopy (SEM) images of the AL particles at (a) 5,000× and (b) 10,000× magnification, confirming their spherical morphology and primary particle size consistent with the manufacturer's specification.

DLS measurement

Replicate	Peak 1 (nm)	Mean ± sd (nm)	Reported size
1	1097		
2	969.3		
3	990.9	1030 ± 40	1.03 ± 0.04 μm
4	1007		
5	1081		
6	1009		

Table S2: DLS Peak-1 diameters of amidine latex particles showing a dominant monomodal sample with a mean size of 1.03 ± 0.04 μm.

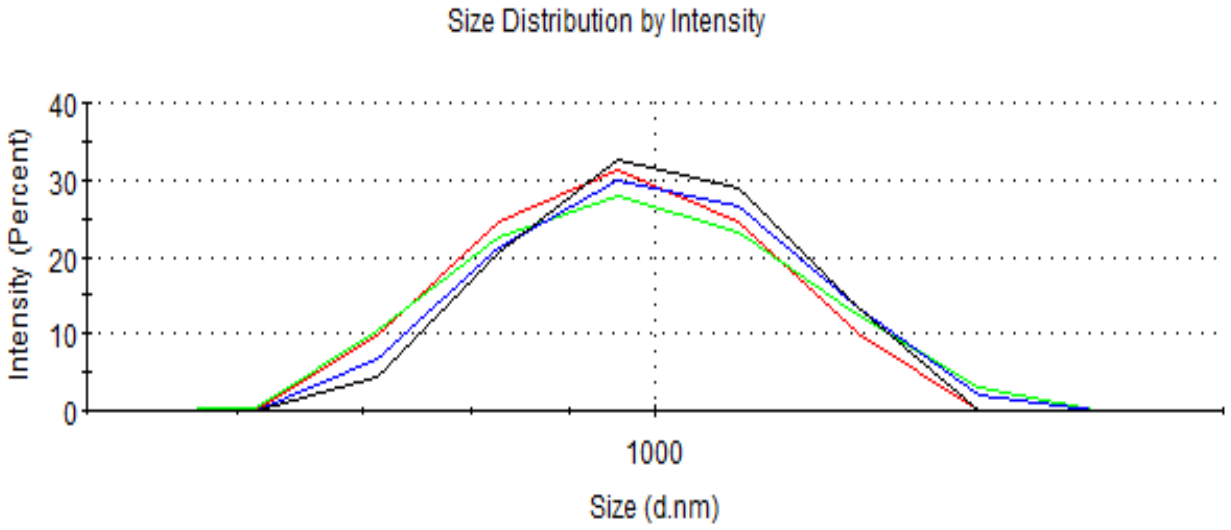


Figure S5: DLS intensity distribution of AL showing a monomodal peak near 1.0 μm. No secondary modes were observed, confirming the absence of measurable aggregates.

	Lot data	Specification
Physical properties of AL		
Density at 20°c	1.055 g / cm^3	N.a.
Refractive index at 590 nm, 20°c	1.591	N.a.
Technical data		
Material lot number	2482781	N.a.
Mean diameter (TEM)	0.98 μm	0.95 – 1.2 μm
Standard deviation of diameter	0.031 μm	N.a.
Coefficient of variation of diameter	3.2 %	≤ 10 %
Percent solids w/v	4.0 %	4.0 ± 0.5 %
Amidine charge titration data	15.3 μEq / g	N.a.
Bioburden test	Meets specification	0 CFU / ml
The calculated data		
Particle number per ml of latex	7.7×10^{10}	N.a.
Specific surface area	5.8×10^4 cm^2 / g	N.a.
Surface charge density	25.4 μC/cm^2	N.a.
Parking area per amidine group	63 A^2 / $C(NH)NH_2$	N.a.
Charge groups per particle	4.8×10^6	N.a.

Table S3. Manufacturer's specifications for the Amidine Latex particles (ThermoFisher Scientific, Lot #2965681, Product #A37322) as provided in the Certificate of Analysis.