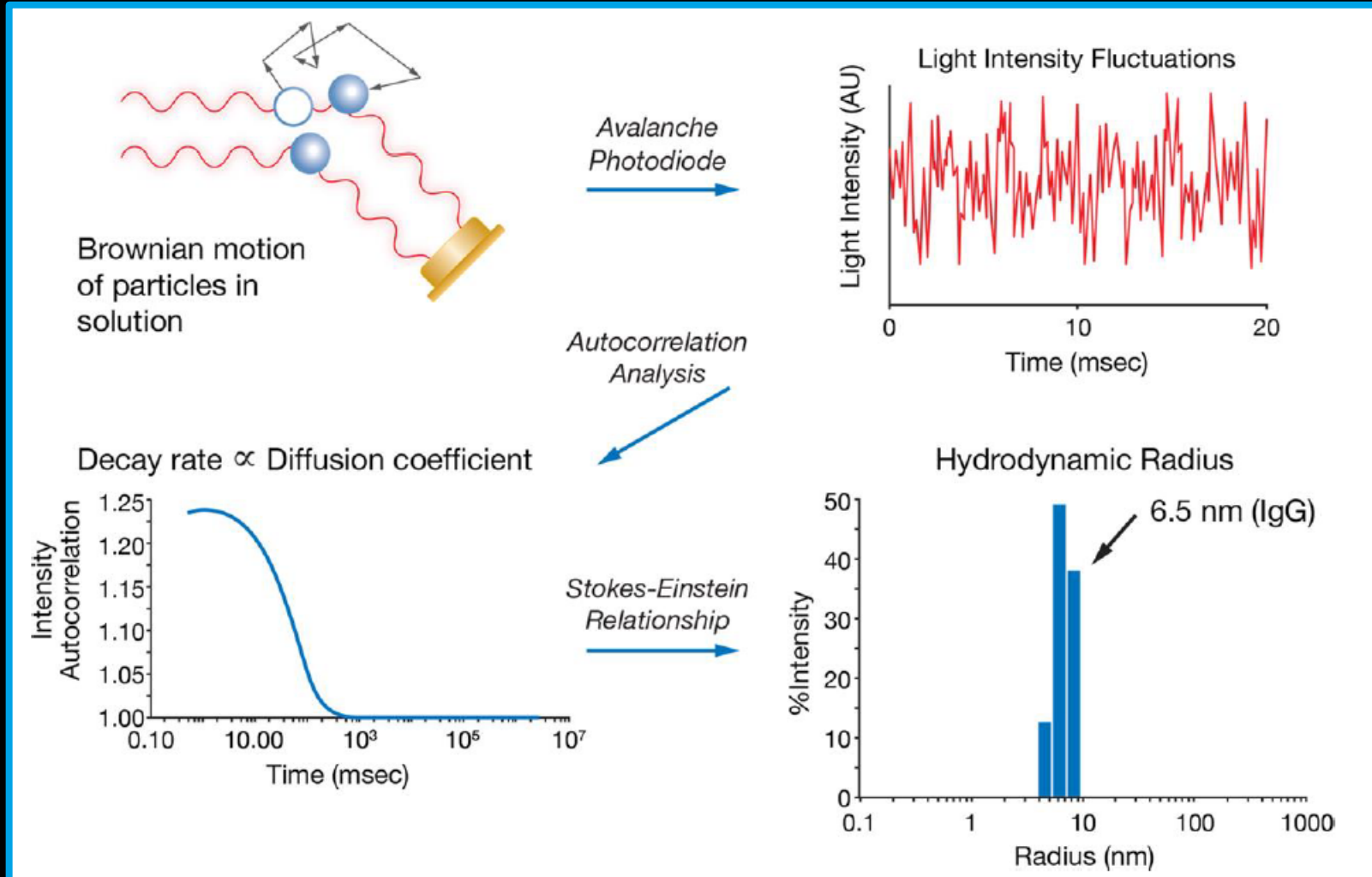


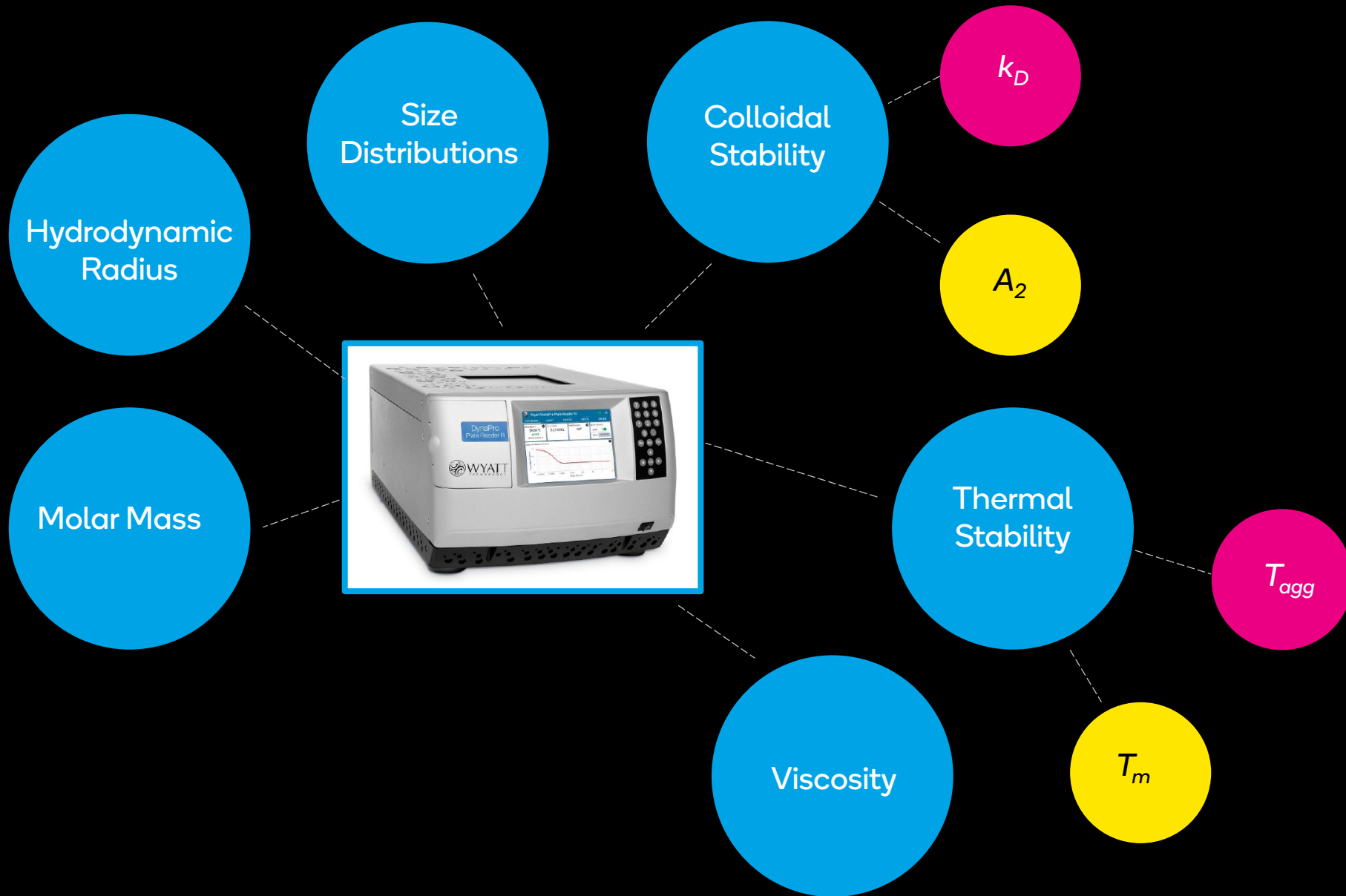
# Dynamic Light Scattering (DLS) Applications

# Introduction: Dynamic Light Scattering



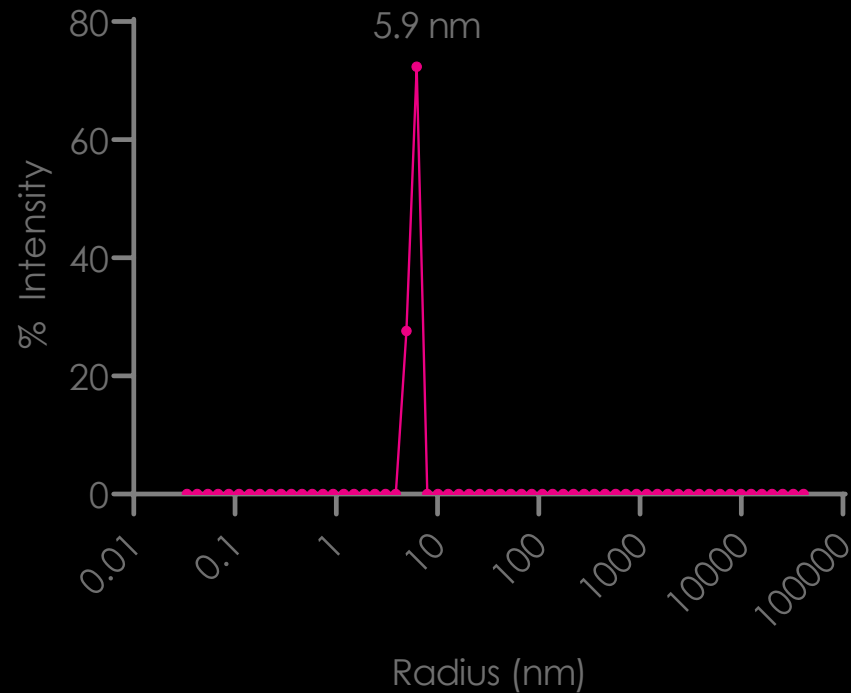
- DLS and SLS capabilities
- 96, 384, 1536 plate compatible
- Measurement range  $R_h$  : 0.5 – 1000 nm  
 $M_w$ : 1 – 1000 kDa
- Temperature control: 4 – 85 °C

# Applications: Dynamic Light Scattering

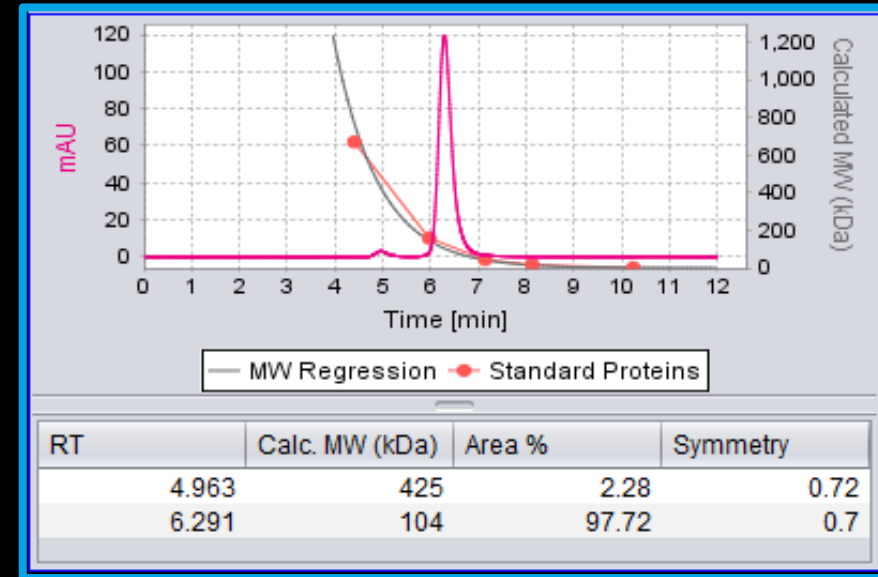


# Application 1A: Hydrodynamic radius

DLS



HPLC-SEC

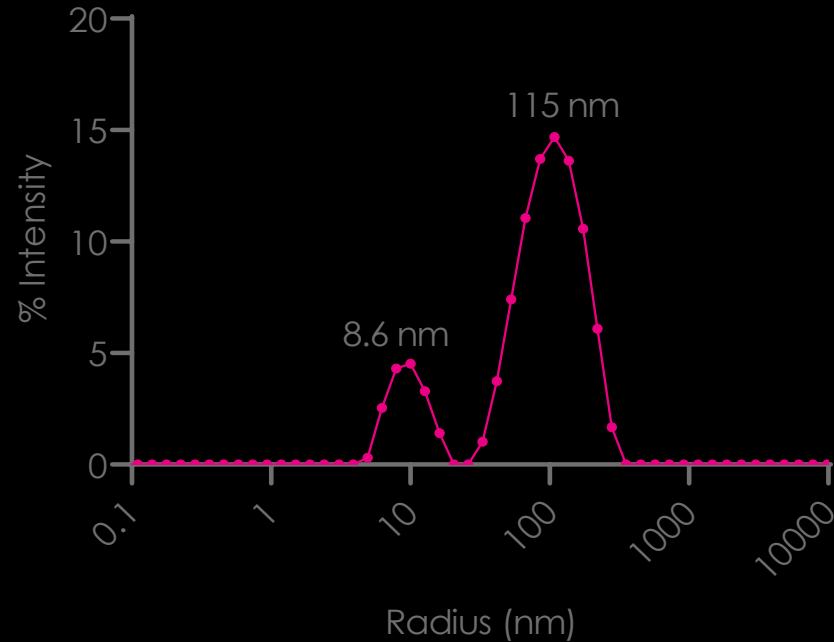


DLS can be used as a quality check to measure particles/aggregates larger than HPLC-SEC's size range (sub-visible particles)

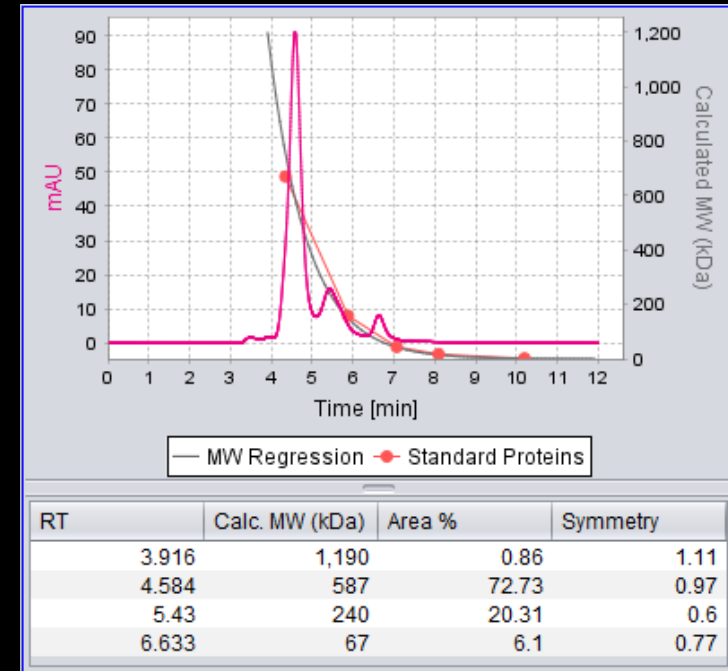
Complementary to HPCL-SEC for aggregate analysis

# Application 1B: Hydrodynamic radius

DLS



HPLC-SEC

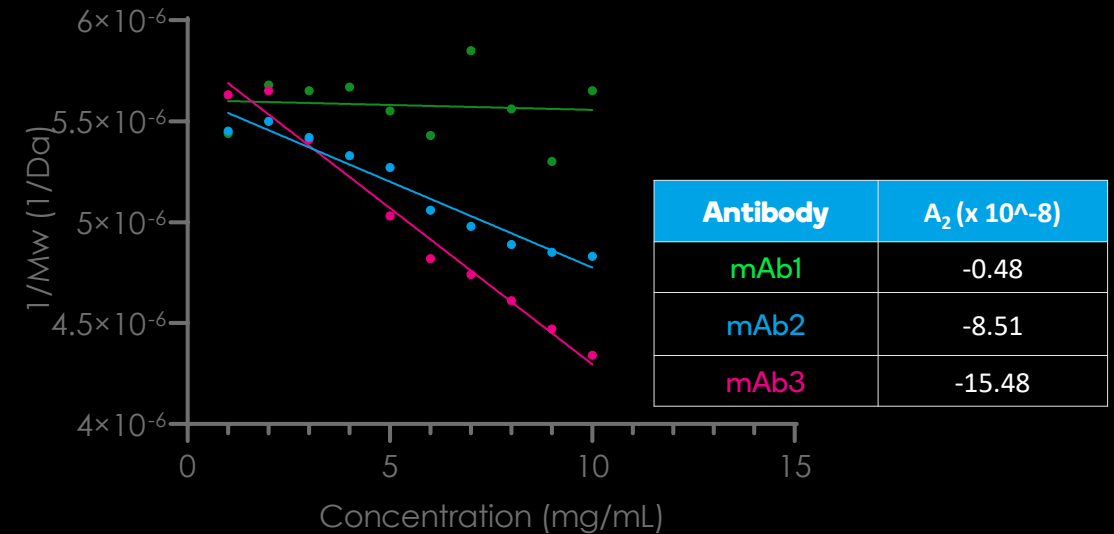
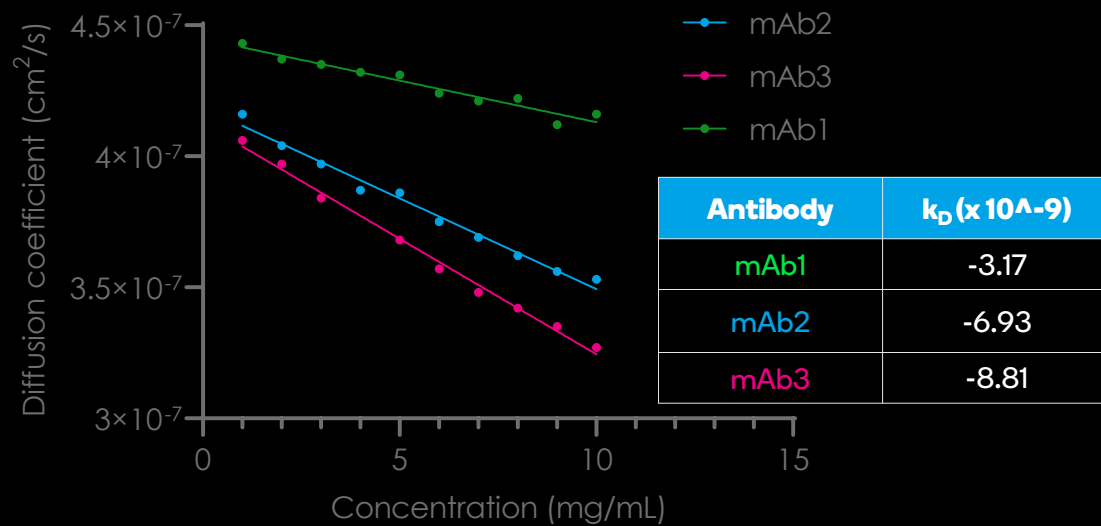


DLS can be used as a quality check to measure particles/aggregates bigger than HPLC-SEC's size range (sub-visible particles)

Complementary to HPLC-SEC for aggregate analysis

# Application 2A: Colloidal Stability ( $k_D$ and $A_2$ )

## Relative molecule ranking of self-association propensity

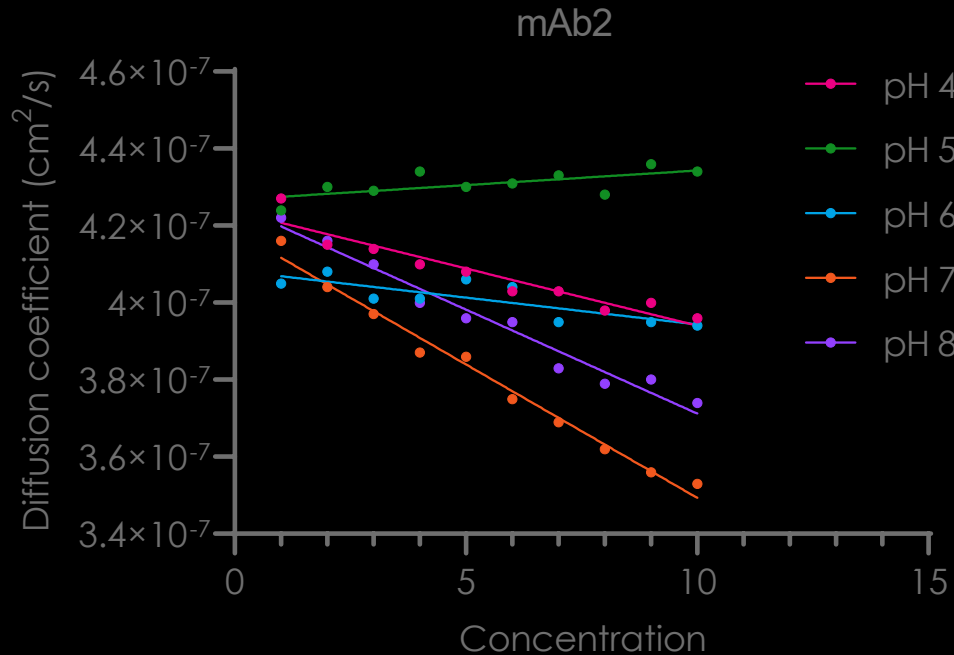


DLS can be used to rank molecules for their propensity to self-aggregate.

Two parameters ( $k_D$  and  $A_2$ ) can be negative (self-association), neutral (no interaction) or positive (repulsive forces).

# Application 2B: Colloidal Stability

## Relative formulation buffer ranking for self-association propensity

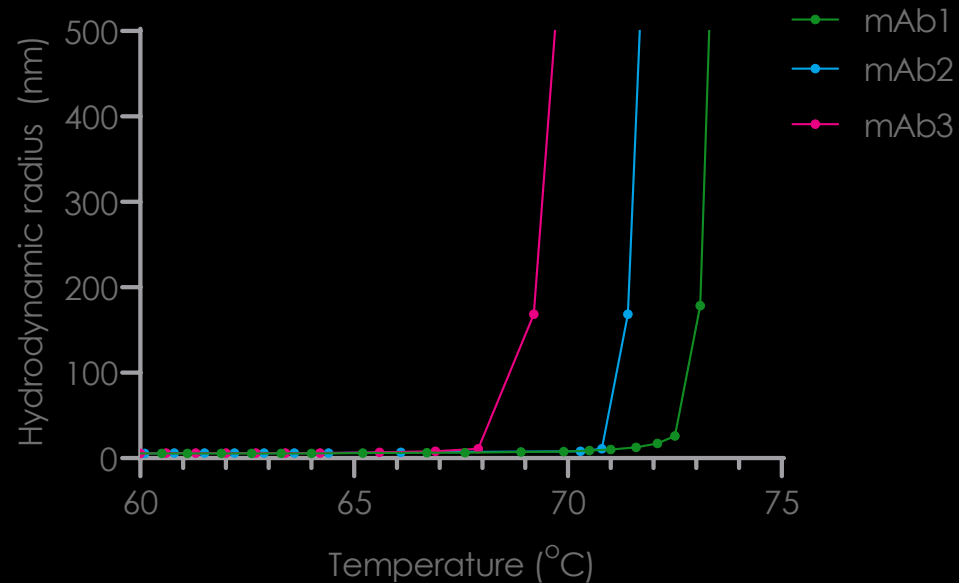


Buffer pH	$k_D$ ( $\times 10^{-9}$ )
4	-3.0
5	0.7
6	-1.4
7	-6.9
8	-5.4

Ideal formulation buffer is expected to arrest self-interactions thereby reducing aggregation propensity. Measurement of  $k_D$  as a function of formulation buffer can be used for relative ranking.

# Application 3A: Thermal Stability

Relative molecule ranking for  $T_{\text{aggregation}}$



Antibody	$T_{\text{agg}}$ °C
mAb1	70
mAb2	71
mAb3	67

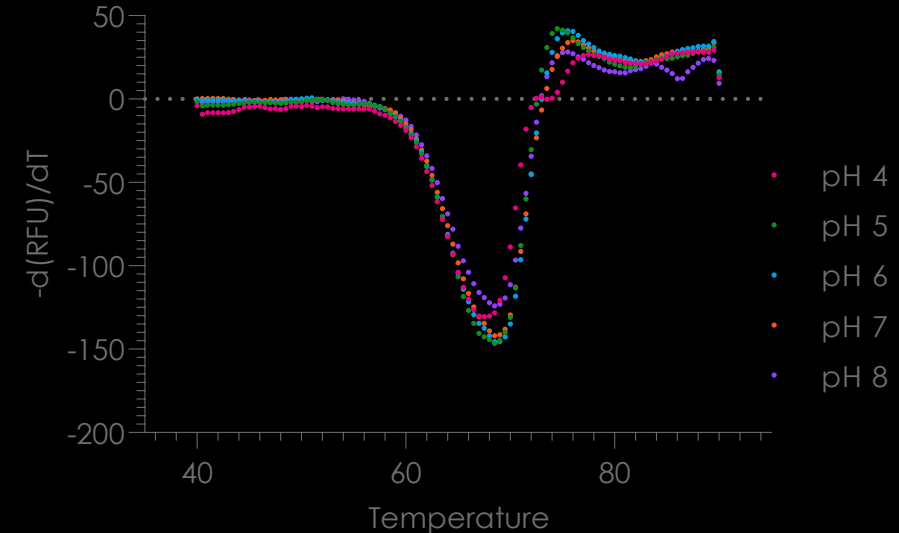
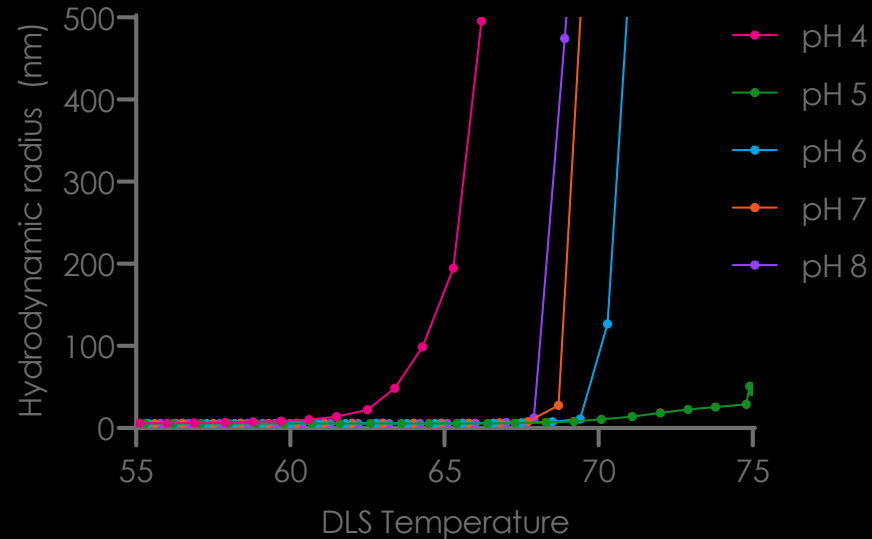
$T_{\text{agg}}$  is the temperature at which a protein begins aggregating ( $T_{\text{onset}}$ ). Here, it is defined as the temperature at which the hydrodynamic radius deviates by 10% from the baseline



# Application 3B: Thermal Stability

Relative formulation buffer ranking for  $T_{\text{aggregation}}$

mAb2



Buffer pH	$T_{\text{agg}}$ (10 mg/ml)	$T_m$ (DSF) (0.5 mg/ml)
4	59.7	67.5
5	68.3	68.5
6	68.5	68.5
7	67.7	68.5
8	67.0	68.5

Ideal formulation buffer is expected to stabilize the protein, thereby increasing the  $T_{\text{agg}}$ . Measurement of  $T_{\text{agg}}$  as a function of formulation buffer can be used for relative ranking.

$T_m$  by DSF maybe limited by resolution in some cases.