GDD/TRD/Bx

Stability Predictions for mAbs Using Arrhenius-Based Kinetics

Mitja Zidar
CMC Strategy Forum Japan – CASSS
October 4th, 2023

NOVARTIS | Reimagining Medicine

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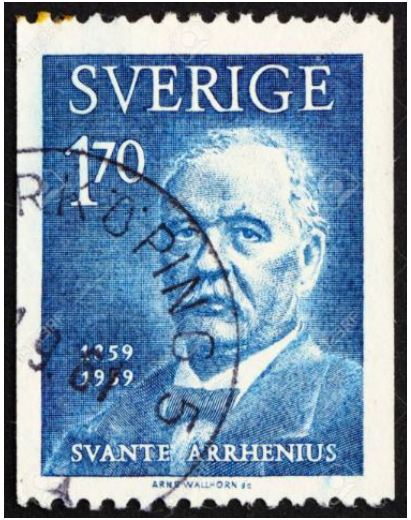
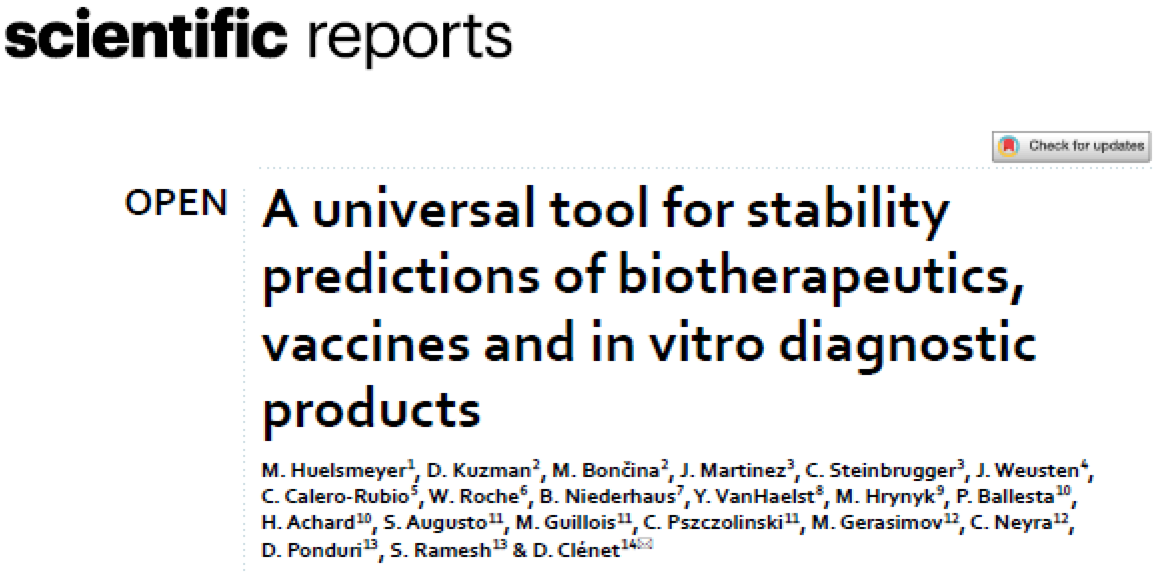
Agenda



- Full AKM model – what *can* be done
- From two to one step Arrhenius based kinetic models
- Does aggregation of proteins follow Arrhenius relation?
- Example of application
- Conclusions

Advanced kinetic modeling in the industry



scientific reports

OPEN

A universal tool for stability predictions of biotherapeutics, vaccines and in vitro diagnostic products

M. Huelsmeyer[1], D. Kuzman[2], M. Bončina[2], J. Martinez[3], C. Steinbrugger[3], J. Weusten[4], C. Calero-Rubio[5], W. Roche[6], B. Niederhaus[7], Y. VanHaelst[8], M. Hrynyk[9], P. Ballesta[10], H. Achard[10], S. Augusto[11], M. Guillois[11], C. Pszczolinski[11], M. Gerasimov[12], C. Neyra[12], D. Ponduri[13], S. Ramesh[13] & D. Clénet[14]✉

Companies involved:

Abbvie

Novartis

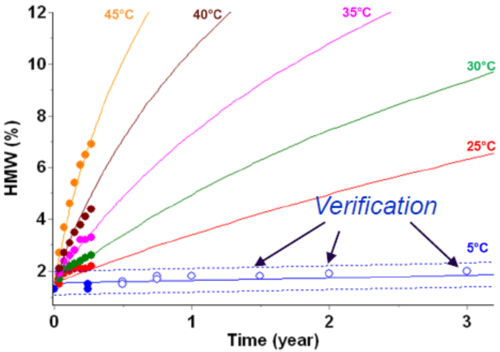
bioMérieux

MSD

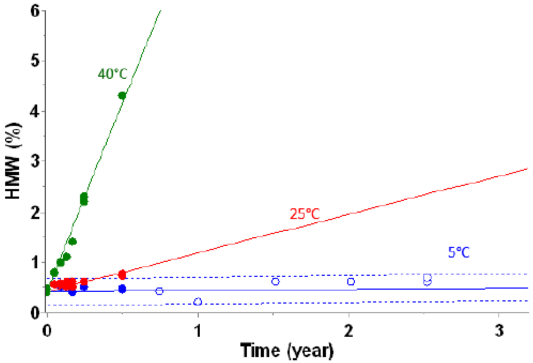
Sanofi

Excellent Agreement With the Experimental Real-time Data

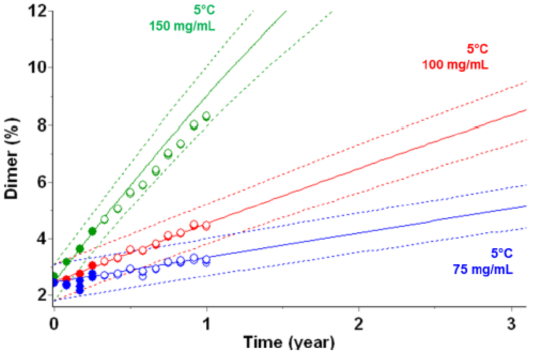
abbvie - liquid mAb
HMW



NOVARTIS – liquid mAb
HMW



sanofi – Single variable domain
Dimers



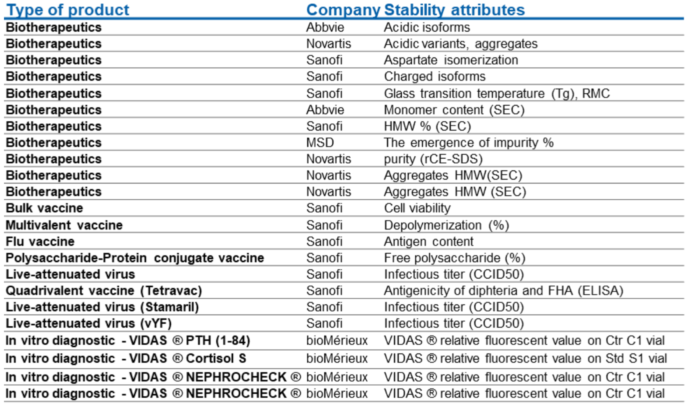
Kinetic rate $d\alpha/dt$ was phenomenologicaly described as the sum of two individual one-step reactions:

$$\frac{d\alpha}{dt} = v \times A_1 \times exp\left(-\frac{E_{\alpha 1}}{RT}\right) \times (1-\alpha_1)^{n1} \times {\alpha_1}^{m1} \times C^{p1} + (1-v) \times A_2 \times exp\left(-\frac{E_{\alpha 2}}{RT}\right) \times (1-\alpha_2)^{n2} \times {\alpha_2}^{m2} \times C^{p2}$$

Excellent Agreement With the Experimental Real-time Data

Evaluation of Arrhenius-based kinetic models through a cross-company perspective on stability modeling:

- key stability indicating attributes
- different types of biotherapeutics, vaccines and biomolecules combined in in vitro diagnostic kits
- five biopharma companies
- stability predictions up to 3 years for products at storage conditions
- temperature excursions outside the cold-chain



Type of product	Company	Stability attributes
Biotherapeutics	Abbvie	Acidic isoforms
Biotherapeutics	Novartis	Acidic variants, aggregates
Biotherapeutics	Sanofi	Aspartate isomerization
Biotherapeutics	Sanofi	Charged isoforms
Biotherapeutics	Sanofi	Glass transition temperature (Tg), RMC
Biotherapeutics	Abbvie	Monomer content (SEC)
Biotherapeutics	Sanofi	HMW % (SEC)
Biotherapeutics	MSD	The emergence of impurity %
Biotherapeutics	Novartis	purity (rCE-SDS)
Biotherapeutics	Novartis	Aggregates HMW(SEC)
Biotherapeutics	Novartis	Aggregates HMW (SEC)
Bulk vaccine	Sanofi	Cell viability
Multivalent vaccine	Sanofi	Depolymerization (%)
Flu vaccine	Sanofi	Antigen content
Polysaccharide-Protein conjugate vaccine	Sanofi	Free polysaccharide (%)
Live-attenuated virus	Sanofi	Infectious titer (CCID50)
Quadrivalent vaccine (Tetravac)	Sanofi	Antigenicity of diphteria and FHA (ELISA)
Live-attenuated virus (Stamaril)	Sanofi	Infectious titer (CCID50)
Live-attenuated virus (vYF)	Sanofi	Infectious titer (CCID50)
In vitro diagnostic - VIDAS ® PTH (1-84)	bioMérieux	VIDAS ® relative fluorescent value on Ctr C1 vial
In vitro diagnostic - VIDAS ® Cortisol S	bioMérieux	VIDAS ® relative fluorescent value on Std S1 vial
In vitro diagnostic - VIDAS ® NEPHROCHECK ®	bioMérieux	VIDAS ® relative fluorescent value on Ctr C1 vial
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Sci Rep. 2023 Jun 21;13(1):10077. doi: 10.1038/s41598-023-35870-6.

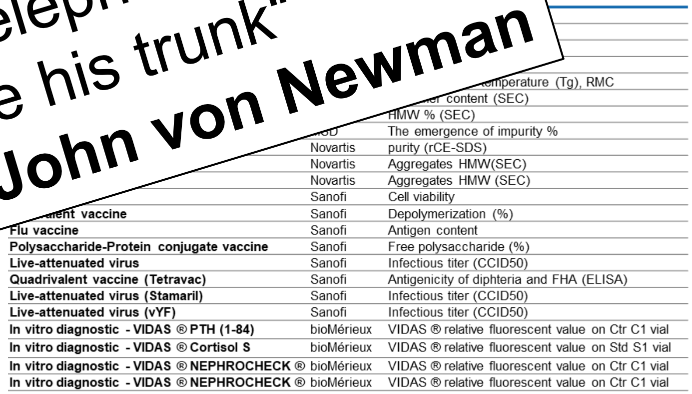
Excellent Agreement With the Experimental Real-time Data

„With 4 parameters, I can fit an elephant, and with 5, I can make him wiggle his trunk“

John von Newman

Evaluation of Arrhenius-based kinetic models throug
cross-company perspective on stability mod

- key stability indicating attributes
- different types of biothe
  biomolecules
- five bi
- stab … storage
  cond
- temp … de the cold-chain



		emperature (Tg), RMC
		er content (SEC)
		HMW % (SEC)
	SD	The emergence of impurity %
	Novartis	purity (rCE-SDS)
	Novartis	Aggregates HMW(SEC)
	Novartis	Aggregates HMW (SEC)
	Sanofi	Cell viability
alent vaccine	Sanofi	Depolymerization (%)
Flu vaccine	Sanofi	Antigen content
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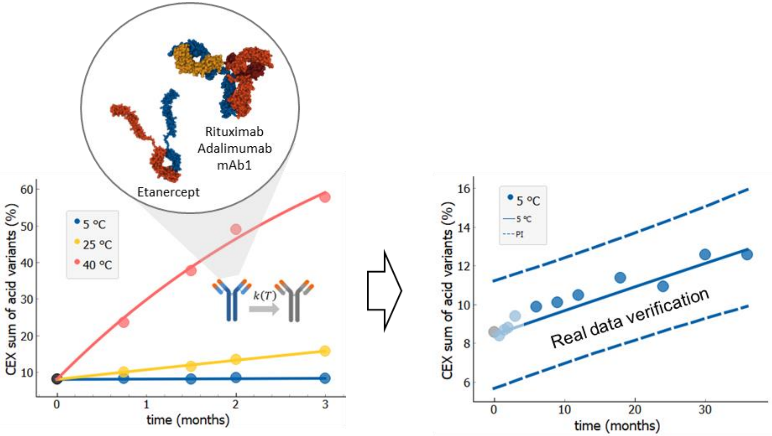
Simplified Biotherapeutic Degradation Models: One Step First-Order Kinetics

$$\frac{d\alpha}{dt} = v \times \boxed{A_1 \times exp\left(-\frac{E_{a1}}{RT}\right) \times (1-\alpha_1)}^{n1} \times \alpha_1{}^{m1} \times C^{p1} + (1-v) \times A_2 \times exp\left(-\frac{E_{a2}}{RT}\right) \times \ (1-\alpha_2)^{n2} \times \alpha_2{}^{m2} \times C^{p2}$$

$$\frac{d\alpha}{dt} = A \times exp\left(-\frac{E_a}{RT}\right) \times (1-\alpha)$$

Stability profiles of Charge profiles, aggregation, fragmentation, potency at standard storage conditions.



Accelerated stability data

Three years prediction

Sci Rep. 2021 Oct 15;11(1):20534. doi: 10.1038/s41598-021-99875-9.

Example Models for a Rituximab batch

Aggregation

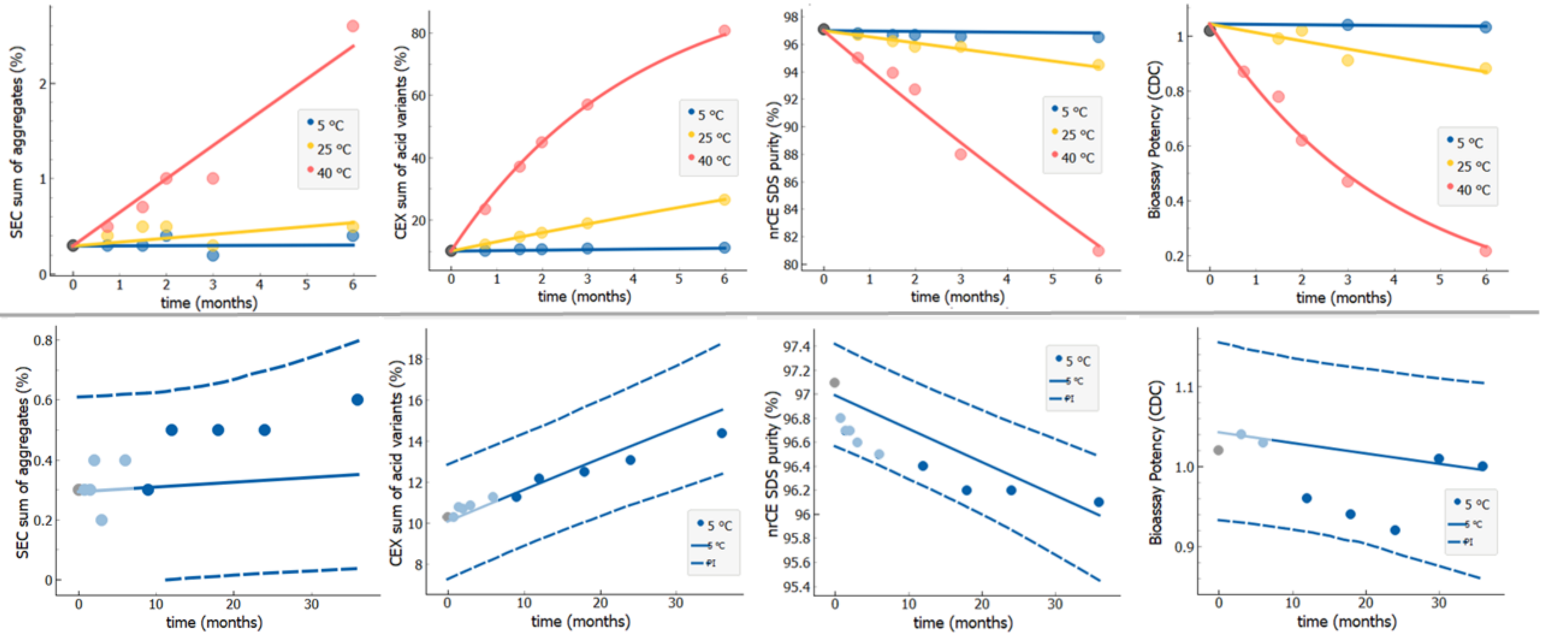
Acid variants

Fragmentation

Potency

Measured six months accelerated stability data and kinetic models

Prediction verification by 36 months experimental stability data



Sci Rep. 2021 Oct 15;11(1):20534. doi: 10.1038/s41598-021-99875-9.

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Long-Term Stability Predictions Verifications with Experimental Data

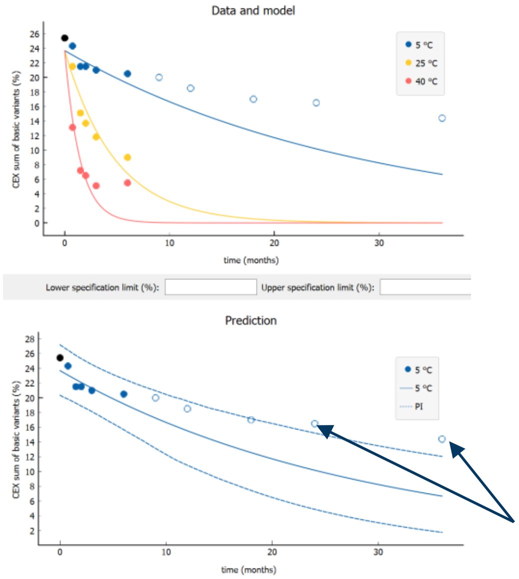
	mAb1	Rituximab	Etanercept	Adalimumab
CEX amount of main variants (%)	100%	91%	100%	99%
CEX sum of acid variants (%)	100%	97%	100%	99%
nrCE SDS purity (%)	100%	95%	100%	98%
nrCE SDS sum of fragments (%)	100%	no data	no data	no data
Relative potency	n.a.	97%	100%	100%
SEC purity (%)	100%	98%	90%	100%
SEC sum of aggregates (%)	100%	94%	90%	97%
CEX amount of basic variants	85%	90%	74%	99%
Number of batches	8	36	11	19

Ratio of experimental long-term data points between 6 and 36 months at 5 °C that fall within the 95 % prediction interval relative to the total number of all measured values during that time (e.g. 100 % - all longterm data fall within prediction interval)

Sci Rep. 2021 Oct 15;11(1):20534. doi: 10.1038/s41598-021-99875-9.

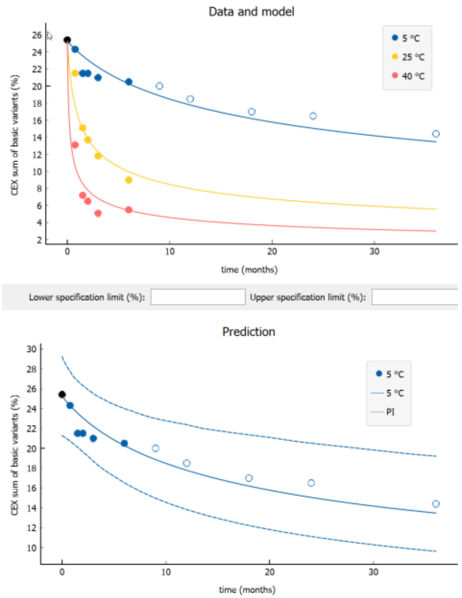
Degradation of „Sum of basic variants“ Described by 2-Step Model

One step – first order model



Above the Prediction Interval

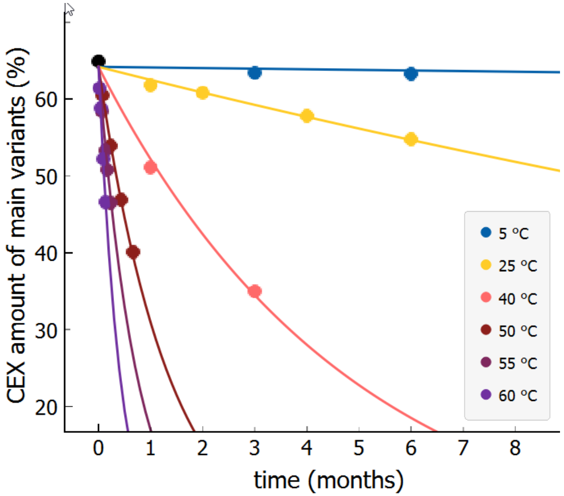
Two steps model



(Non) Arrhenius Degradation

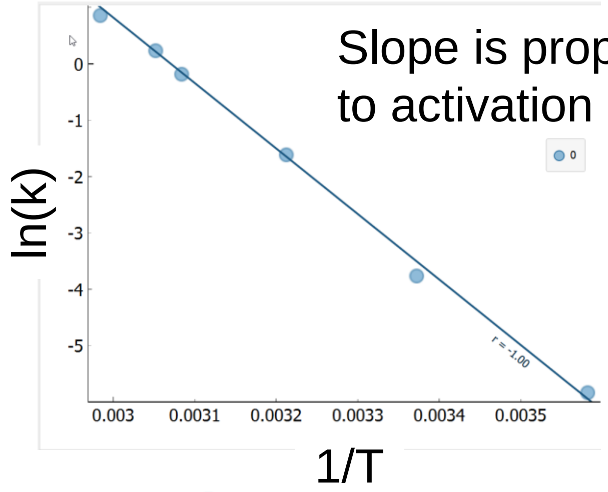
Arrhenius relation for temperature dependent kinetic rate of degradation proces:

$$k = A \times e^{-\frac{E_a}{RT}}$$



Arrhenius plot:

$$\mathbf{ln(k)} = \mathbf{ln}\ (\boldsymbol{A}) - \frac{\boldsymbol{E_a}}{\boldsymbol{R}} \times \frac{\mathbf{1}}{\boldsymbol{T}}$$



Slope is proportional to activation energy

Aggregation: Non-Arrhenius Plots from Literature

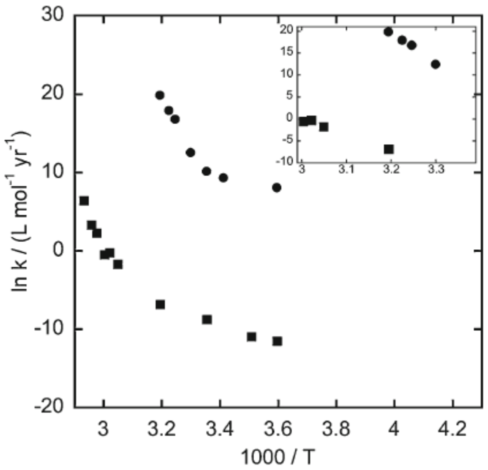


Fig. 4. Arrhenius plot of observed rate coefficient values for aggregation of bG-CSF (*circles*) and a MAb (*squares*). See main text for additional details

Wang & Roberts: AAPS Journal, Vol. 15, No. 3, July 2013

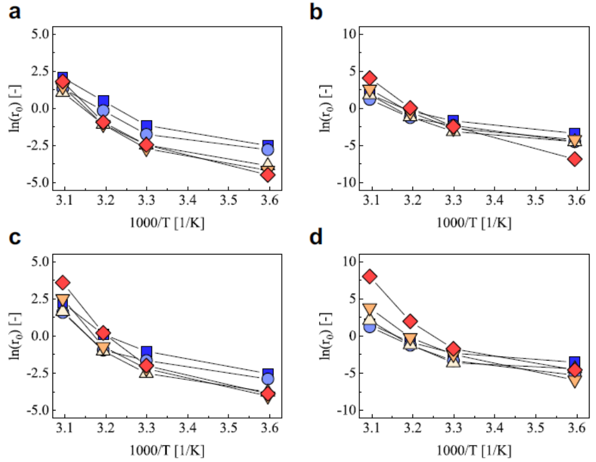
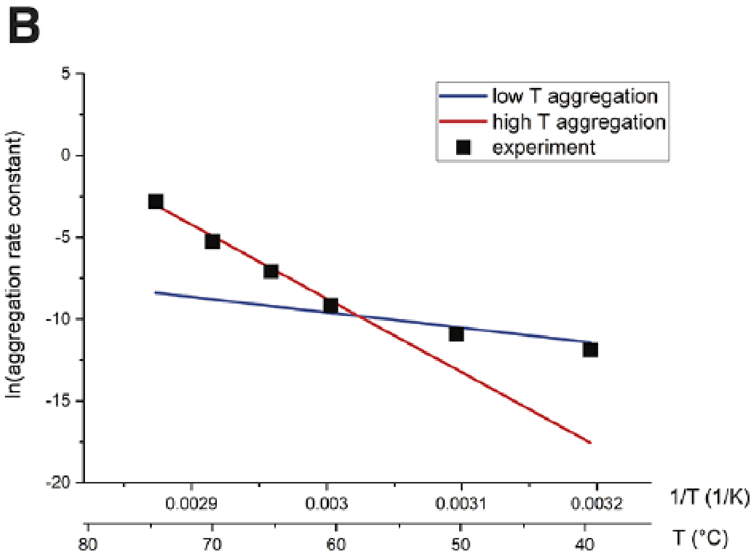
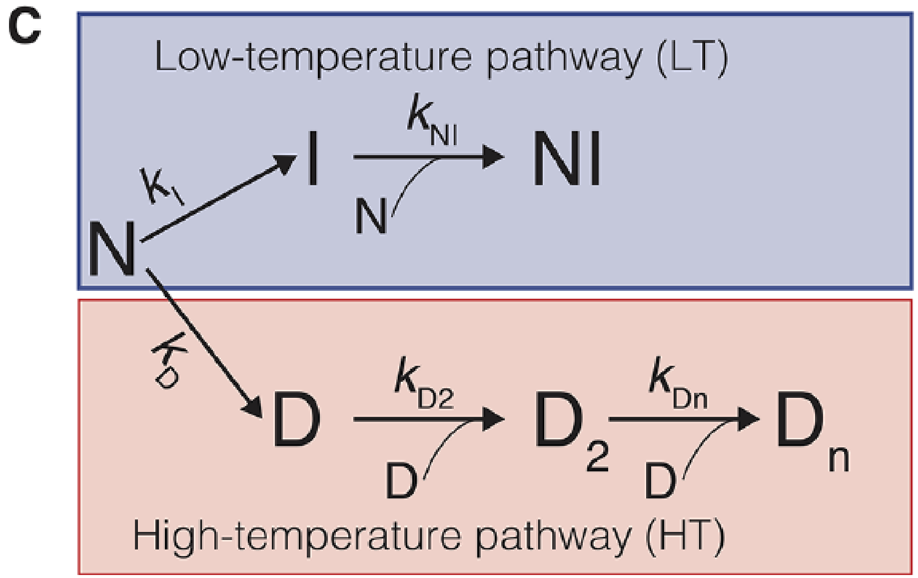


Figure 2. Arrhenius plot of the initial aggregation rate at 100 mg/mL mAb concentration and pH = pI (blue squares), pI − pH = 1 (light blue circles), pI − pH = 2 (yellow upward triangles), pI − pH = 3 (orange downward triangles), and pI − pH = 4 (red diamonds). (a) mAb-1 without excipient, (b) mAb-2 without excipient, (c) mAb-1 with 150 mM sodium chloride, (d) mAb-2 with 150 mM sodium chloride.

Wälchli: Journal of Pharmaceutical Sciences 109 (2020) 595-602

Temperature is Promoting Different Aggregation Pathways

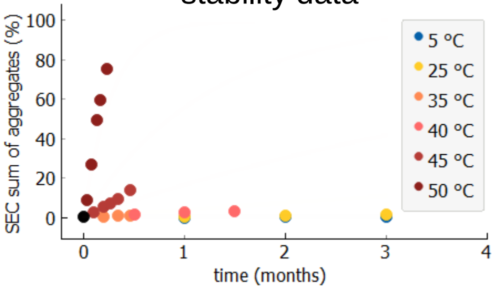




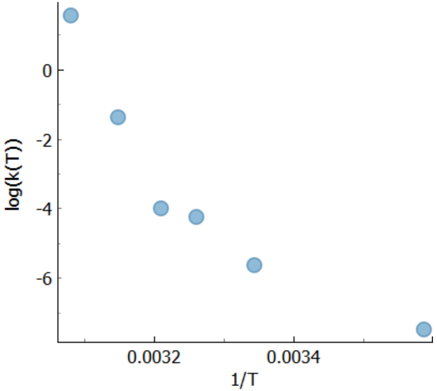
Journal of Medicinal Chemistry 2022 65 (3), 2623-2632,
DOI: 10.1021/acs.jmedchem.1c02010

Aggregation Prediction for a Fusion Protein

Accelerated stability data

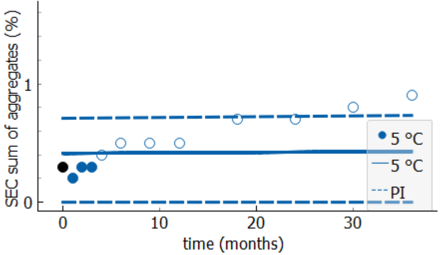


Arrhenius plot



T [5 - 50 °C]

Ea = 59.6 kcal/mol



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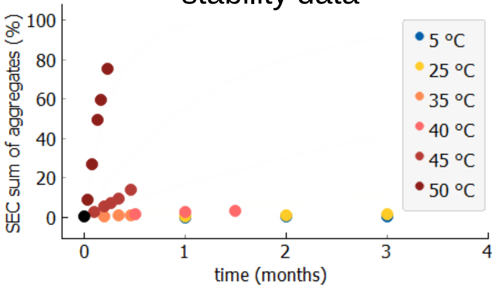
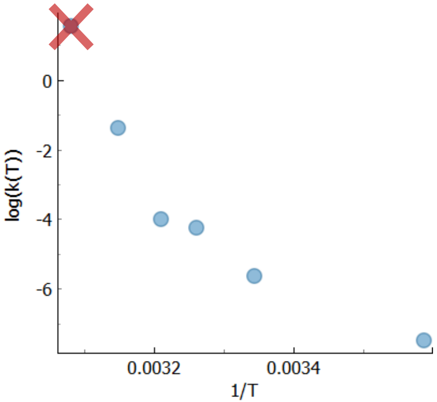


Arrhenius plot



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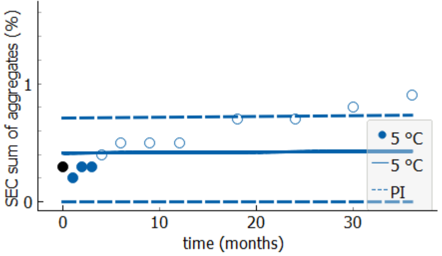
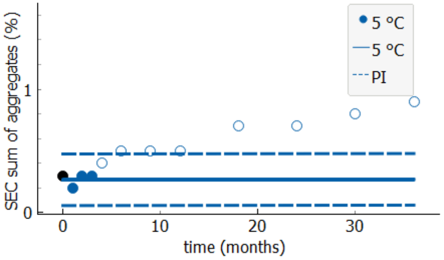
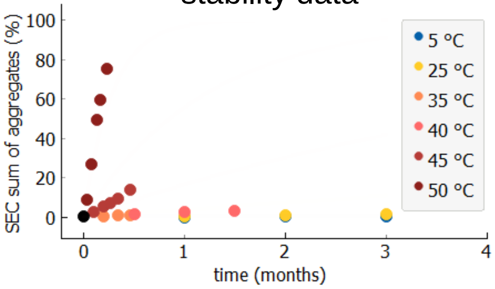


T [5 - 45 °C]
Ea = 54.0 kcal/mol

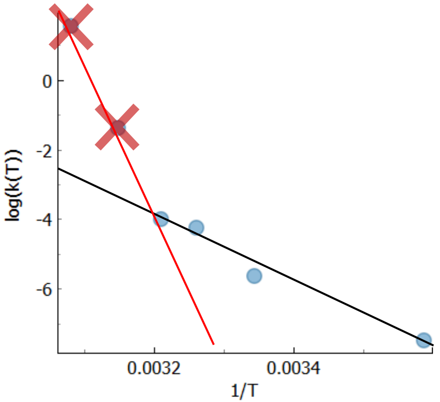


Aggregation Prediction for a Fusion Protein

Accelerated stability data



Arrhenius plot



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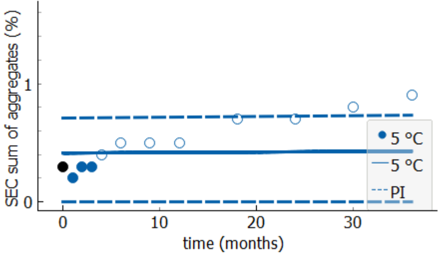

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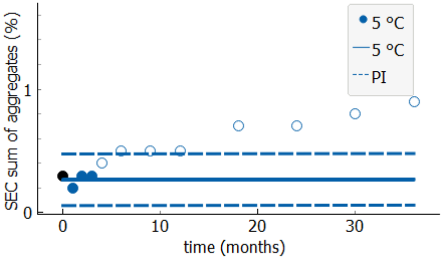
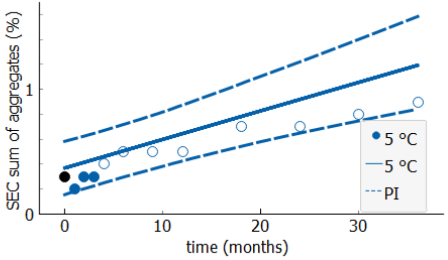
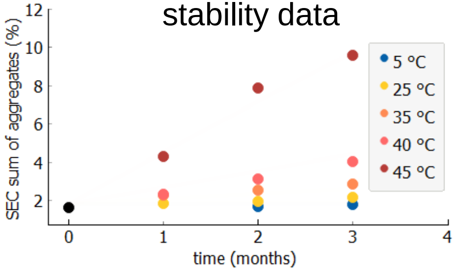


T [5 - 40 °C]
Ea = 21.4 kcal/mol

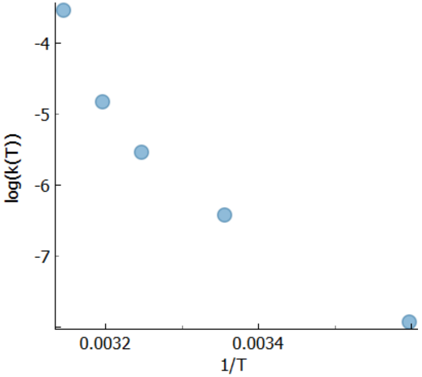


Aggregation Prediction for a mAb

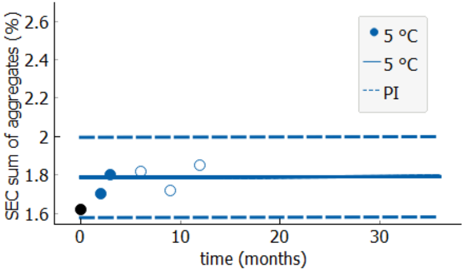
Accelerated stability data



Arrhenius plot

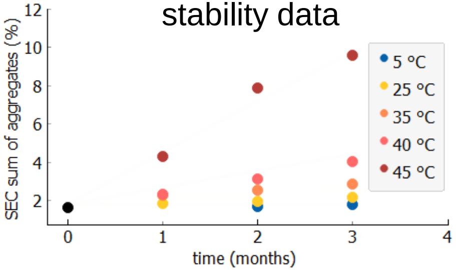


T [5 - 45 °C]
Ea = 44.9 kcal/mol

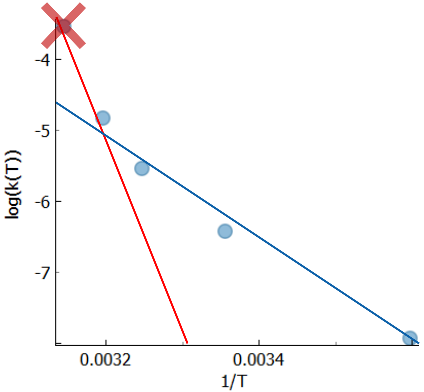


Aggregation Prediction for a mAb

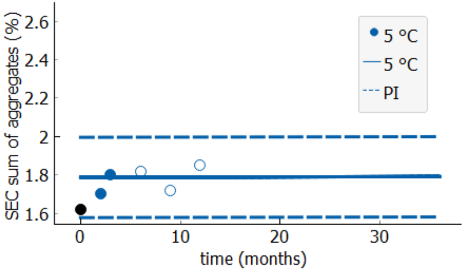
Accelerated stability data



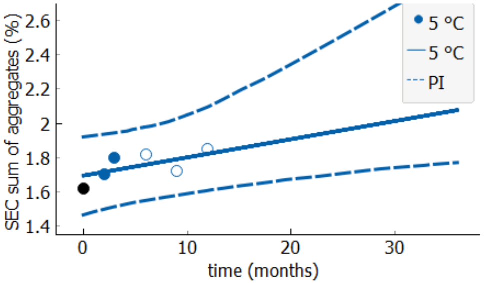
Arrhenius plot



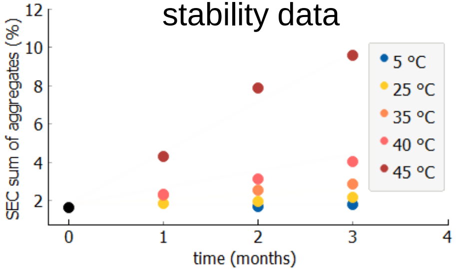
T [5 - 45 °C]
Ea = 44.9 kcal/mol



T [5 - 40 °C]
Ea = 21.0 kcal/mol

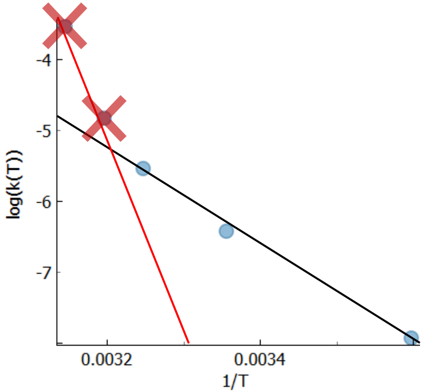


Aggregation Prediction for a mAb



Accelerated stability data

Arrhenius plot



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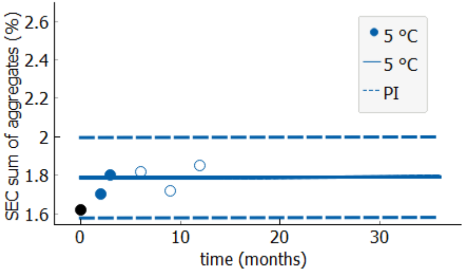
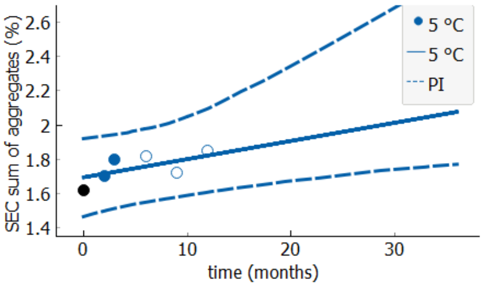
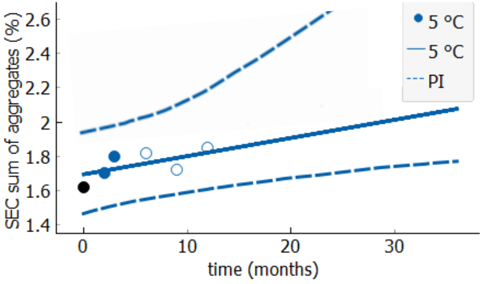


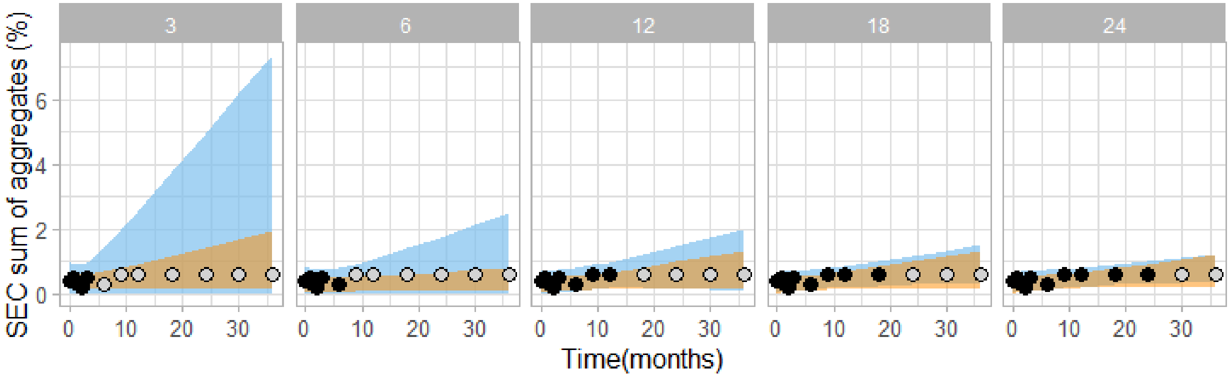
T [5 - 40 °C]
Ea = 21.0 kcal/mol



T [5 - 35 °C]
Ea = 18.3 kcal/mol



Improved robustness, speed and accuracy of stability predictions



Aggregation of Rituximab: comparison of Arrhenius kinetic model (orange) and linear extrapolation (blue) prediction intervals by using 3, 6, 12, 18 or 24 month experimental stability data (black data points) respectively. In grey, remaining measured data points at intended storage are shown for visual verification of the prediction intervals.

Sci Rep. 2021 Oct 15;11(1):20534. doi: 10.1038/s41598-021-99875-9.

Conclusions

- Simplified one-step Arrhenius-based kinetics for mAbs longterm stability predictions (6 months accelerated stability data -> 3 years prediction verified by real data)
- Arrhenius relation applies to protein aggregation, with attention to temperature range
- Practical applications: formulation development, temperature excursions
- Valuable insights for assessing shelf life and ensuring product quality and safety

Check-out publications:
2021: D. Kuzman *et al.*, *Long-term stability predictions of therapeutic monoclonal antibodies in solution using Arrhenius-based kinetics* (Sci. Reports)
2022: M. Bunc *et al.*, *Aggregation Time Machine: A Platform for the Prediction and Optimization of Long-Term Antibody Stability Using Short-Term Kinetic Analysis* (J. Med. Chem.)
2023: M. Huelsmeyer *et al.*, A universal tool for stability predictions of biotherapeutics, vaccines and in vitro diagnostic products (Sci. Reports)

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Thank you

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