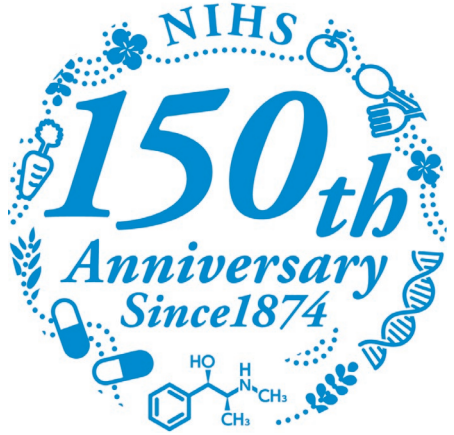
CMC Strategy Forum Japan 2023
Dec 4th, 15:40-16:00

# Current situation and issue on stability prediction of biopharmaceuticals from regulatory perspective

**Hiroko Shibata, PhD.**
**Division of Biological Chemistry and Biologicals,**
**National Institute of Health Sciences**

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# Outline

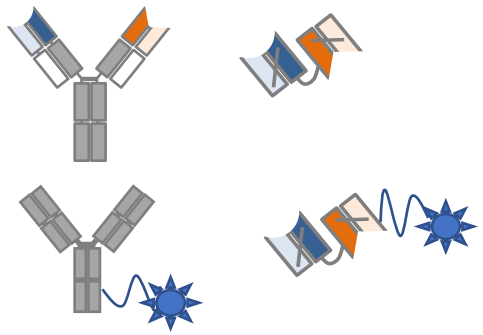
- Issues on stability evaluation in accelerated access programs
- Stability evaluation of biopharmaceuticals
- Utilization and challenges of stability prediction tools and models on biopharmaceutical development

  1. molecular design, 2. formulation screening, 3 shelf life setting

- Future prospects

# Scope

## Biotechnological and biological products



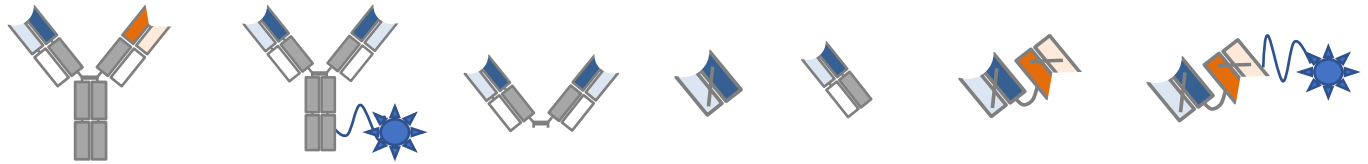
- Proteins and polypeptides, their derivatives, and products of which they are components (e.g., conjugates)
- These proteins and polypeptides are produced from recombinant or nonrecombinant cell-culture expression systems and can be highly purified and characterized using an appropriate set of analytical procedures.
(From ICH Q6B Scope)

## Biologics

Biological products include a wide range of products such as vaccines, blood and blood components, allergenics, somatic cells, gene therapy, tissues, and recombinant therapeutic proteins. (From FDA Web Site: What Are "Biologics" Questions and Answers)

# Issues on stability evaluation in early access programs

Development of next-generation antibodies in a wide variety of formats



and more

- ◆ Breakthrough new drugs are expected to take advantage of accelerated access programs such as PRIME, Breakthrough Therapies.
- ◆ On the other hand, setting the shelf life of biopharmaceuticals requires long-term stability test data at actual storage temperatures and periods.
- ◆ Setting a retest period for drug substances, or extrapolation to extend the retest period or shelf life beyond the period covered by long-term data, as is permitted for chemical products, are not permitted.
- ◆ Obtaining stability data for application may be a critical path for accelerated approval program.

（ICH Q5C: Minimum 6 months at time of application）

- ◆ In addition, stability evaluation is essential when developing formulation and manufacturing process.

⇒Extrapolation from data acquired in a short-term and construction of a predictive model are required.

# Issues on stability evaluation in early access approaches

**Workshop with stakeholders on support to quality development in early access approaches (i.e. PRIME, Breakthrough Therapies)** 26 November 2018, EMA, London
Published on 25 July 2019. EMA/CHMP/BMP/812924/2018

**Session 7a. Stability**

Standard stability requirement in line with Q5C may not be feasible for accelerated access product as this may delay the submissions**.**

**Main points identified for further follow-up:**

- **Predictive stability models**

  The proposal is interesting and may help to set a commercially acceptable shelf life to a product even if full time, product specific data have not been submitted. Further work is needed to understand the possibilities and weaknesses of the proposed model.

- **Reliance on accelerated /stress data**

  Accelerated and or stress stability data have in the past not been acknowledged to the same extent for biological products compared to small molecules. A lot of data has however been gained over the years and it would be useful to further discuss the predictability of accelerated/ stressed data to support a claimed shelf life beyond what has been shown by real time data. Stressed data may also help in understanding if the product will follow the predictive stability model as described above.

# Issues on stability evaluation in early access approaches

## EMA Draft toolbox guidance on scientific elements and regulatory tools to support quality data packages for PRIME marketing authorisation applications

2 Februay 2021 EMA/CHMP/QWP/IWP/694114/2019 Committee for Human Medical Products

**4.6.1. Stability models generated from stability of structurally similar molecules (Biotech)**

- In accelerated development programs, standard stability data packages may not be feasible and alternative paths may be needed while still assuring the stability of the product.
- For a biologic PRIME product, trends in stability data, and therefore the claimed shelf life, could be extrapolated using predictive stability models generated from prior knowledge of the stability of structurally similar molecules.
- In such cases, it may be possible to approve a shelf life which is longer than the available product-specific real time stability data.

## Concept Paper, Targeted Revisions of the ICH stability Guideline Series (Guidelines ICH Q1A-F, ICH Q5C)

- Address new technologies and modern tools/strategies used as part of enhanced product
  Modeling techniques, statistical approaches, Artificial Intelligence modeling, understanding accelerated stability conditions and prior knowledge which may support extrapolation, etc.

# Issues on stability evaluation in early access approaches

**Keywords: extrapolation, stability prediction models**

For small molecules

Extrapolation, re-test period (ICH Q1 series)

*New* Accelerated Stability Assessment Program (ASAP)

- ✓ A prediction model that adds a humidity term to the conventional Arrhenius equation for solid chemical formulations.
- ✓ Dedicated software has already been developed and it is becoming widely used.

For large molecules

- ✓ Any prediction models have not been established.
- ✓ There has not been sufficient discussion on whether extrapolation or prediction model can be used for stability testing and setting shelf life.

# Stability evaluation of biopharmaceuticals

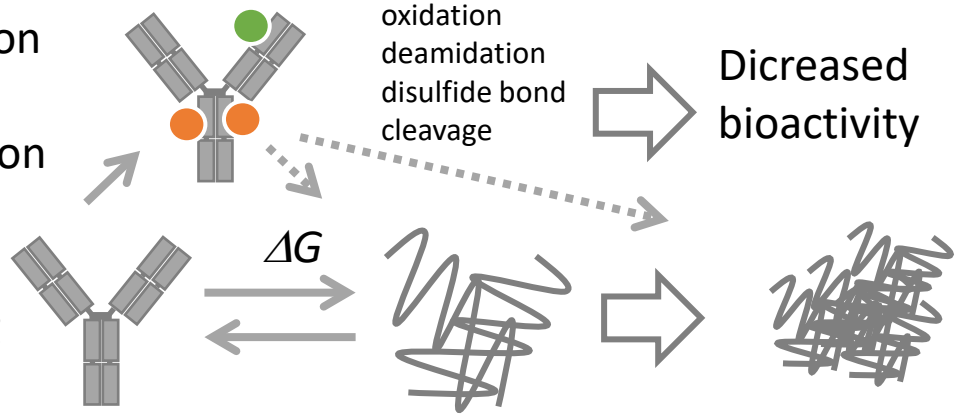
## Stability of protein in solutions

### Conformational stability

Degradation
Chemical
modification

oxidation
deamidation
disulfide bond
cleavage

Dicreased
bioactivity




**Lumry-Eyring model**

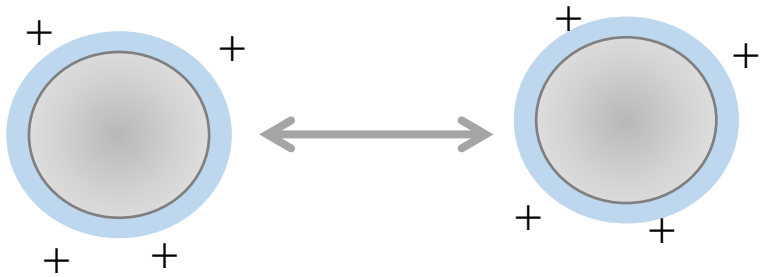
**Native** **Denatured** **Aggregates**

Various physical and chemical stresses can cause chemical and structural changes of proteins.

The larger the energy difference between the natural state and the denatured state, the lower the abundance of the denatured state and the higher the structural stability.

Physicochemical parameter: $T_m$, $T_{agg}$, $\Delta G$, $\Delta H$ *etc.*

### Colloidal stability

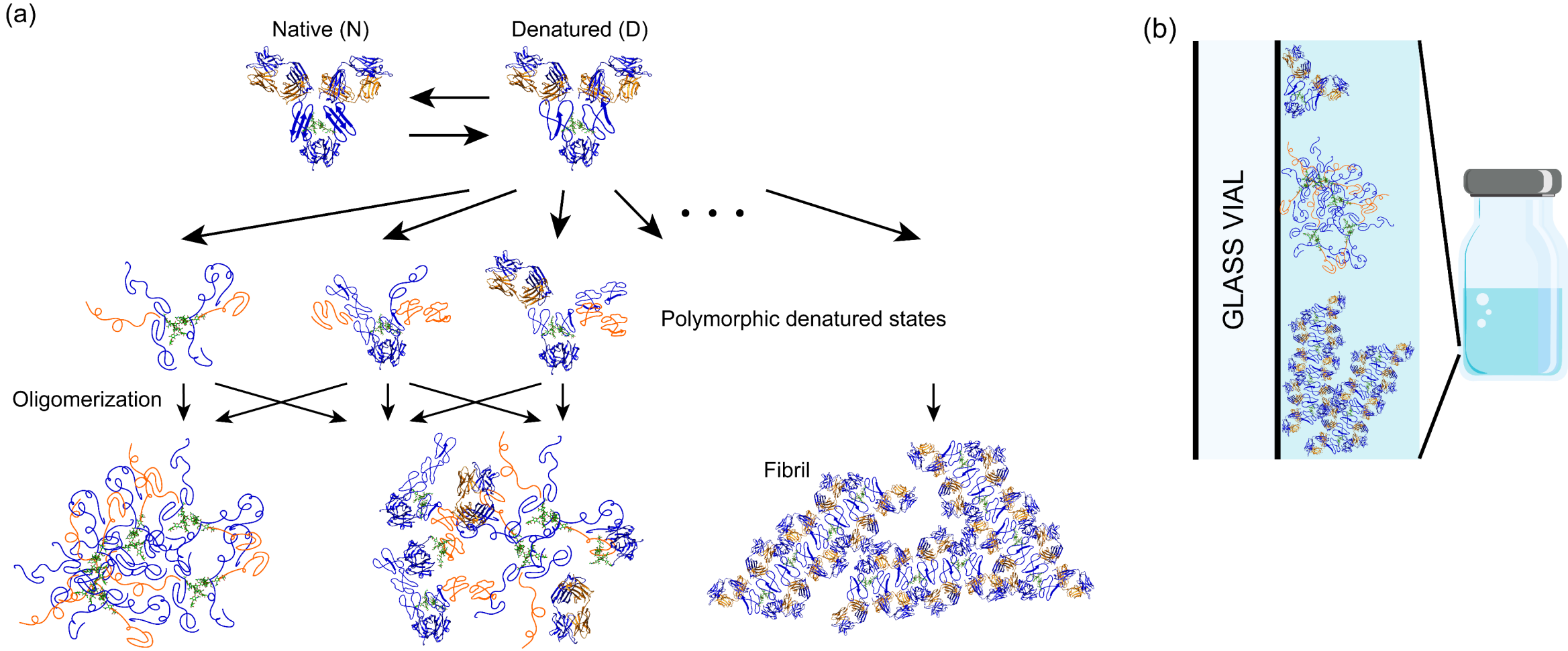


Dispersibility between molecules

Changes in H and ionic strength also affect the dispersibility of protein molecules in solution. When colloidal stability decreases, the repulsion between protein molecules becomes weaker, making it easier for molecules to form aggregates.

Physicochemical parameter: $B_2$, $K_{diff}$ *etc.*

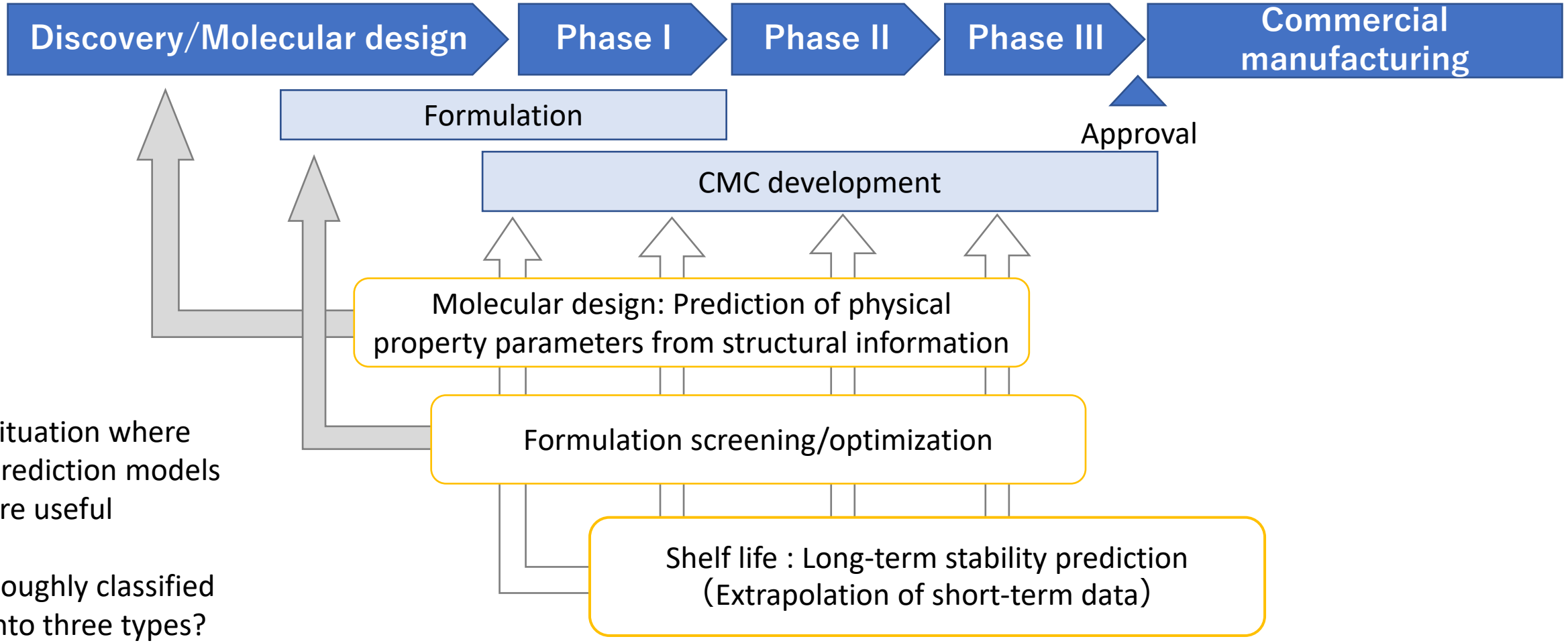
# Stability evaluation of biopharmaceuticals




Formation of protein aggregates in different pathways
(may happen in manufacturing process, storage, logistics and clinical site)

At various development stages, it is necessary to identify and evaluate risks related to the stability of biopharmaceuticals to take measures of risk reduction.

# Utilization and challenges of stability prediction tools and models on biopharmaceutical development




Situation where prediction models are useful

Roughly classified into three types?

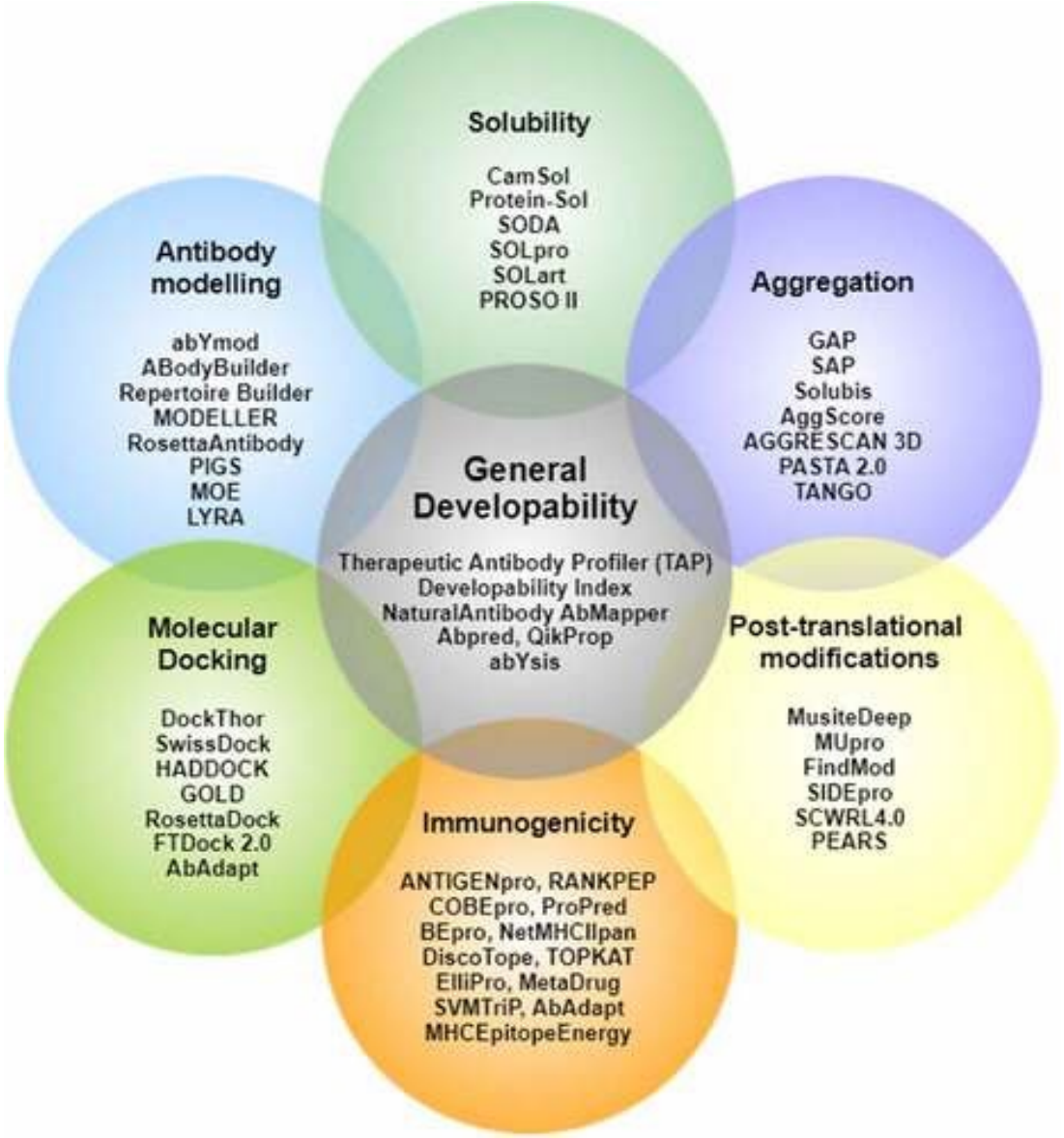
Stability prediction using in silico tools and models at each development stage

Potential to reduce stability-related risks in CMC development

# Models for molecular design

Clones with low solubility and stability,  and prone to aggregation

Risks that require significant effort and time to develop a control strategy




Advantage

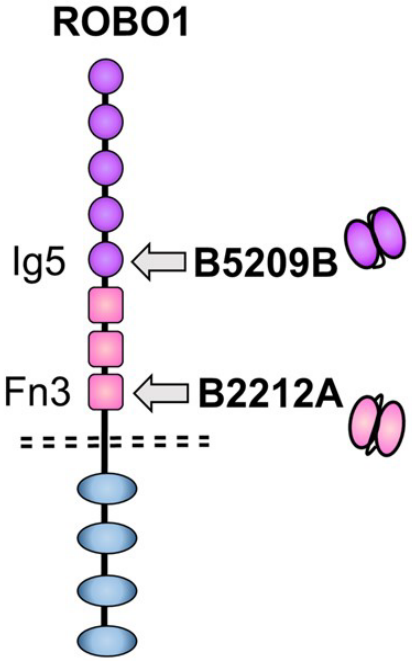
- ✓ Possibility to shorten the time required for molecular design
- ✓ Possibility to improve the accuracy of molecular design optimization for full IgG

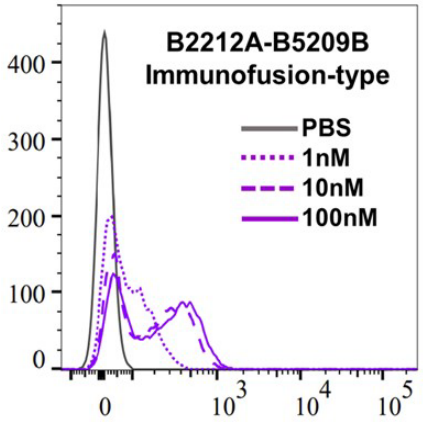
Cautions

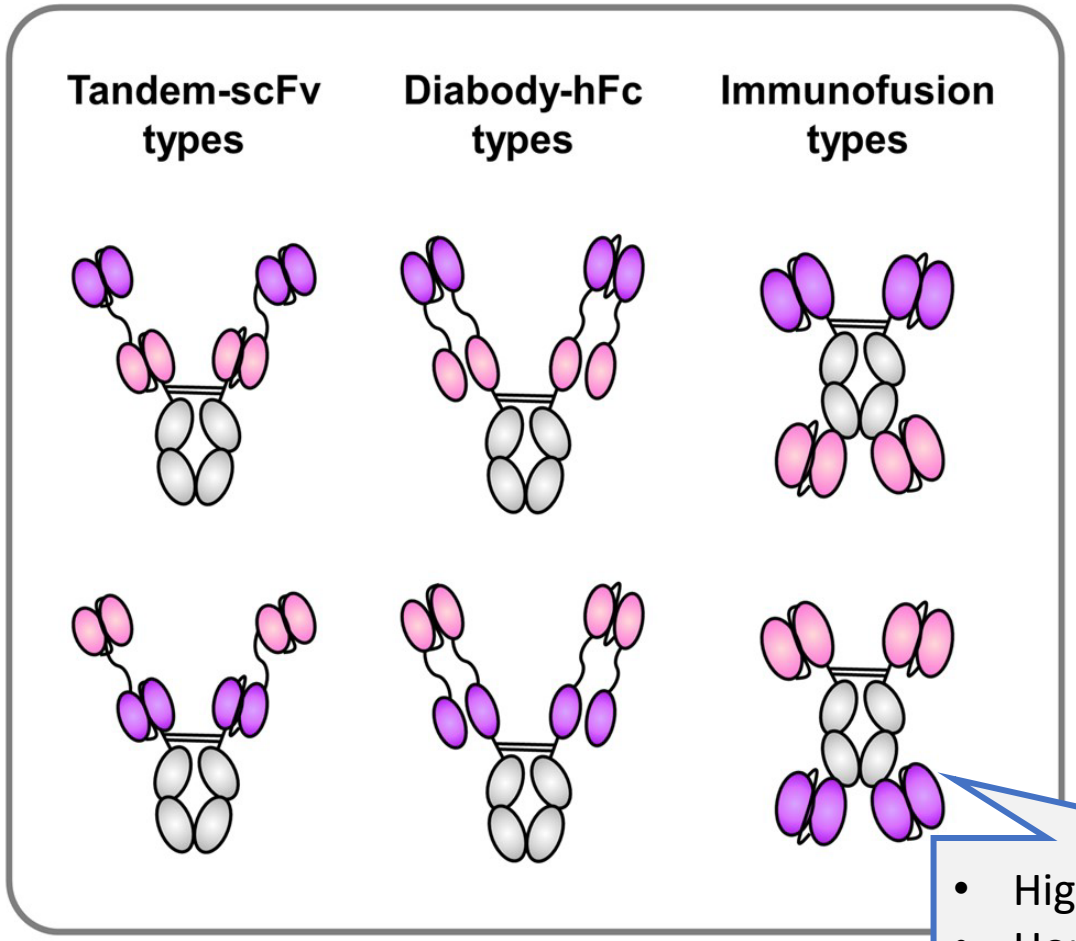
- ✓ When combining domains artificially, a next-generation antibody with the desired properties may not be obtained.
- ✓ Combining multiple domains may amplify errors caused by predictions for each domain.

MABS 2022, VOL. 14, NO. 1, e2020082 (22 pages) https://doi.org/10.1080/19420862.2021.2020082

# Models for molecular design




In our AMED research, when double paratopic antibodies were created using individually designed scFvs, the yield and thermal stability were significantly reduced depending on the length of the linker and the way the domains were combined. We also confirmed that there are cases where the desired binding affinity cannot be obtained.

- Highest yield
- Homogeneous interaction with higher affinity

Tokyo Univ. Prof. Tsumoto Lab.

J Biochem, Volume 170, Issue 2, August 2021, Pages 307–315, https://doi.org/10.1093/jb/mvab049

# Models for formulation screening

Optimization of formulations for drug substances and drug products at early stage of development

Screening index

- Conformational stability: $T_m$, $T_{agg}$
- Colloid stability: $B_2$, $K_{diff}$

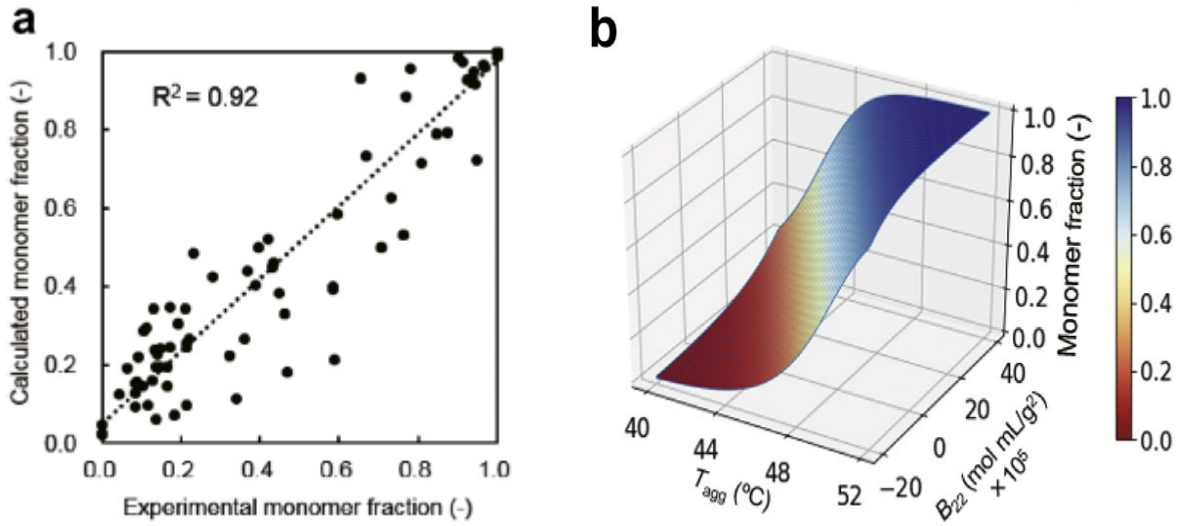
Issues related to formulation screening

- ✓ No correlation between thermal stability and colloidal stability
- ✓ No correlation in stability trends between screening parameter and actual aggregate formation

Oyama H., Koga H., Tadokoro T., et al. Relation of Colloidal and Conformational Stabilities to Aggregate Formation in a Monoclonal Antibody. J Pharm Sci. 2020 Jan;109(1):308-315. doi: 10.1016/j.xphs.2019.10.038.

**Fitting equation that can calculate aggregate formation over time at 40° C**

$$m = \sqrt[-2]{2t \cdot \exp(36.8 - 10800B_{22} - 0.871T_{agg}) + 1}$$




Features

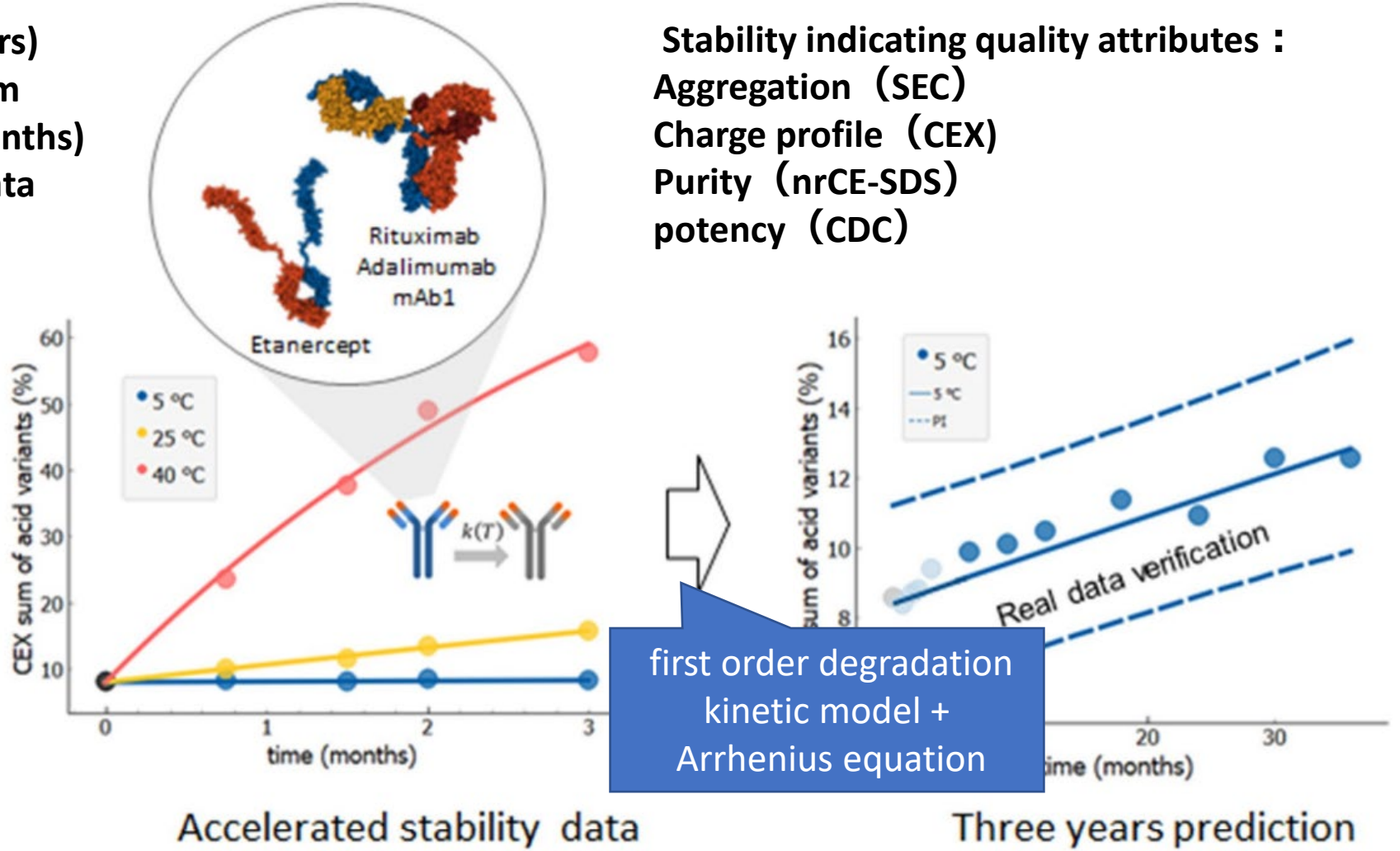
- ◆ Incorporation of $T_{agg}$ and $B_2$ into single equation
- ◆ Quantitative method to estimate the proportion of monomers at a certain time

Details in the next presentation

# Models for shelf life setting

Kuzman, D., Bunc, M., Ravnik, M. et al. Long-term stability predictions of therapeutic monoclonal antibodies in solution using Arrhenius-based kinetics. Sci Rep 11, 20534 (2021). https://doi.org/10.1038/s41598-021-99875-9

**Long-term (up to 3 years) stability prediction from short-term (up to 6 months) accelerated stability data**



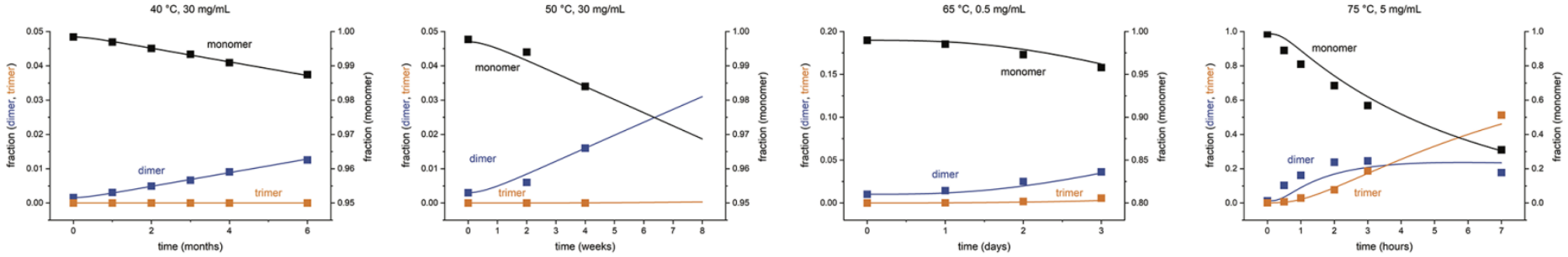

**Stability indicating quality attributes：**
**Aggregation（SEC）**
**Charge profile（CEX）**
**Purity（nrCE-SDS）**
**potency（CDC）**

Details in the next presentation

# Branched kinetics model can describe aggregate formation of mAb over a wide range of temperatures

**A** Temperature dependent aggregate formation of mAb

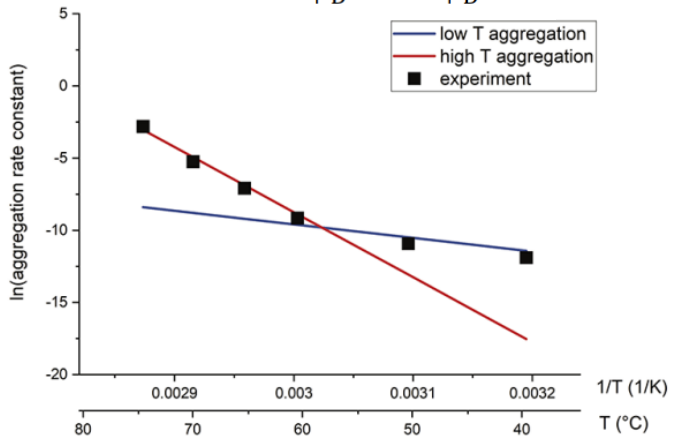
Solid lines show the fit using model C




T=40 °C > months | T=50 °C > weeks | T=65 °C > days | T=75 °C > hours

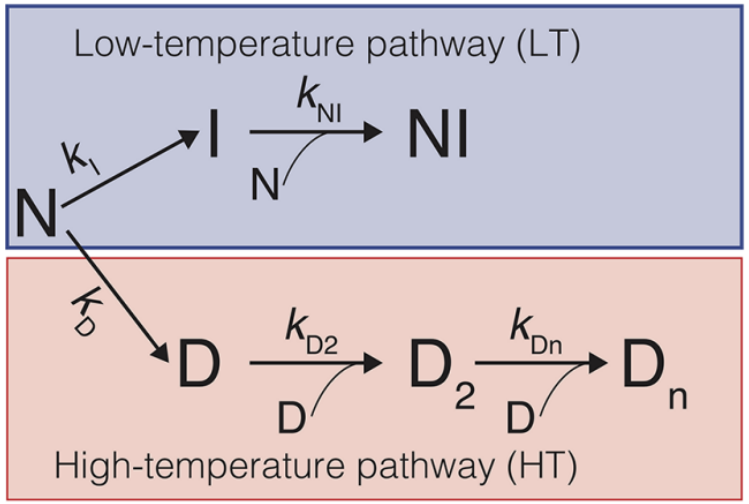
**B**

$$N \xrightarrow{k_D} D \xrightarrow[+D]{k_{D_2}} D_2 \xrightarrow[+D]{k_{D_3}} D_3$$




Arrhenius plow show s a biphasic behavior ⇒two competitive kinetic pathways with difference temperature dependencies

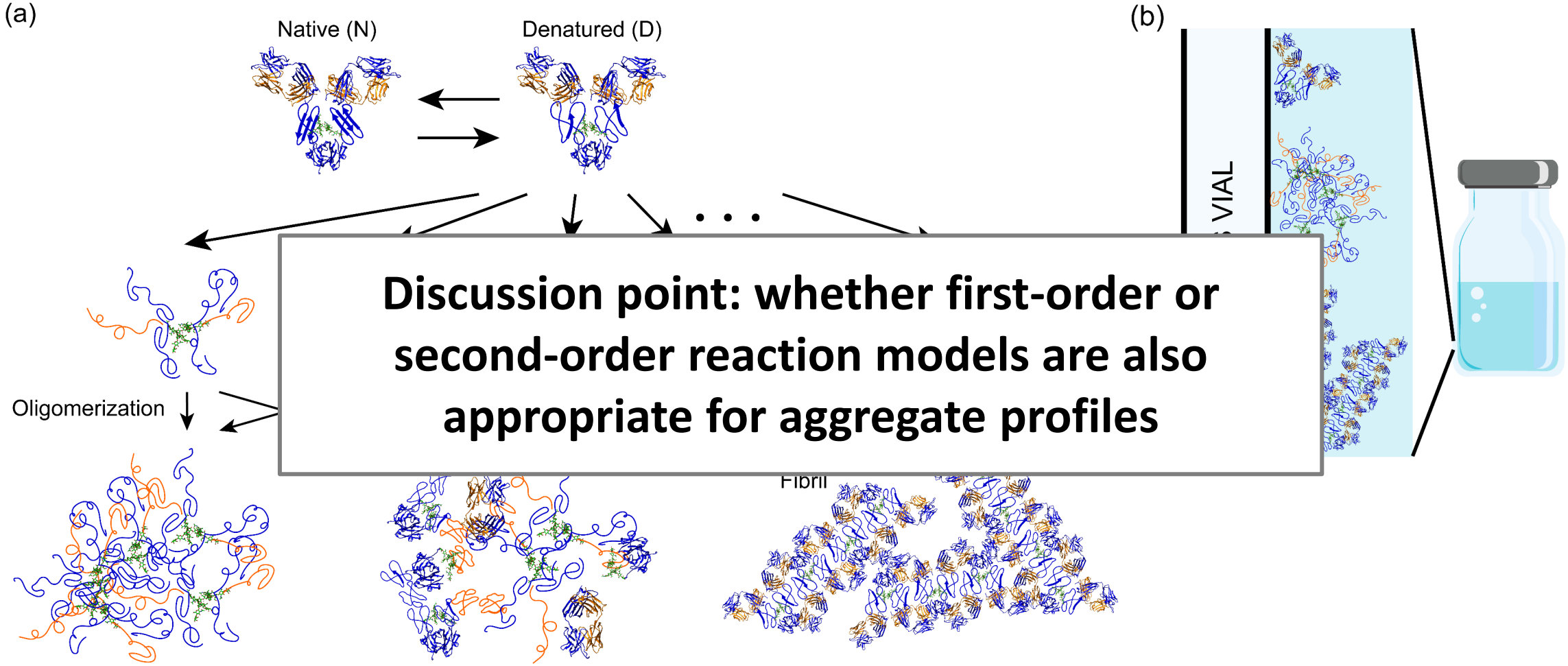
**C**




The simplest physically realistic model that successfully describes the entire range of experimental data with a single parameter set

J. Med. Chem. 2022, 65, 2623−2632 https://doi.org/10.1021/acs.jmedchem.1c02010

# Formation of protein aggregates in different pathways




**Discussion point: whether first-order or second-order reaction models are also appropriate for aggregate profiles**

Formation of protein aggregates in different pathways
(may happen in manufacturing process, storage, logistics and clinical site)

# Our experiences on stability evaluation : antibody fragments

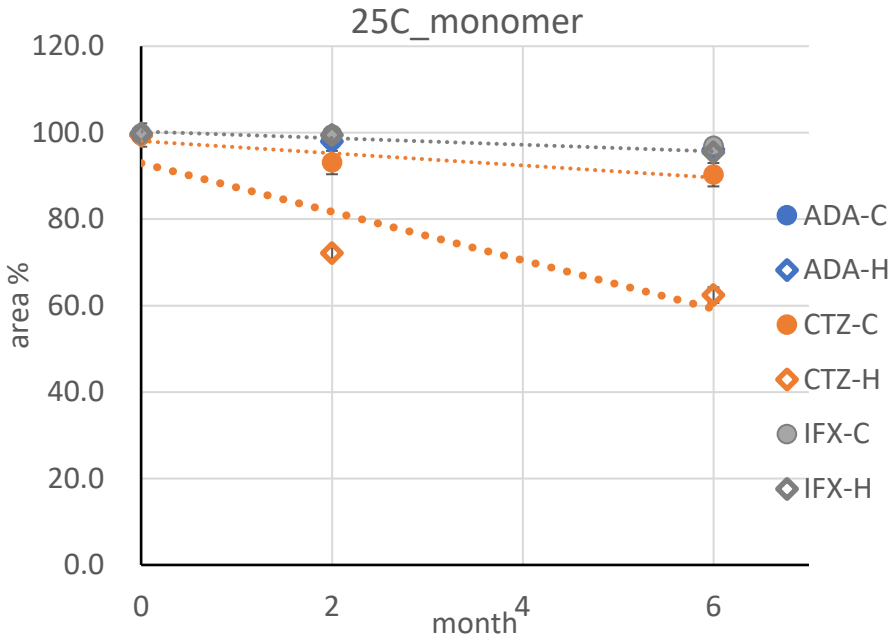
Antibody fragments did not form aggregates and only the fragmentation reaction proceeded.

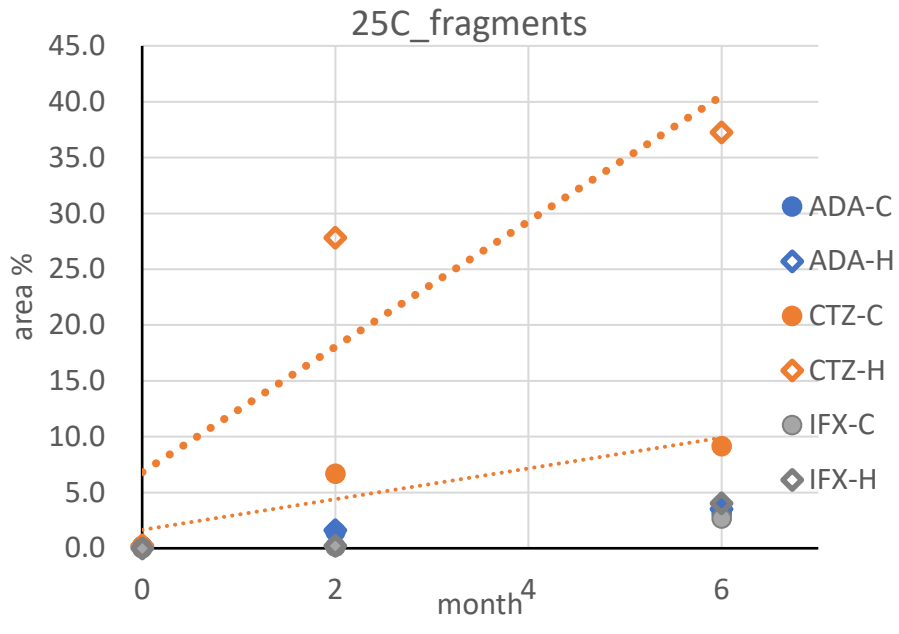
**Different reaction pathways may exist for highly engineered antibodies** such as multispecific antibodies and small-molecule antibodies

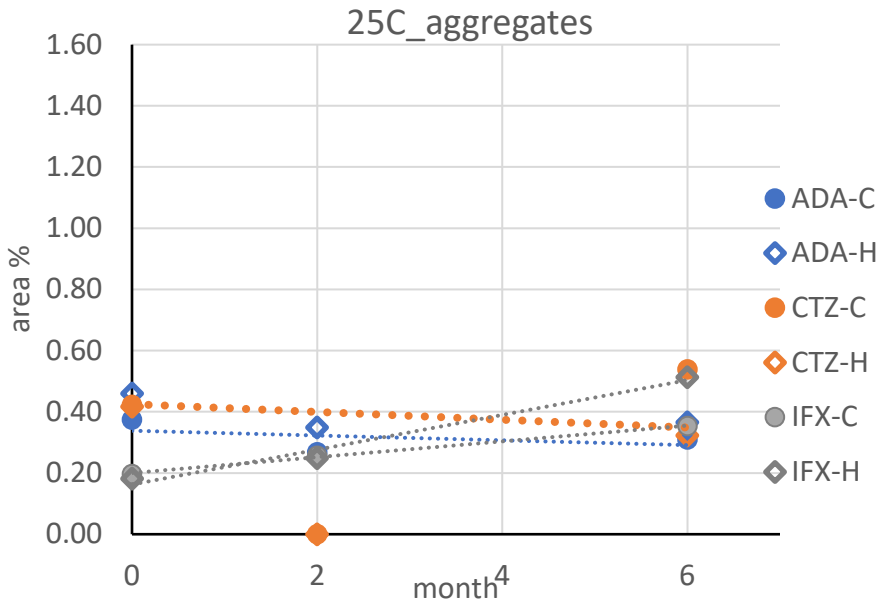
Certolizumab pegol
(PEGylated Fab')

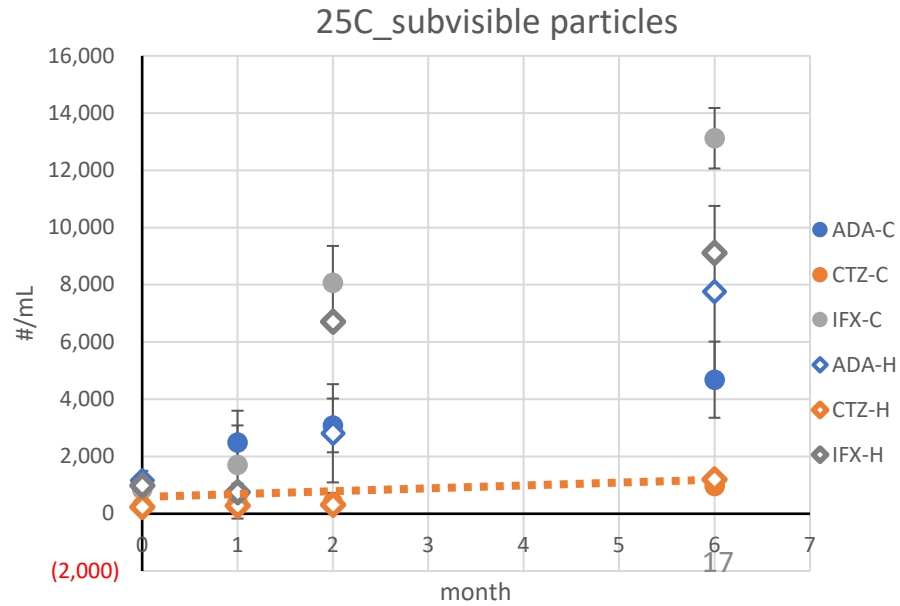
Adalimumab
(Full IgG1)

Infliximab
(Full IgG1)



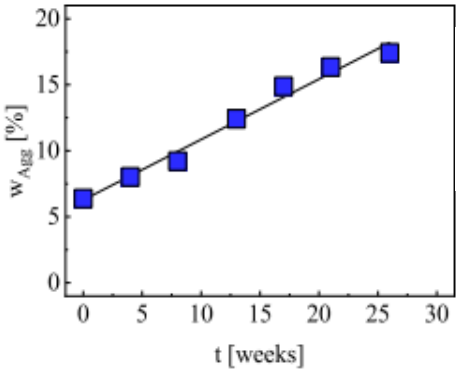
# Our experiences on stability evaluation :aggregation evaluation by SEC

## Accelerated Aggregation Studies of Monoclonal Antibodies: Considerations for Storage Stability

Ruben Wälchli [1], Pieter-Jan Vermeire [2], Jan Massant [2], Paolo Arosio [1,*]

[1] Department of Chemistry and Applied Biosciences, Institute for Chemical and Bioengineering, ETH Zurich, Vladimir-Prelog-Weg 1-5/10, 8093 Zurich, Switzerland

[2] UCB Pharma, BioTech Sciences, Formulation Development, Chemin du Foriest, 1420 Braine-l'Alleud, Belgium



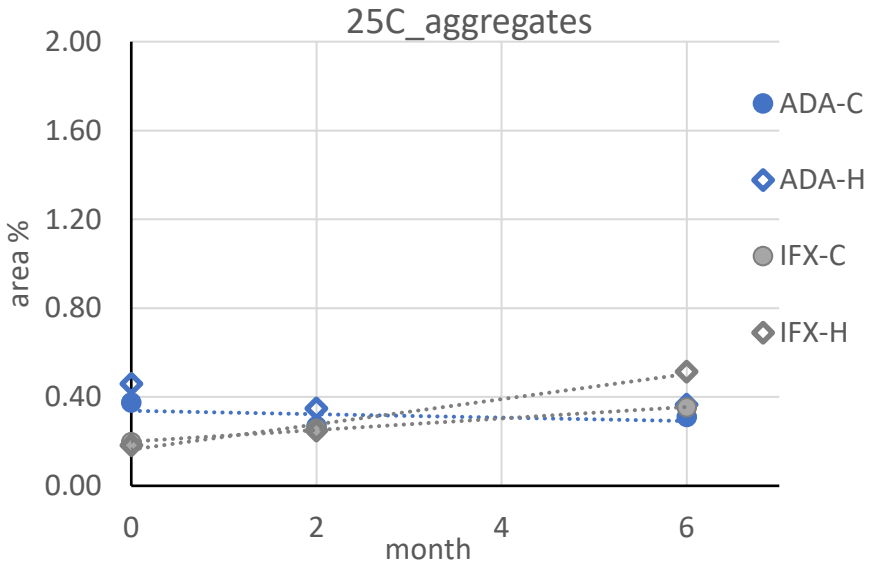

$$w_{Agg}(t) = \frac{A_{Agg}(t)}{A_{Tot}(t)} + \frac{A_{Tot}(0) - A_{Tot}(t)}{A_{Tot}(0)}.$$

Atot (0): total peak area at 0 time

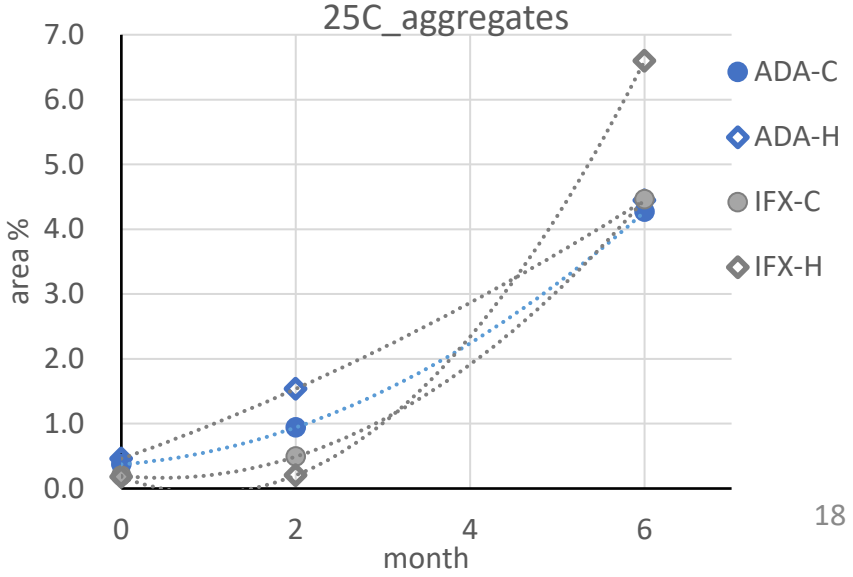
Atot (t): total peak area at certain time point

Aagg (t): aggregate peak area at certain time point

By considering the **reduction rate from the total peak area** at the 0 time point, it is possible to cover aggregates that are **difficult to evaluate by SEC**, such as adsorption to the container, precipitation, aggregates trapped in the column filter, and insoluble aggregates




considering the reduction rate of total peak area



# Models for shelf life setting

## Discussions

- Although it is necessary to verify the applicability on the molecules actually developed, we believe that predictions using a first-order or two-order reaction model that incorporates the Arrhenius equation are likely to be successful for chemical deterioration reactions such as oxidation and deamidation.
- For other quality characteristics, especially aggregates, it is necessary to discuss whether it is appropriate to apply the prediction models based on Arrhenius kinetics for all biopharmaceuticals.
- Especially in drug products, aggregates and subvisible particles are considered one of critical quality attributes. It is difficult to evaluate aggregates in the submicron ranges only using SEC.
- One solution is to measure the amount of monomer remaining without denaturation from time zero, and estimate the amount of multimers and insoluble particles formed during storage from the rate of decrease in monomer.

# Utilization of stability data obtained from similar molecules

## General approach in Japan

Prior knowledge and data that can be used when setting the shelf life of biopharmaceuticals

- ◆ Long-term stability test results for the product before the manufacturing method change

  If the formulation and storage conditions are the same and comparability has been demonstrated before and after the manufacturing process change, the results of long-term stability tests before the manufacturing process change can be used to determine the shelf life.

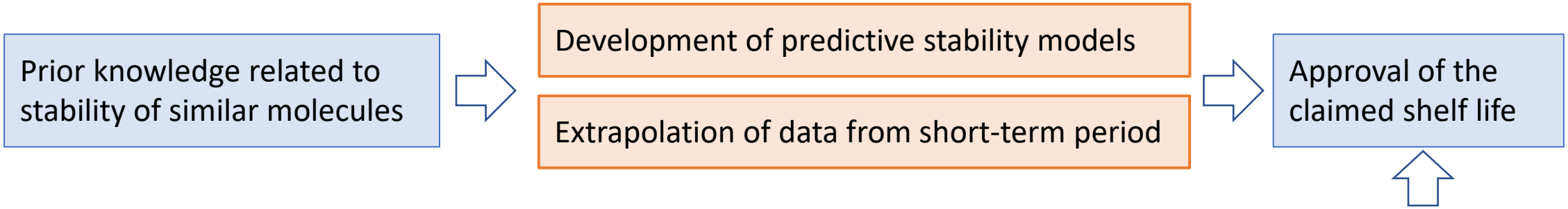
- ◆ Long-term stability test results for different content weight product of the same item

  For the item with multiple content weights, for example, long-term stability test data for the largest and smallest content products can be used to set the shelf life of the product in between.

  However, in data from long-term stability tests and accelerated tests, if stability trends differ before and after manufacturing method changes or depending on content weights, it is difficult to use prior knowledge and data even for the same item.

# Utilization of stability data obtained from similar molecules

**EMA Draft toolbox guidance on scientific elements and regulatory tools to support quality data packages for PRIME marketing authorisation applications**




- The evaluation and justification of risks in the extrapolation will be required.
- Commitments to report deviations from the expected stability trends and out of specification results will be required.
- The data used to generate the predictive stability model should be provided.
- The trend in the stability model is considered of greater importance.
- The principles will be difficult to apply to other groups of products.

Observation

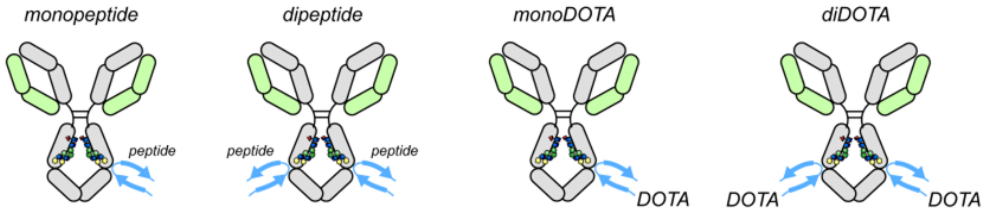
✓ This seems to be a useful approach for products with similar molecules and their sufficient prior data.

✓ However, it is necessary to discuss to what extent molecules are considered similar molecules.

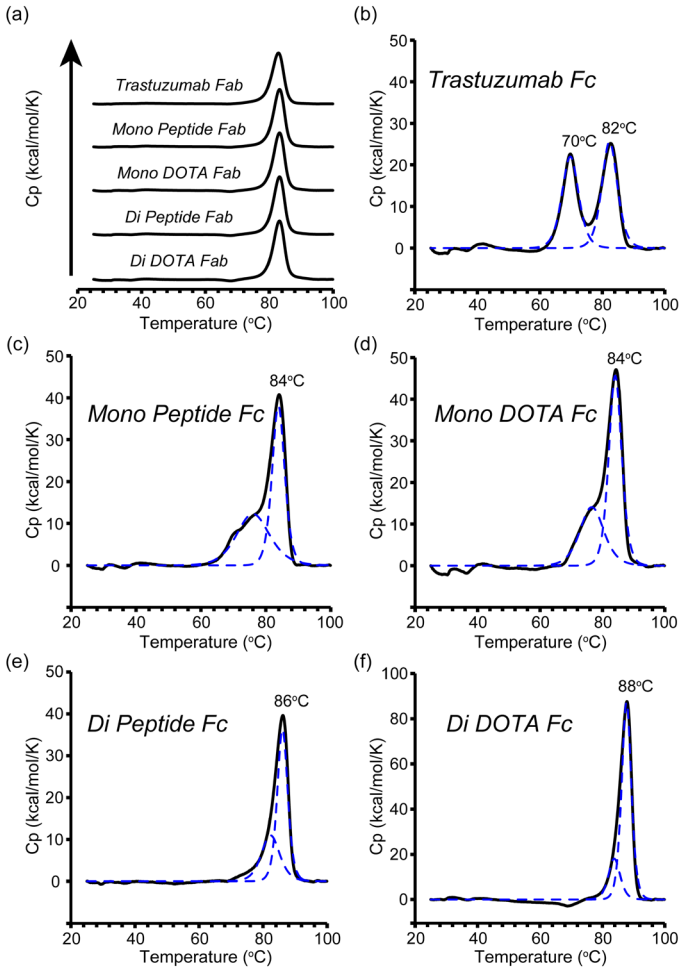
✓ The properties of next-generation antibodies vary greatly between individual molecules, and there are limits to the use of data on similar molecules.

# Our experiences on stability evaluation : next-generation antibodies

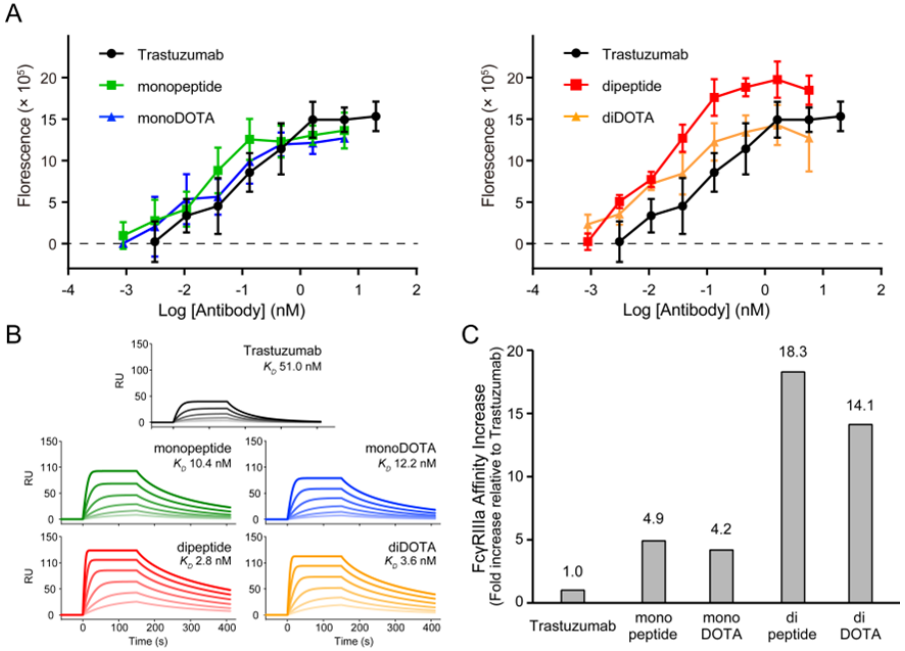
## Peptide-conjugated antibodies



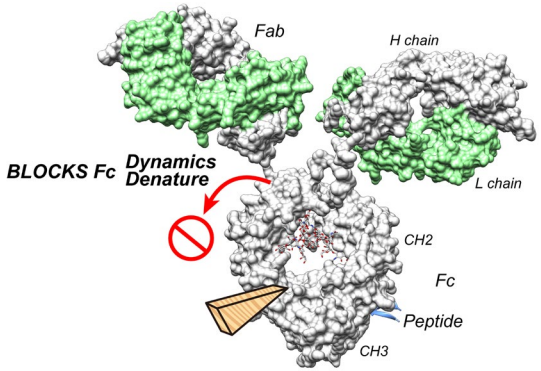

## Increased thermal stability




## Enhanced ADCC and binding affinity to FcγR




In peptide-conjugated antibodies, the thermal stability and binding affinity to Fcγ receptors change significantly just by attaching the peptide.




Kiyoshi, M., Nakakido, M., Rafique, A. et al. Specific peptide conjugation to a therapeutic antibody leads to enhanced therapeutic potency and thermal stability by reduced Fc dynamics. Sci Rep 13, 16561 (2023). https://doi.org/10.1038/s41598-023-43431-0

# Utilization of stability data obtained from similar molecules

## Discussions

- For next-generation antibodies, it is useful to establish a predictive stability model by utilizing data on the physical and chemical properties as well as data from accelerated tests and stress tests of the molecules actually developed.
- In addition to structural similarities, studies based on experimental data using multiple molecules are also desired regarding what kind of data should be accumulated in order to establish a appropriate stability prediction model.

# Future perspective

- Progress of stability prediction model using AI including machine learning
- Need to accumulate studies on stability prediction models for setting shelf life from short-term stability test data
- Possibility of mutual utilization of accumulated stability data among pharmaceutical companies, academia and regulatory agencies while assuring their confidentiality

  ⇒contribution to efficient verification on stability prediction models or extent of similar molecules
- Understanding and measures for utilization of models and AI for setting shelf life by regulatory sides

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### Current situation and issues for stability prediction of biopharmaceuticals

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