Predicting product shelf-life by using advanced kinetics and statistical analyses on forced degradation data

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Abstract

The stability of vaccines is of great interest for the vaccine industry, government institutions, and philanthropic organizations attempting to increase the distribution of vaccines to people living in countries with poor infrastructure and unreliable transportation and storage facilities. We have used protein-based vaccines, live virus vaccines, and experimental adjuvants to evaluate an advanced kinetic modeling approach. This approach uses a systematic and simple procedure for the selection of the most appropriate kinetic equation to determine the degradation rate of compounds due to accelerated temperature exposure. One-step and two-step reactions with unlimited combinations of kinetic models were screened for the products under evaluation. The most appropriate mathematical model for each product is chosen based on the values of residual sum of squares and weight parameters. Then, long term reaction progress was determined and statistical analysis was additionally performed to define accuracy of predictions. Depending on the complexity of degradation pathways, relatively simple n-th order kinetic model or more complex two-step model were required to fit the degradation of products. Generally, a prediction error lower than 10% was obtained. [1]

The modeling approach described here could be used for multiple purposes: stability prediction for expiry date estimation, temperature excursion during transportation, formulations ranking, batch to batch comparability and support to manufacturing process changes. To the best of our knowledge, this is the first procedure mixing a global kinetic approach and statistical analyses to accurately determine a vaccine degradation rate from product exposure to temperature greater than those recommended for storage.

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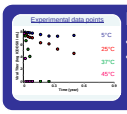

Design forced degradation study

- 20 to 30 experimental data points
- At least 3 different temperatures (5°C, 25°C, 37°C, ...)

2

Screen infinite number of models

- Run the fitting procedure including *one-step* and *multi-steps* equations (truncated Šesták-Berggren formula)
- Ockham razor principle is applied All the models are ranked according to an automatic statistical analysis

3

Rank kinetic models

- The best / simplest model is identified according to the higher AIC (Akaike) and BIC (Bayesian) statistical scores
- Then, predict reaction progress in any time/temperature domains: long term reaction progress at recommended storage temperature (stability), excursions of temperature, ...

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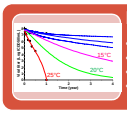


Determine accuracy of predictions

- Predictive bands (CI 95%) obtained by statistical analysis (boostrap)
- Shelf-life evaluation
- Batch to batch comparison according to their long term stability
- Formulations ranking

Advanced kinetics and statistical analyses

The *AKTS-Thermokinetics* software considers a non-limited amount of models using "one step" (n-th order, autocatalysis) and "two steps" (combinations of one step) kinetics as proposed in B. Roduit *et al.*, 2014. [2] An universal equation is used to automaticaly fit forced degradation data. By changing the value of the parameters n1, m1, n2, m2 the number of possible combinations becomes infinite.

Models are ranked based on Akaike (*AIC*) and Bayesian (*BIC*) statistic criterion. Those considerations are translated in a *w* index (*w* for 'weight') between 0 and 1 attributed to each model combination, with the sum of individual *w* values equal to 1. The model combination that exhibits the highest *w* value is chosen to describe the kinetic reactions.

It is then used to extrapolate the long term stability behavior of the product under any storage temperature. The confidence intervals was calculated according to a bootstrap analysis. This statistical approach, which is based on data re-sampling, was applied with 1000 loops and provided confidence intervals in the form of upper and lower 95 percentiles (P.B. 95%) of each of the fitted parameters.

$$\frac{d\alpha}{dt} = A_1 \exp\left(-\frac{E_1}{RT}\right)(1-\alpha)^{n1}.\alpha^{m1} + A_2 \exp\left(-\frac{E_2}{RT}\right)(1-\alpha)^{n2}.\alpha^{m2}$$

One step

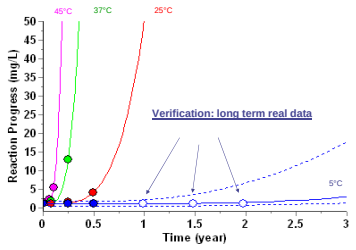
0 order
1st order
2nd order
3rd order
4th order
...
n-th order
Perez-Maqueda

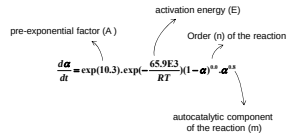
Two steps

Two *n-th* order
Sourour Kamal
...and more

1. Identify convenient storage condition

- Oil oxidation followed by HPLC up to 6 months
- An autocatalytic type kinetic model was identified as expected for oxidation reaction
- Product exhibits 2 years stability at 5°C and a sensitivity to higher storage temperature

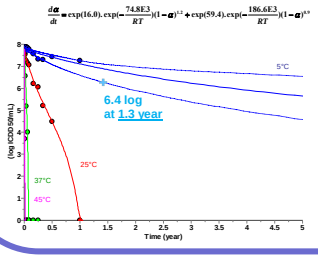




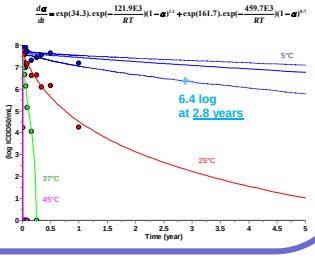
2. Formulations ranking

- Virus-based vaccines: loss of viral activity followed by CCID50 infectious titer for a formulation with or without stabilizer
- Two-steps kinetic model identified in agreement with literature [3]
- Considering 6.4 log as the lowest acceptable value, shelf life would be around 2 x in presence of stabilizer excipient (formulation B)

Formulation A



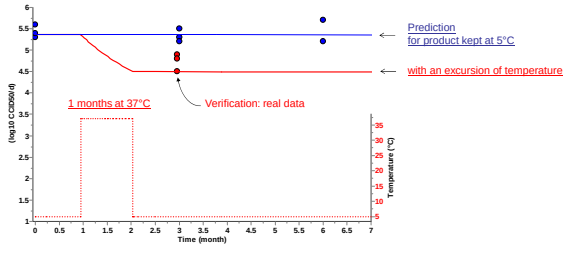
Formulation B



3. Impact of excursion of temperature

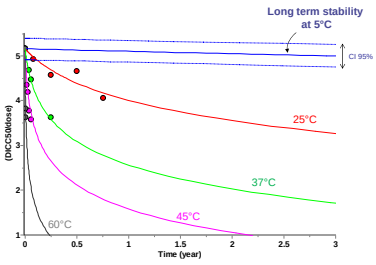
- Infectious titer for virus based vaccine
- A one-step model (third-order) is determined to describe degradation reaction
- In agreement with real stability data, kinetic model predicts loss of viral titer after one month excursion at high temperature (37°C)

Best kinetic model $\frac{d\alpha}{dt} = \exp(36.6).\exp\left(-\frac{136.1E3}{RT}\right)(1-\alpha)^3$



4. Getting more confidence in shelf-life extension

- Loss of viral activity followed by CCID50 infectious titer in forced degradation conditions (above ambiant temp.)
- One-step kinetic model identified (n^{th}-order reaction)
- Kinetics and statistical analyses predict several years of stability at 5°C and a high sensitivity at 25°C



Best kinetic model $\frac{d\alpha}{dt} = \exp(37.2).\exp\left(-\frac{130.5E3}{RT}\right)(1-\alpha)^{1.54}$

Conclusion

We used a novel approach to analyze the experimental data, to fit them by computed kinetic parameters, and finally, to predict the long term stability of products. Presented results indicate that shelf-life of vaccines can be predicted with accuracy. This procedure was applied to evaluate the degradation kinetics of protein and viruses based vaccines, and adjuvanted formulations. Accelerated stability studies at three or more isothermal temperatures were conducted and the resulting data were fitted with one-step or two-steps kinetic models. The most appropriate model was identified by statistical analysis and was afterwards used to extrapolate the long term degradation at any required storage temperature. It is important to mention that the selection of the kinetic models is based purely on statistical analysis and goodness of fit without requiring that the chosen model is established as correctly describing the mechanism(s) of degradation of the complex systems under evaluation. Presented kinetic approach could be employed for many practical applications, including shelf life prediction, estimation of impacts of excursions of temperature from the "cold chain", lot-to-lot comparison, and evaluation of stabilizers and antimicrobials for biologics and vaccines during formulation development. Future studies should look into the contributions and interactions of individual components in the vaccine to the overall kinetics of degradation of the vaccine as a whole.

[1] D. Clenet *et al.*, Advanced Kinetic Analysis as a Tool for Formulation Development and Prediction of Vaccine Stability, J. PHARM. SCI. 103:3055–3064, 2014
[2] B. Roduit *et al.*, Prediction of thermal stability of materials by modified kinetic and model selection approaches based on limited amount of experimental points, Thermochimica Acta 579: 31–39, 2014
[3] Higashikawa F, Chang L *et al.*, Kinetic analyses of stability of simple and complex retroviral vectors. Virology 280:124-131, 2001

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