Degradation and Stability Profiling in Pharmaceutical Analysis: Review

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ABSTRACT: Forced degradation is a degradation of new drug substance and drug product at conditions more severe than accelerated conditions. It is required to demonstrate specificity of stability indicating methods and also provides insight into degradation pathways and degradation products of the drug substance and helps in elucidation of the structure of the degradation products. Forced degradation studies show the chemical behavior of the molecule which in turn helps in the development of formulation and package. In addition, the regulatory guidance is very general and does not explain about the performance of forced degradation studies. Thus, this review discusses about the current trends in performance of forced degradation studies by providing strategy for the conduct of studies, degradation mechanisms and also describes the regulatory aspects of force degradation and the study of stability and also the analytical methods helpful for development of stability indicating method.

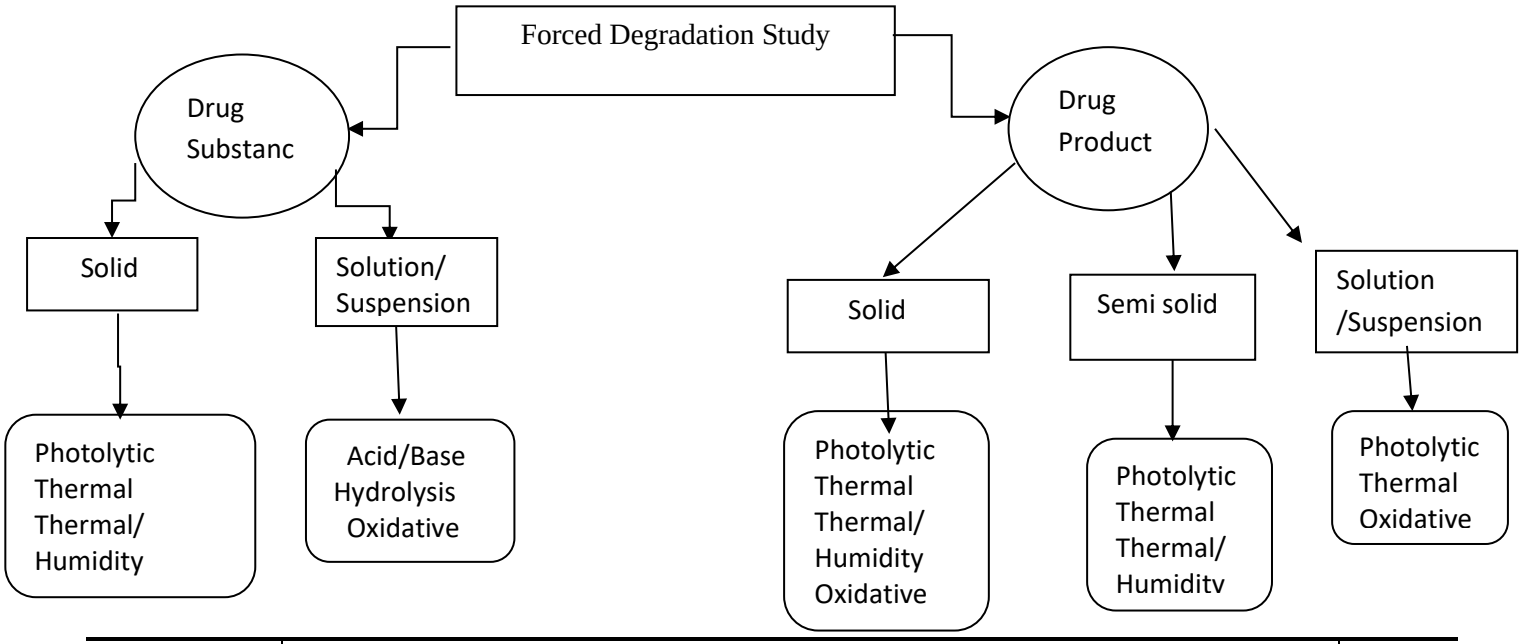
KEYWORDS: Forced degradation, Stability indicating methods, ICH guidelines

INTRODUCTION:

Chemical stability of pharmaceutical molecules is a major concern it has an impact on the efficacy and safety of the drug product. The FDA and ICH guidelines state need for stability testing data to realize how a drug substance's and drug product's quality changes across time under the influence of numerous ecological conditions. Assessing a molecule's stability aids in determining the ideal formulation and package as well as providing proper storage conditions and shelf life, this is vital for regulatory certification. Forced degradation is a process that involves degradation of drug products and pharmacological compounds at conditions that are more severe than accelerated conditions and thus generates degradation products that can be studied to determine the stability of the molecule.[1]

Studies on forced deterioration are conducted to meet the following goals:
1. To establish drug substance breakdown pathways and drug supplies.
2.To differentiate degradation products that are related to drug items made from those derived from non-drug products when formulating.
3. To clarify the composition of degradation products.
4. To determine a medicinal substance's intrinsic stability in formulation [2]

Fig (1) Forced degradation can be studied with the help of following flow chart-[3]



Degradation Conditions:

Conditions Usually Applied For Forced Degradation Studies (Table1)[4]

Degradation type	Experimental Conditions	Storage Conditions	Sampling time(Days)
Hydrolysis	Control API 0.1 M HCl 0.1M NaOH Acid/ base Control Ph : 2,4,6,8	40^0C, 60^0C 40^0C, 60^0C 40^0C, 60^0C 40^0C, 60^0C 40^0C, 60^0C	1,3,5 1,3,5 1,3,5 1,3,5
Oxidation	3%H_2O2 Peroxide Control Azobisisobutyronitrile (AIBN)	25^0C, 60^0C 25^0C, 60^0C 40^0C, 60^0C	1,3,5 1,3,5 1,3,5
Photolytic	Light 1^xICH 3^x ICH Light	NA NA NA	1,3,5 1,3,5 1,3,5
Thermal	Heat Chamber Heat Chamber/RH Heat Chamber	60^0C 60^0C/75%RH 80^0C	1,3,5 1,3,5 1,3,5

Stability studies and their classification:

Stability studiesare critical criteria for ensuring the final product's quality, efficacy, and integrity.

1) Physical stability studies:

The physical assessment of the solution is especially significant for intrathecal, ophthalmic, and intra-arterial routes. Additionally, the bodily changes may be harmful. Physical stability studies are particularly important because a tablet's bioavailability may suffer if it becomes soft and unsightly or extremely hard and shows a very sluggish rate of disintegration. Therefore, a more in-depth physical investigation is crucial to determine the kinetic patterns of aggregation of therapeutic proteins utilizing turbidimetry, light blockage, dynamic light scattering, or microscopic examination.[3]

2) Chemical Stability Studies:

Moisture is a common reactant in chemical processes and frequently serves as the solvent vector. Because moisture has stronger thermal conductivity than solids, which allows for better heat transmission, molecules have more kinetic energy and more breakdown is seen. Moisture is the root cause of all of these processes, including hydrolysis, oxidation, and fermentation.

3) Microbiological stability studies:

Even though many natural polymers are the origin of microorganisms, as microorganisms not only affect or assist in making moisture but also solid dosage forms made from naturally occurring polymer.[5]

Regulatory guidelines for stability testing [6]

Guideline	Title
Q1A(R2)	Stability testing of new drug substances and products
Q1B	Stability testing: photo stability testing of new drug substances and products
Q1C	Stability testing for new dosage forms
Q1D	Bracketing and matrixing designs for stability testing of new drug substances and products
Q1E	Evaluation for stability data.
Q1F	Stability data package for registration applications in climatic zones iii and iv.
Q5C	Quality of biotechnological products: stability testing of Biotechnological/ biological product

ICH Q1A summary of stability parameters [7]

Study Type	Condition	Storage Condition	Period In Months	Comments
General Case	Long-term Intermediate	25°C±2°C/60% RH±5% RH or 30°C±2°C/65% RH±5% RH 30°C±2°C/65% RH±5%	12 6	Must cover retest or shelf-life period at a minimum and includes storage, shipment and subsequent use
Refrigeration	Long-term Accelerated	5°C±3°C 25 °C±2°C/60% RH±5% RH	12 6	Must cover retest or shelf-life period at a minimum and includes storage, shipment and subsequent use.
Freezer	Long term	-20°C±5°C	12	Must cover shelf life period at a minimum and includes storage, shipment and subsequent use.

Stability testing method:

\ At various stages of drug substance product development, stability testing is a standard procedure used. In the beginning, an accelerated stability study (at relatively high temperatures and/or humidity) is carried out to assess the type of degradation products that may be produced after long-term storage. It is advised to test a product's long-term shelf life in exacting circumstances, such as at a high temperature, in order to determine its shelf life and expiration dates. The main goal of pharmaceutical stability testing is the provision of an acceptable declaration that the products will continue to be at an acceptable level of fitness/quality for the duration that they are on the market and available for supply to the patients, and will be fit for their consumption until the patient uses the last unit of the product.The following categories of stability testing techniques have been established based on the purpose and measures taken.

1) Real-Time Stability Testing:

Real-time stability testing includes longer-term deterioration of the test medications to enable degradation under advised storage settings. The test's duration is determined by the product's stability, which should be lengthy enough to reliably demonstrate the absence of quantitative degradation and distinguish it from inter-assay variation. To distinguish between daily uncertainty and instability, data is gathered during testing at an appropriate frequency. The addition of a single batch of a reference material with known stability properties can improve the accuracy of data interpretation.

2) Accelerated Stability Testing:

During accelerated stability studies, a material is subjected at a specific high temperature (hot than ambient) to ascertain how much heat is generated. Required to degrade the product. The comparison of alternative options' relative stability. The expected formulas and shelf life follows. Temperature and moisture combined, Gravity, agitation, pH, light, and other factors are the strain circumstances put in place during accelerated stability evaluation. The samples in this technique are simultaneously assessed and subjected to stress, then chill after stressing. The comparatively, the measurement system is more modest because of the real-time stability testing. The analysis only lasts a brief time. Additionally, the same assay is used to compare the unstressed product to the stressed material, and the stressed sample recovery is represented as a percentage of the unstressed sample recovery. By avoiding denaturing stress temperatures, projections of the stability of thermolabile and proteinaceous components are generated with a fair amount of accuracy. It is advised to carry out the accelerated stability projections at four distinct stress temperatures for statistical reasons.

3) Retained Sample Stability Testing:

In this investigation, at least one batch every year is chosen for stability sample retention storage. When there are more than 50 batches being marketed, stability samples from two batches are advised to be taken. When a batch is first released into the market, stability samples may be taken; thereafter, this may be reduced to only 2 percent to 5 percent of marketed batches. For example, if a product has a shelf life of five years, it is routinely tested at three, six, nine, twelve, eighteen, twenty-four, thirty-six, forty-eight, and sixty months. The constant interval method is a common technique for obtaining stability information on samples kept in storage. This approach is more practical because it tests the product in both the real market and the idealized conditions for storing retained samples.

4) Cyclic Temperature Stress Testing
This is not used as a standard testing procedure for products that are advertised. Product knowledge is used to construct cyclic temperature stress tests that imitate similar market conditions for storage. Since the earth has a 24-hour day and night cycle, this is also the design time that most commercially available medications are most likely to experience while in storage. The lowest and maximum temperatures for the cyclic stress testing are chosen based on the specific physicochemical features of each product as well as other significant considerations like recommended storage temperatures. Typically, 20 cycles have been advised.[8,9]

Factors influencing stability of dosage form:
1) PH
This is not used as a standard testing procedure for marketed items. Storage cyclic temperature stress tests are designed with the goal of simulating similar market conditions. Since the earth's diurnal rhythm lasts for 24 hours, this is also the length of most cycle designs, which is what makes medications on the market most susceptible to odour during storage. The lowest and maximum temperatures for the cyclic stress tests are chosen based on a product-by-product basis, as well as significant considerations like recommended storage temperatures and particular physicochemical degrading features of the products. 20 cycles have often been advised.

2) Temperature
It is one of the most important elements in the stability of drugs. The pace of the degrading reactions may increase by 2 to 5 times with an increase in storage temperature of roughly 10°C. For some compounds, the optimal temperature range for physicochemicalstabilityis only a small range; outside of this, significant degradation is shown. For the majority of active components, the kinetics of degradation processes adhered to the Arrhenius law. The stability of the formulation at room temperature can therefore be determined by conducting stability studies at high temperatures (at 40°C, for example).

3) Surfactant
Although various surfactant types (anionic, cationic, or nonionic) create micelles in solution, this entanglement of the active ingredient the bioavailability of component molecules changes a resolution. The usage of the surfactants protect the active ingredient and reduce its deterioration an active component in hydrolytic groups like hydroxyl.

4) Oxygen
Instability may result from the oxidation of one of the drug's components caused by oxygen in the preparation. Use of antioxidants and appropriate production Techniques like those used under nitrogen are crucial. An a suitable container with its guaranteed integrity essential components for stopping the incursion over time, of oxygen.

5) Light
An significant aspect is that light can make photosensitive compounds chemically unstable. If precautions are taken, especially during manufacturing, such as the choice of suitable packing material, it is preventable, and It is crucial to make sure they are upheld over time.

6) pH Rate Profile
The pH-rate profile, also known as pHstability, is the relationship between a compound's particular rate constant of degradation and pH. It is called a profile, rate-pH profile, or easily shown by a log(k) versus pH plot. The pH-rate profiles assist with creating formulations for more reliable solutions and lyophilize items also offer perceptions into the catalytic character of a response.[10,11]

Degradation prediction tools:

1) CAMEO
A computer programme called CAMEO (Computer Aided Management of Emergency Operations) makes predictions about the end results of chemical processes based on the circumstances, reagents, and starting components. The studies include the following crucial degradation circumstances: acidic/electrophilic, radical, and basic/nucleophilic additionally to oxidative/reductive, photochemical, and mechanical explanations for these responses. In The CAMEO algorithms have generally been made to correct product mixes that are off forecasting greater amounts of breakdown byproducts observed.[12]

Analytical tools used in stability indicating method development:
The development of stability indicating methods is made simpler by advancements in analytical instrument techniques. The modern techniques the medicine and its separation must be effective. Material, its byproducts that degrade and its impurities Additionally, it should have excellent sensitivity and great precision while evaluating psychoactive substances.[13]

1) HPLC:
The separation of the peak for the active ingredient from the peak for the degradation product, as well as the detection of the same. Whenever a sample is created using an appropriately constructed and implement forced and can be used to construct the LC during degradation. Method. The many grounds for separation depending on the solvent type and mobile phase pH, the chromatographic method, temperature and type of column. Analysis solubility utilized buffer, solvent's UV value, and the security of solvent are the

criteria for choosing a solvent. In the stability-indicating assay, systematic and planned evaluation of experimental parameters including pH, mobile phase flow rate, column type and temperature, chromatogram mode, sample concentration and amount injected, solvent employed during this method development.

2) Calorimetry:
A chemical assay of samples kept at high temperatures for proper analysis is typically part of the methodology for accelerated stability testing. Periods. Mostly driven by the desire to intensify sample processing and thermal analysis particularly differential scanning methods in studies, calorimetry (DSC) has been used. On the kinetics of explosives' breakdown and research on the stability of pharmaceutical solids. The sensitivity restrictions, however, necessitate high temperatures in both scanning mode and irradiation mode.

3) Chemo metric Method:
This approach uses UV- spectrophotometry as its foundation, and chemo metrics is used to process the ensuing, highly overlapping responses. In this approach; various chemo metric techniques were used to simultaneously determine the presence of medicines and their products of degradation, such as PCR and PLS methods. Multivariate calibrations were used in these helpful in the spectrum analysis since the simultaneous use of several spectral data strongly prefer wavelengths to single wavelength increased predictability and accuracy. For assessment of the predictability of the models were created, and many diagnostic tools were use: concentration plot comparing predicted and actual values (diagnostic model and sample); concentration plot of residuals vs actual concentration and diagnostic sample) and root mean square RMSEP, or prediction error (model diagnostic), the validation's anticipated concentrations samples were determined.

4) LC-MS/MS:
The APIs or a drug's degradation products can be identified and characterized using the superior and sophisticated analytical tool known as LC-MS/MS product.These methods are combined to create becoming more prevalent in the analytical structural synthetic chemistry. Applications of analysis to HPLC and MS are also well-known. Using HPLC converting the complex of chemicals into its components, and MS as a great for chemical characterization.

5) First Derivative of Ratio Spectra Spectrophotometric Method:
The method's key benefit is that it cancels out the entire spectrum of the interfering material. Consequently, the selection of the It is not crucial what wavelength is used for calibration. The optimal outcomes are displayed in terms of signal to sensitivity, noise ratio, and selectivity.[14]

Drug shelf-life estimation:
The amount of time following manufacture during which the typical drug characteristic (such as potency) of the drug material still meets the approved specifications known as its shelf life or expiration date. United States Food and Drug Administration administration (USFDA) furnishes each container's label. The drug product must display the medicine's expiration date substance. Typically shelf-life is assessed depending on results of the drug's assay and a drug's characteristic Since a product's exact shelf life is frequently unknown, often from a stability analysis carried out during the process of developing drugs.[15]

Garret and carper method:
The shelf-life estimation process used in this in line with the Arrhenius plot. The notion that based on the mathematical outcome, shelf life produced with the use of the Arrhenius formula equation that incorporates the impact of impact of chemical reaction temperature on rate at 1/T of the thermodynamic temperature, constant k it was straight when viewed. The amount k derived from temperature readings by extrapolation based on this line's slope. This k Value is replaced with insignificance. The sequence of response demonstrates the degree of degradation occurs during the specified time. Hence, the main Operations are necessary to ascertain. This sequence of event.

Hold time stability studies result evaluation:
It is a stability-establishing tool for all phases of the production of pharmaceutical products. Hold time stability is a consideration in the development of medicinal products. An important tool for assessing the in-process hold time. Each hold period stability is being assessed. Stage of the manufacturing process. Ensure stability to determine the required period of time, study is used. An appropriate way to keep the mixture or bulk stage before it moves on to the next level. Time when appropriate Limits for when each stage of manufacturing must be completed must be defined to ensure the product quality of the medicine. Process of makingthe drug's product Compounds and products determine the setting up the hold time study. The important Study time is indeed one of the criteria included in the procedure. Points, conduct study phases, and conduct drug analysis test.[15]

Hold time studies result evaluation:
Evaluation of the shelf life of the medicine and its component at each stage of manufacturing requires the holding of study data. The durability of if the period is up to 45 days, the particular stage 60 days have passed since the hold time samples began.[15]

CONCLUSIONS:

Forced degradation is a crucial stage in the formation of new drugs. The evolution of stability indicating methods, the formation of more stable formulations, the establishment of drug product degradation pathways, the confirmation of the structure of degradation products, the revelation of drug product degradation mechanisms such as hydrolysis, thermolysis, photolysis, and oxidation, and the comprehension of chemical properties all require forced degradation studies. Stability testing of emerging drug products is regulated within ICH guideline Q1A. Highlights necessity approved stability-indicating testing methodologies to be used for analyzing features that are subject to change during storage and may have an impact on quality, safety, or efficacy.This analysis also facilitates in the specification and shelf life of a medicinal product. The study's findings will aid in the improvement of the product's formulation, manufacturing process, and storage conditions. A properly designed and managed to carry out forced degradation study would fetch a representative sample for the development of a stability indicating method. As a result, the forced degradation study must be demonstrated throughout method development and before the regulatory dossier is forwarded to the FDA.

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