Original Article

A review on forced degradation studies, stability indicating method and stability profile of few antiviral drugs

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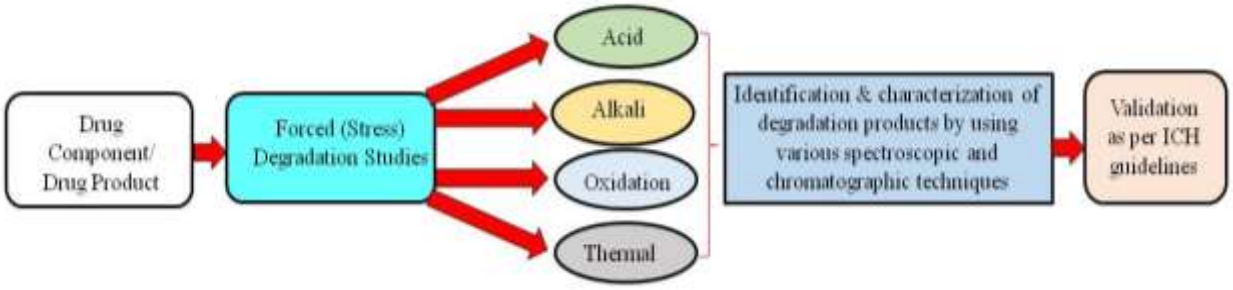
Abstract

The aim of the study is to provide with available literature on regulatory aspects and protocols for forced degradation investigations and stability indicating methods of various drug substances and their drug products too. Also, it provides information on stability profile of various antiviral drugs. Forced degradation is a procedure in which the drug material or drug product is exposed to conditions that are more severe than accelerated once, resulting in the creation of different degradants which helps in determining the stability profile of molecules. Stability indicating technique is a process in which the reduction in the amount of active pharmaceutical ingredient (API) in drug product (DP) due to degradation is determined quantitatively. The use of antiviral drugs has increased to treat various viral diseases. This has resulted in development of resistance of the drugs towards the targeted virus. These drugs have emerged as a new challenge for estimation in general and researchers. So, there is a pressing need to focus on this antiviral class of drugs and also to study about stability behaviour of these drugs so far. The current literature discusses about concept of forced degradation studies, stability indicating methods and degradation profiles of numerous antiviral medicine in bulk and varied dosage forms by using conventional as well as newly developed methods. The current article also discusses the strategies and stepwise approach for developing stability indicating methods. Forced degradation techniques are helpful in determining the degradation pathway and structure elucidation of degradants produced which in turn helps for selecting proper storage conditions as well as to modify the manufacturing process of drug moiety or drug product.

Keywords: Forced degradation; Stability indicating methods; Antiviral; Regulatory aspects.

INTRODUCTION



Stability testing is vital in the medication development process since stability is a critical quality attribute of pharmaceutical products. Stability testing is used to establish a retest period for a drug substance or a shelf-life for a drug product, as well as recommended storage conditions, by demonstrating how the quality of a drug substance or drug product changes over time under the influence of a variety of environmental factors such as temperature, humidity, and light [1].

The purity, potency and safety are affected by the drug component or drug product stability which seems to be critical. Any change in drug stability can result in the formation of a toxic substance or reduces its potency. So, it is crucial to know the effect of various environmental conditions on drug component or drug product which in turn will help us to know its purity profile [2].

OBJECTIVES OF STABILITY STUDIES

Stability studies can be used to determine the storage conditions and shelf life of a drug component or drug product. According to Committee for Proprietary Medicinal Products (CPMP) recommendations, it explains how the quality of the drug component or the drug product gets changed over time as a result of various environmental conditions. The stability of drug component actually means "controlled, documented and acceptable change" rather than "fixed" or "not likely change" [3,17].

FORCED DEGRADATION (STRESS TESTING) STUDIES

Stress testing is defined as the stability testing of drug component and drug products under conditions that are more severe than those used in accelerated testing [4].

"Stress testing of the drug component can help identify the likely degradation products, which can in turn help establish the degradation pathways and intrinsic stability of the molecule, and validate the stability indicating power of the analytical procedures used," according to ICH guidance on stability testing [1]**.**

This definition of stress testing (forced degradation) is useful as it distinguishes from two other types of stability studies: accelerated stability studies and long -term stability studies. So, it is one of the predictive tool for identify stability issue related to drug component or drug product which aids in the development of stability-indicating methods [5]**.**

Dolan J.W, 2002, has explained about stability indication, in which he has mentioned that any OTC product has a shelf life mentioned on it. That means the product should be effective and stable within the stated shelf life period under normal storage conditions. To determine this shelf life, he mentioned to measure two aspects of drug after it has been stressed. First aspect is to measure the potency or the active ingredient and the second one is to determine the degradants or impurities resulting due to ageing. The second aspect is more important as it becomes necessary to determine what type of degradants are present and find out all those possible degradants having concentration of more than 0.1% of parent drug. Further he explained about developing a stability indicating technique which involves three aspects, obtaining a representative sample, choosing the separation technique, and selecting the detector [6].

When designing stability-indicating procedures, forced degradation or stress testing is used to establish specificity, especially when little information about probable degradation products is available. These studies also reveal the mechanisms of degradation and the degradation products that may occur during storage. Forced degradation technique will help us to focus on areas such as manufacturing, packaging, and formulation development and also the information about chemical behavior can improve drug product quality [7].

When there is limited knowledge on probable degradation products, stability-indicating procedures such as forced degradation or stress testing are used to demonstrate specificity. This will help us to provide knowledge about degradation products that may occur during its storage.

During drug development process, stress testing is a critical component and by forming important stress testing samples we can predict about the degradation conditions early in the process and this will be helpful to the drug manufacturers in terms of time and money. Selection of more-stable drug substance salt forms and drug formulations also becomes easy. This method is also becoming increasingly useful in the testing of new pharmacological compounds. By applying stress testing, we can develop suitable method and the stability data is gained form these methods which will affect on actual compound of interest chosen for method development [8].

Stress conditions are applied to a drug product to approximate long-term stability in a short period. The tentative idea of long-term stability in a short period can be obtained by applying stress conditions to a drug component or drug product [3].

During preformulation, pharmaceutical companies perform these stress testing (or forced degradation studies) to select appropriate parameters such as sample production for developing stability-indicating analytical methods, compounds and excipients for further development, and formulation optimization. The information about degradation mechanisms and potential degradation products obtained from stress testing can be used to build manufacturing methods or choose appropriate packaging and prepare reference material for degradation products found. Although in early-phase drug development, it involves

preformulation studies but stress testing can be repeated during product composition, manufacturing processes, and analytical procedures [4].

In case of solid-state studies, the drug substances are exposed to light, heat and humidity and heat while in case of solution-state studies, it is exposed to extensive range of pH values. Whether the suggested approach is "Stability Indicating," or not will be determined by the experimental samples produced, i.e., the method is capable of identifying the loss in content of the active component followed by increase in degradation products [9].

An approximately 5–20% degradation of the drug component or drug products should be obtained by the stress conditions or it should indicate a feasible maximum condition achievable for the drug substance or drug products. A comparison should be made of the stress sample with the control and the suitable blank. It is not necessary that the drug component or drug products may degrade for a given stress conditions. In such case, no further stressing is suggested. From the complicated combinations of degradants generated from stress studies only significant degradants should be considered for method development [10].

To help develop and establish the specificity of such stability-indicating technologies, forced degradation studies (chemical and physical stress testing) of new chemical entities and medicinal products are required [11].

Both FDA and ICH suggests to perform forced degradation or stress testing of drug component and drug products by using conditions such as temperature, acid and base hydrolysis, photolysis and oxidation but neither FDA nor ICH have given proper guidelines on how to perform these stress studies and have completely left over the pharmaceutical companies to choose stress conditions design of these studies. In general, at least four samples should be produced in stress testing for each stress condition: (a) a zero-time sample containing the drug that is stored under normal conditions; (b) a drug solution that has been treated to stress treatment. (c) a blank solution that has been stressed in the same way as the drug solution has been; (d) a blank solution that has been stored in normal conditions [3].

The stability studies are of 2 type's viz. long term (12 months) and accelerated stability studies (6 months). Also 3rd study can be performed at conditions milder than those used in accelerated studies i.e. Intermediate studies. Usually these studies require longer time for the entire process of degradation. Comparatively forced degradative studies requires much lesser than the other stability studies. The SIM that was produced using samples from stress experiments can then be used to investigate samples from accelerated and long-term stability investigations [12].

It is critical to determine which of the potential degradation product formed under FDS conditions in either drug component or drug product and also same produced under long term or accelerated storage conditions and then selecting appropriate measures to reduce these degradants [13].

Forced deterioration experiments are carried out for the following reasons

a) To Develop and validate stability-indicating methodology drug component and drug products [14].

b) To find out the degradation pathways of drug component and drug products [12].

c) To differentiate degradation products those are related to drug products from those that are generated from excipients in a formulation [12].

d) To search for the mechanisms of degradation such as hydrolysis, oxidation, thermolysis or photolysis of the drug component and drug product [12].

e) To generate degradation profile that would be similar to the one observed in a formal stability study under ICH conditions [12].

f) To determine whether a drug component or a drug product is intrinsically stable [15].

g) To study the chemical properties of drug component or drug product [15].

h) To produce stable formulations [15].

i) To determine the structure of degraded products [15].

j) To solve stability related problems [15].

Degradation conditions

Hydrolysis

One of the most common degradation chemical reactions is hydrolysis over an extensive range of pH. It involves deterioration of a drug component or drug product by reaction with water. Ionization of functional groups present in a drug component or drug product gets catalysed when exposed to hydrolysis under acidic and alkaline conditions. When subjected to acidic or alkaline conditions, a drug component or drug product undergoes forced deterioration, which produces primary degradants. The kind and quantities of acid or alkali are chosen depending on the stability of the drug component or drug product. Most usually recommended reagents for hydrolysis are sulfuric acids or hydrochloric acid (0.1–1 M) for acid hydrolysis and potassium hydroxide or sodium hydroxide (0.1–1M) for alkali hydrolysis. Stress study should be performed at room temperature and if required degradation is not achieved then combination of stress condition (e.g. temperature) can be applied. If the compounds have poor solubility in water, then co-solvents can be used. Based on the drug component structure the co-solvent is selected. The degradation samples should be neutralised with the same concentration of alkali and acid to minimise further degradation reactions and to protect the HPLC column from damage [12, 16].

Oxidation

Hydrogen peroxide is most commonly used for oxidation of drug substances in forced degradation studies but other oxidizing agents such as metal ions, oxygen, and radical initiators e.g. azobisisobutyronitrile (AIBN) can also be used. An electron transfer occurs during the oxidative breakdown of drug components and drug products, resulting in reactive anions and cations. The type and concentration of an oxidising agent are chosen in accordance with the drug component or drug product. Most commonly hydrogen peroxide solution in the scale of 0.1–3% at neutral pH is used. Hydrogen peroxide study should not be carried out with Combination stress studies (E.g. elevated temperature). Chemically, the O-O bonds in hydrogen peroxide (H-O-O-H) are not so stable, and in fact hydrogen peroxide decay at even room temperatures. If heated the bonds of H_2O_2 decompose even faster which leads to hydrolysis rather than oxidation. Degradation samples produced during oxidation should be neutralised with a sodium metabisulfite solution of the same concentration [12, 16].

Photolytic conditions

The photostability testing of drug substances or drug products must be carried out to demonstrate that a light exposure does not produce any unacceptable change. Photolytic conditions can induce photo oxidation by free radical mechanism. The principal degradants of drug components or drug products are produced when exposed to fluorescent or ultra violet conditions are. ICH guidelines Q1B describes photostability testing of new drug components and products. It is recommended that UV-VIS light exposure of at least 1.2 million lux hours and integrated near ultraviolet energy of at least 200 watt hours/square metre be used. The same samples should preferably expose to near ultraviolet and cool white fluorescent lamp. The most frequently acknowledged wavelength of light for photolytic deterioration is between 300 and 800 nm. The maximum of 6 million lux hour's illumination is recommended. In the lack of specific instrument, natural light will be an alternative. But variations due to the time of day, weather conditions and atmospheric pollution affects the intensity of daylight as well as spectral distribution, due to which natural light is not suitable for testing [12,16].

Thermal conditions

Conditions more extreme than recommended ICH Q1A accelerated testing conditions should be applied for thermal degradation (e.g., dry & wet heat). In case of solid samples, exposure to heat in both humidified and non-humidified conditions can be done, whereas in case of liquid samples, exposure to heat without humidity is recommended. Because heat stress causes liquid samples to lose water, the concentration of the drug ingredient in the solution can fluctuate. By generating multiple time point's results, knowledge about rate of degradation, primary and secondary degradation products can be studied. The Arrhenius equation can be used to investigate the effect of temperature on drug substance or drug product thermal degradation:

$k = Ae^{-Ea/RT}$

Where k is the specific reaction rate, A is the frequency factor, Ea is the energy of activation, R is the gas constant (1.987cal/deg mole), and T is the absolute temperature. A study of thermal degradation is conducted at 40–80ºC [12,16].

Microwave-assisted chemistry has resulted a very efficient and powerful technology to heat reaction mixtures in a sealed reaction vessel. Modern microwave instrumentation has been proven to significantly cut processing times compared to traditionally heated tests under reflux circumstances [13].

Humidity

One of the most critical parameters in the degradation process is humidity. In forced degradation experiment, the drug component when exposed to 90% humidity for 1 week leads to degradation [15].

Factors affecting degradation of drug substances or drug product

Moisture

Water-soluble substances may get dissolved when there is moisture which will results into physicochemical changes within the molecule.

Excipients

Few excipients may contain high content of water/moisture which may leads to increase level of water in the formulation which later on hampers on drug component stability. Sometimes, the decreased stability is observed due to the chemical interaction between the excipients and the drug component or drug products [16].

Temperature

Temperature plays an important role on the drug component stability. Usually the rate of drug hydrolysis is found to be increased with increase in temperature.

pH

pH also plays a vital role on the drug component stability. To reduce the pH induced degradation rate of drugs by hydrolysis, buffer solutions of pH with maximum stability are used during formulation of the drug component.

Oxygen

Oxygen leads to cause oxidation in few drug substances. This increased rate of deterioration of drug component or drug product are controlled by purging nitrogen or carbon dioxide in the vessel where the drug is to be stored.

Light

Few drugs are light sensitive in nature and decompose when exposed to light. The sensitivity to decomposition by light can be found out by comparing the stability of the drug component or drug product when exposed to light as well as in dark. So, it is highly recommended that the light sensitive drug component or drug product should be kept in the amber coloured containers or should be stored in dark [15].

Prediction of degradation pathways and product

Our prediction procedure begins when a sample with a degradation problem is submitted. It involves 2 steps viz. in-silico (computer software) & in-cerebro (chemistry knowledge).

In-silico prediction

Predictive software's mostly used are as follows:

A. CAMEO (Computer-Assisted Mechanistic Evaluation of Organic reactions) [13]

B. DELPHI (Degradation Expert Leading to Pharmaceutical Insight) [13]

C. Zeneth [13]

In-cerebro prediction

In-cerebro prediction tools of major utility have been published in reviews and books in the primary literature. It explains the major chemical degradation mechanisms of medicines with different functional groups which include dehydration, hydrolysis, isomerization/epimerization, oxidation, dimerization, decarboxylation, photolysis and polymerization, and transformation products involving contact with excipients/salt forms. Drug degradation prediction will improve as the field of degradation chemistry matures with documentation of investigational experiments in the literature.

Drug degradation database

This concept was developed at Pfizer and it contains structure-elucidation data which allows researchers to recollect data easily based on change of structure and degradation chemistry conditions. This was achieved by using CambridgeSoft Corporation's ChemOffice WebServer product, a comprehensive Windows-based program, which was capable of achieving the degradation database goals. Degradation findings are gathered into one organised record per API, allowing fractional and full chemical structure searches, as well as searches depending on degradation conditions [13].

Regulatory Guidelines

The compendia of many nations or regions have developed general rules for Good Manufacturing Practices (GMPs) for the sector. Some organizations, such as the Food and Drug Administration (FDA), the International Conference on Harmonization (ICH), the World Health Organization (WHO), the European Medicines Agency (EMA), the Japanese Pharmacopoeia (JP), and the Agencia Nacional de Vigilancia Sanitaria (ANVISA), have provided stability testing guidelines for new drug component and drug product stability testing. The criteria for the United States are found in 21 CFR part 211 Section 166, which states that a written testing method designed to determine the stability characteristics of a drug component or drug product must be in place [3, 15].

Regulatory perspectives of forced degradation

Issues addressed in regulatory guidance's include

- One batch of material is normally used in forced deterioration studies.
- Forced deterioration circumstances are more severe than accelerated stability testing, such as temperatures above 50°C, relative humidity below 75%, light levels above ICH, high and low pH, oxidation, and so on.
- The design of a forced degradation research should include photostability.

- It may not be necessary to identify or analyse the structure of degradation products that do not occur in accelerated or long-term stability.

- It's important to think about mass balance.

Issues that aren't addressed in the regulatory advice

- For forced deterioration research, exact experimental parameters (temperatures, time, and extent of degradation, for example) are not given.

- The applicant has complete control over the experimental design [10, 13].

ICH GUIDELINES

The ICH guidelines which discuss about forced degradation studies are ICH Q1A, Q1B, and Q2B, Q3A, Q3B, M4Q (R1) [15] which are enlisted in Table 1.

Table. 1 ICH guidelines for forced degradation studies

Guidelines	Title
Q1A(R2)	Stability testing of new drug substances and products
Q1B	Photostability testing of new drug substances and products
Q2(R1)	Validation of analytical procedures: Text and methodology
Q3A(R2)	Impurities in new drug substances
Q3B(R2)	Impurities in new drug products
M4Q(R1)	The common technical document for the registration of pharmaceuticals for human use.

STABILITY INDICATING METHOD (SIM)

One of the most often used analytical procedures is drug degradation, which may be quantified using SIM. A stability-indicating method is a validated quantitative analytical procedure that can be used to identify how the stability of drug component and drug products changes over time, according to FDA guidance. This method accurately analyses the change in the active ingredient concentration in presence of excipients, deterioration products and impurities. The most extensively used analytical tool for separation and quantification of contaminants is the RP-HPLC paired with a UV detector [12].

The United States Food and Drug Administration (US-FDA) stability guideline of 1987 and the draught guideline of 1998, on the other hand, contain detailed descriptions of stability-indicating technique. Stability-indicating methods according to USFDA 1987 guideline were defined as the 'quantitative analytical methods that are based on the characteristic structural, chemical or biological properties of each active ingredient of a drug product and that will distinguish each active ingredient from its degradation products so that the active ingredient content can be accurately measured.'[18]

This definition in the draft guideline of USFDA 1998 reads as: 'a validated quantitative analytical methods that can detect the changes with time in the chemical, physical, or microbiological properties of the drug substance and drug product, and that are specific so that the contents of active ingredient, degradation products, and other components of interest can be accurately measured without interference.' [18] The primary modifications in the new guideline are (i) the addition of a validation requirement, and (ii) the requirement of examination of degradation products and other components, in addition to the active ingredients (s) [18].

Techniques used in analysis of stability samples

Titrimetric and spectrophotometric

In this technique, only the interested drug is analysed in presence of additives, excipients, impurities, degradation products, etc., and also other drug components in case of the combination products. Simplicity and low cost is the main advantage but sometimes they are found to be less sensitive. These days there are hardly any reports on their use due to lack of its specificity.

Chromatographic

Its advantages include multicomponent separation, greater accuracy and sensitivity. Various chromatographic methods that have been used are, gas chromatography (GC), capillary electrophoresis (CE), thin-layer chromatography (TLC), HPLC, high-performance thin-layer chromatography (HPTLC).

Miscellaneous

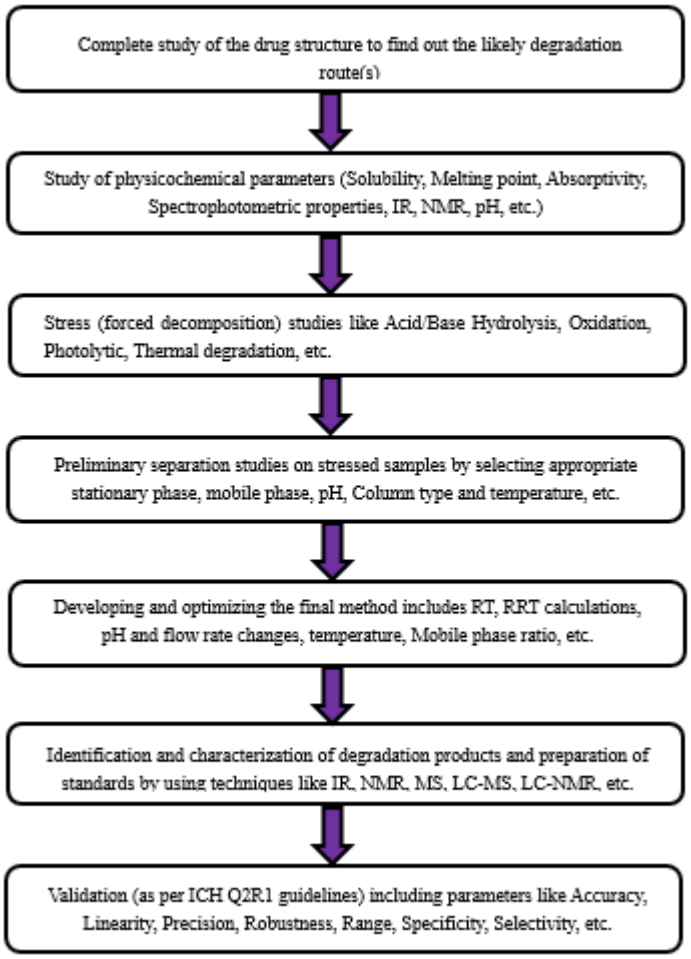
Proton nuclear magnetic resonance (H-NMR) spectroscopy is used for the establishment of SIAMs has been found in earlier reports while now CE is also one of the latest techniques for the development of SIAMs. It has the advantage of high resolution, sensitivity, and high efficiencies with minimal peak dispersion. The various hyphenated techniques like GC-MS, LC-MS, LCMS-MS, LC-NMR and CE-MS techniques for identity confirmation of known and unknown degradation products and their selective determination [18].

Establishment of stability indicating method

Although regulatory regulations specify the requirements for stability-indicating procedures, neither regulatory guidelines nor pharmacopoeias provide information on the basic stages to be followed in the development and validation of stability-indicating methods [13].

Steps involved for development of SIM are shown as in Fig. 1

Fig. 1. Steps involved for development of SIM.



Few critical issues to be considered when developing and validating of SIAMs

Specific and Selective stability-indicating assay methods

A specific stability-indicating assay method (Specific SIAM) is defined as “a method that can measure the drug(s) definitively in the presence of any degradation products, excipients, and additives that are predicted to be present in the formulation.” On the other hand, the Selective stability-indicating assay method (Selective SIAM) is defined as "a method capable of measuring indisputably the drug(s) and all degradation products in the presence of excipients and additives predicted to be present in the formulation." [13]

When to use ‘Specific or Selective SIAM’

In case of older and established drugs, significant information is available and hence specific SIAM can be very well used. Due to this reason, the pharmacopoeias, other than USP, have a policy to continue with the titrimetric and spectrophotometric analysis for the assay of drugs, while having control on important degradation products through related substance or impurity tests. In case of new drugs since much information is not available, so selective SIAM will be more applicable.

Should the development of a SIAM be limited to key degradation products?

Sometimes even in single stress situation large no of degradation products are formed and for all stress conditions, very large no of degradant products will be formed and developing a selective SIAM for such situation becomes difficult. So, the question arises, Should the development of a SIAM be limited to key degradation products? The ICH guidelines provide a comprehensive answer to the question as 'However, If it has been proven that particular degradation products do not occur under expedited or long-term storage conditions, it may not be required to look for them specifically.'

Can stress studies be conducted on formulations instead of drug substance while developing SIAM?

There is no proper guidance on conduct of stress studies directly on formulation other than photostability testing (ICH Q1B) and particular testing on certain products, (e.g. emulsions, metered dose inhalers, refrigerated aqueous liquid products, creams).

The trauma of establishment of SIAMs for combination drug formulations

Formulation which contains more than two drugs, separation of individual drug from the combination, followed by separation from degradation products (which might be of any number) becomes very difficult and hectic. Thus, only degradation products formed during long term, accelerated stability testing conditions should be focused on.

The importance of mass balance in development of SIAMs

It is the sum of the assay value and levels of degradation products to see how closely these add up to 100% of the initial value, along with consideration of the margin of analytical error. In case of degradation products that are stable, can be separated easily and standards for which are available, then inception of mass balance becomes easy. But there are situations where establishing mass balance difficult due to one of the following reasons:

- Formation of more than one degradation products, involving complex reaction pathways and drug excipient interaction products,
- Incomplete visualisation due to loss of UV chromophore or lack of universal detection,
- Loss of drug/degradation products due to its volatile nature,
- Diffusive losses into or through containers
- Elution/resolution problems
- Inappropriate or unknown response factors due to lack of standards
- Variations in drug content and assay errors

If mass balance establishment during SIAM development is possible then it's good enough. But in some cases, where mass balance is not observed then here a method should be valid for other characteristics like precision, specificity, ruggedness, robustness, etc. and the difficulties experienced in inception of mass balance must be stated and indicated in the pertinent part in the registration application.

Are pharmacopoeial methods stability-indicating?

In the compendial monograph, the assay method prescribed were stability specific and not selective in nature. However, advances in technology and with the introduction of ICH guideline Q1A (R2), the degradation products can be simultaneously analysed. The USP also contains large number of assays and several of which are selective by nature. USP also defines Category II analytical procedures that are meant for determination of impurities in bulk drug substances or degradation compounds in finished pharmaceutical products and provides data elements required for validation of these. In case of BP they are attempting for shifting to 'Selective' methods.

New approaches for analysing stability samples are being developed

Conventional techniques used for testing of stability samples are GC, HPLC, CE, or NMR, etc. Recent techniques involve use of hyphenated techniques like CE-MS, LC-MS or LC-MS-MS, GC-MS, LCNMR, etc. Fourier transform near-infrared (FT-NIR) spectroscopy is another emerging technique than hyphenated technique. It allows the analysis of drugs directly in the dosage forms which is its unique advantage. Technically the assay using FT-NIR are specific, but can be made selective if only few degradation products are formed on storage.

SIAMs developed and optimised using computer simulation

The manual method for development of SIAM by HPLC is tedious and challenging process. Also, there are huge numbers of interconnected parameters involved during method development. Due to these limitations of manual HPLC the adoption of computer-based solutions to automate various parts of the HPLC process is on the rise. The advantages of computer simulation over manual method development are:

a. Obviates the majority of the necessary experimental work in the development and optimization of chromatographic methods.
b. It can continue in an unattended manner once the process has begun.

Few examples of software's used in HPLC method development are:

DryLab, ENHANCER, ICOS, DIAMOND, PESOS, PRISMA model, ELUEX, CHROMSWORD, HPLC-METABOLEXPERT, ProDigest-LC, CHROMDREAM [18].

Need for antiviral drugs study

The newly found viral infections are found to be resistant to the available antiviral drugs and hence the available antiviral regimen turns to be unsatisfactory which in turn become an alarming threat and a serious health concern. Because of increasing occurrence of viral infections and especially of resistant viral strains, there is a need for improving the available antiviral regimens and also there is a need for challenge to discover novel antiviral agents to counter the new resistant viral strains [19-22].

From earlier reports, the force degradation studies of different drug molecule, the stability profile and the degradants produced for the drug molecule are summarized in the Table 2. [51-54].Also the same is shown in Fig. 2

Table. 2 Stability profile of few antiviral drugs

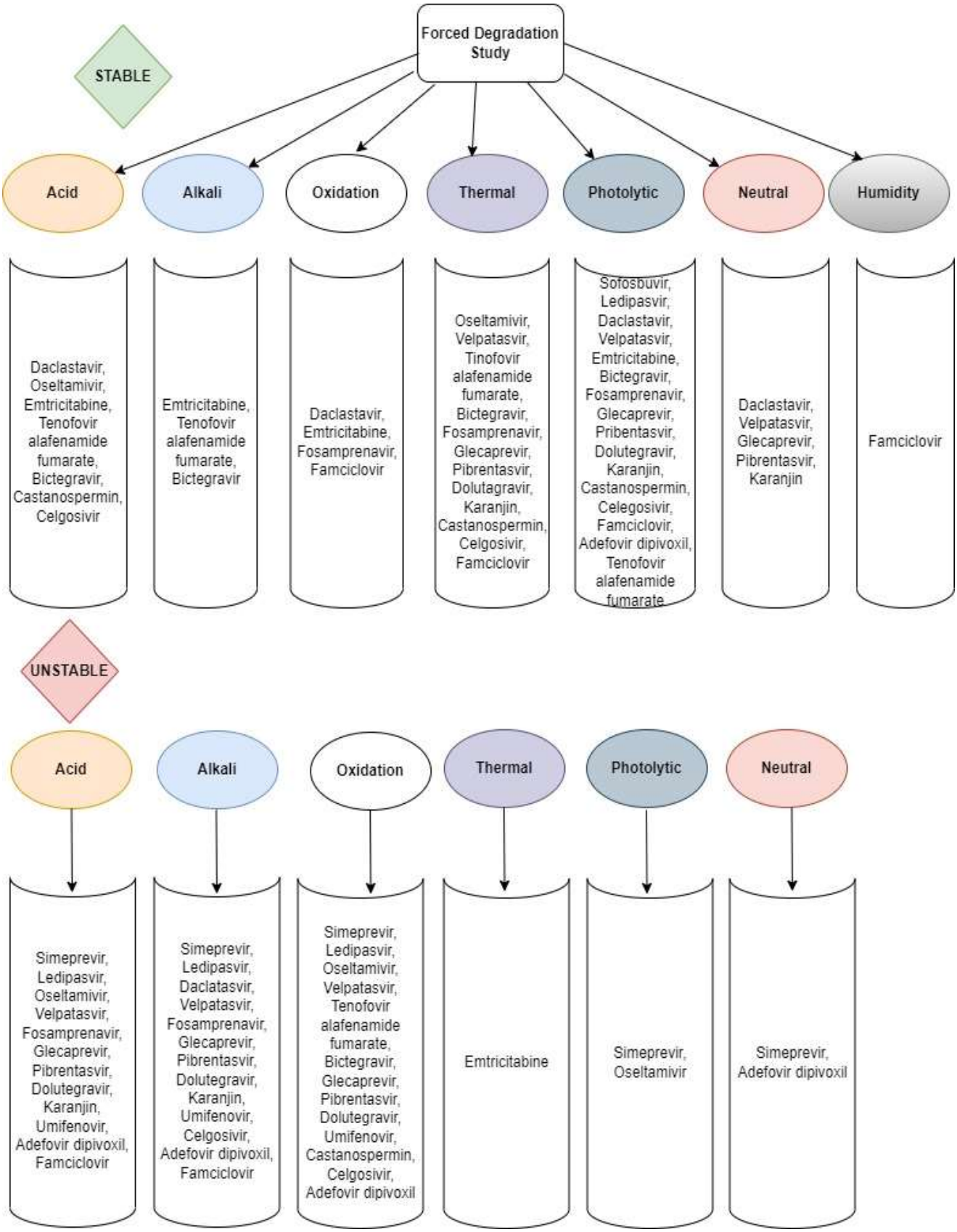
Sr. No.	Year	Name of the drug	Stable under	Unstable under
	2021	Bictegravir, Emtricitabine and Tenofovir Alafenamide Fumarate	Emtricitabine-Acidic, alkali, oxidative & Photolytic situations. Tenofovir AF and Bictegravir- Acidic, alkali, thermal & Photolytic situations.	Emtricitabine-Thermal situations. Tenofovir AF and Bictegravir-Oxidative situations

	2020	Simeprevir (in pharmaceutical dosage form and in spiked human plasma)	-	acidic hydrolytic, alkaline hydrolytic, oxidative routes, photochemical degradation and neutral hydrolysis
	2020	sofosbuvir (SF) and ledipasvir (LD)	Photolytic situations	acidic hydrolytic, alkaline hydrolytic, oxidative routes
	2020	Daclatasvir (DCV)	acidic, neutral, oxidative and photolytic situations	basic situations
	2020	Oseltamivir	acid hydrolysis and thermal degradation	photolysis, oxidative and alkali degradation
	2020	Velpatasvir	Neutral, photolytic and thermal stress situation	acid, base and oxidative situations
	2019	Tenofovir Alafenamide fumarate	Thermal degradation and photolytic degradation situations	Oxidative conditions, Acid and Alkali hydrolysis
	2019	Fosamprenavir	Oxidative, photolytic & thermal degradation studies are not reported.	Acidic and basic stress situations
	2019	Glecaprevir (GLE) and Pibrentasvir (PIB)	Neutral, UV and thermal situations	Alkaline, acidic and oxidative stress situation
	2019	Dolutegravir	Thermal degradation and photolytic degradation situations	Acid hydrolysis, Alkali hydrolysis, & Oxidative situations
	2019	Dolutegravir	basic, thermal and photolytic stress situations	acidic and oxidative situations
	2019	Daclatasvir dihydrochloride (DAC) in combination with Sofosbuvir	Acidic situation, photolytic situations -day light	Alkali, oxidative and photolytic situations -UV radiation & direct sunlight
8	2019	sofosbuvir and ledipasvir	sofosbuvir - photolytic situations and ledipasvir-not stable under any situations	sofosbuvir - acid hydrolysis, base hydrolysis, oxidation degradation and thermal degradation and ledipasvir- acid

				hydrolysis, base hydrolysis, oxidation degradation, thermal degradation, and photolytic degradation
8	2019	Daclatasvir	Heat acidic, oxidative	basic and light situations
8	2019	daclatasvir dihydrochloride	photolytic and thermal stress situations	acid, base, neutral and oxidative situations
	2019	Daclatasvir (in pharmaceutical dosage form)	photolytic and thermal situations	acid, base and peroxide stress situations.
	2018	Karanjin	Neutral, photolytic and thermal situations	Acidic and Alkaline situations
8	2018	Sofosbuvir (Pharmaceutical Dosage form)	Not stable in any situations	acidic, alkaline, oxidative, thermal, photolytic situations.
9	2018	Sofosbuvir and Velpatasvir (Combination)	Sofosbuvir & Velpatasvir drugs are stable at all situations like alkaline, thermal, oxidative, humidity & UV stress situations	Only Velpatasvir is degraded in acidic situations
8	2018	Umifenovir	Not stable in any situations	Acidic, Alkali, oxidation, and hydrolysis situations
3	2018	Ledipasvir	Oxidative & UV degradation	Acid, alkali, photolytic (direct sunlight) situations
9	2018	Velpatasvir (VLP)	daylight situations	acidic, alkaline, oxidative, sunlight and UV situations
6	2018	Tenofovir alafenamide	-	acidic, basic, oxidative (3% H2O2) and neutral hydrolysis
8	2018	Sofosbuvir (SOF)	heat, photo and thermal situations	acid, base, water and oxidation situations
8	2018	castanospermin and celgosivir	Acidic, UV-Visible and thermal situations	Castanospermin-oxidative and celgosivir –oxidative and base hydrolysis
0	2018	Adefovir dipivoxil (ADL)	photolytic and thermal stress situation	acid, alkaline, neutral and oxidative stress situations

	2018	tenofovir disoproxil (in tablet dosage form)	photolytic, humid and acidic situations	basic, neutral, thermolytic and oxidative situations
8	2017	daclatasvir dihydrochloride (in drug component and drug prodruct)	Acidic, neutral, and photolytic degradation.	Alkali and oxidative situations
7	2016	Sofosbuvir	photolytic and thermal situations	acidic, basic and oxidative situations
1	2013	Tenofovir disoproxil fumarate (TDF), Emtricitabine (FTC), and Efavirenz (EFV)	EFV is stable to acidic & neutral situations, FTC and EFV are stable to alkali, all 3 drugs are stable under thermal & photo degradation.	FTC and TDF susceptible to acidic & neutral situations, TDF is more prone to alkali
9	2009	Oseltamivir	Neutral and Thermal situations	base, acid and hydrogen peroxide
9	2008	Famciclovir (in bulk drug and in the form of pharmaceutical dosage)	thermal, humidity, peroxide and photolytic degradation	acid hydrolysis and base hydrolysis

Fig. 2. Steps involved for development of SIM



CONCLUSION

This review is an attempt to focus on the concept, regulatory aspects and techniques used for the forced degradation studies and stability indicating methods. Stress testing studies are helpful in providing the information about prospective degradation pathways of new drug components and drug products and to demonstrate the analytical methods stability indicating capacity. These proposed techniques are helpful for selection of suitable storage conditions meeting drugs protocol and improving its manufacturing processes. The review has also made an attempt to explain the importance of antiviral class of drugs and summary of the stability profile of few antiviral drugs under variety of stress conditions has been reported. It is envisaged that the above discussion would be of significant interest to the general public.

DECLARATION OF INTEREST

The authors report no conflicts of interest. The authors alone are responsible for the content and writing of this article.

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