WORLD JOURNAL OF PHARMACY AND PHARMACEUTICAL RESEARCH

Review Article

Coden USA: WJPPCL

2026
Volume: 03
Issue: 05
Page: 63-101

IMPURITY PROFILING IN PHARMACEUTICALS: STRATEGIES AND TOOLS FOR THE DETECTION, QUANTIFICATION, AND CONTROL OF IMPURITIES IN DRUG SUBSTANCES AND PRODUCTS

B. S. Preethi[1], G. Kranthi Kumar[2]*

[1]M. Pharmacy Student Department of Pharmaceutical Analysis, Raghavendra Institute of Pharmaceutical Education and Research (RIPER) K.R. Palli Cross, Chiyyedu, Anantapuramu -515721, Andhra Pradesh, India.

[2*]Assistant Professor, Department of Pharmaceutical Analysis, Raghavendra Institute of Pharmaceutical Education and Research (RIPER) K.R. Palli Cross, Chiyyedu, Anantapuramu -515721, Andhra Pradesh, India.

Received: 18 February 2026 **Revised: 10 March 2026** **Accepted: 31 March 2026**

Corresponding Author: G. Kranthi Kumar
Address: M. Pharmacy Student Department of Pharmaceutical Analysis, Raghavendra Institute of Pharmaceutical Education and Research (RIPER) K.R. Palli Cross, Chiyyedu, Anantapuramu -515721, Andhra Pradesh, India. **DOI:** https://doi.org/10.5281/zenodo.19940738,

ABSTRACT

Impurity profiling is a critical aspect of pharmaceutical quality control, encompassing the identification, characterization, quantification, and regulation of impurities in both drug substances and drug products. Impurities may originate from synthetic processes, degradation pathways, residual solvents, or interactions with packaging materials, and their presence can significantly impact drug safety, efficacy, and regulatory compliance. A wide range of advanced analytical techniques, including chromatographic methods (HPLC, GC, UPLC), spectroscopic tools (NMR, IR), and mass spectrometric approaches (LC-MS/MS, GC-MS), are employed for impurity profiling. International regulatory frameworks, particularly ICH guidelines Q3A–Q3D and M7, provide clear specifications for impurity limits and control strategies. This report outlines the various sources and classifications of impurities—organic, inorganic, residual solvents, and genotoxic—and emphasizes the role of Quality by Design (QbD), method validation, and risk assessment in establishing robust impurity control.

KEYWORDS: Impurity profiling, drug substance, drug product, analytical techniques, HPLC, GC, mass spectrometry, regulatory guidelines, LC-MS/MS, forced degradation, QbD, pharmaceutical quality, impurity control.

1. INTRODUCTION

1.1. Impurity Profiling:

Impurity profiling is an essential part of pharmaceutical development and quality assurance. It is defined as the detection, characterization, and quantitation of impurities in active pharmaceutical ingredients (APIs) or finished drug product.[1] Impurities can have their origin from raw materials, intermediates, solvents, reagents, or degradation products. The existence of impurities, even in minute quantities, can affect the safety, efficacy, and stability of a drug product. Hence, the understanding and regulation of impurities are critical to product quality and patient safety.[2] Impurity profiling aids in formulating decisions, process optimization in manufacture, and shelf-life estimation. It also assists in the establishment of specifications for drug substances and products.[3] Analytical methods such as HPLC, GC, MS, NMR, and IR spectroscopy are routinely used to identify and characterize impurities with high sensitivity and accuracy.[4] By means of strong impurity profiling, the pharmaceutical industry can prove regulatory compliance, improve process knowledge, and reduce the risk of product recalls or drug toxicity.[5]

1.2. Importance of Impurity Control:

Impurity control throughout drug development is required to ensure drug quality, patient safety, and regulatory compliance.[6] Some impurities may have severe toxicological effects, including genotoxicity, mutagenicity, or carcinogenicity, even at low levels. Thus, an obvious plan for impurity control ensures harmful impurities are minimized or eliminated, enhancing the final product's safety profile.[7] This is especially important during scale-up and production stages where raw material changes, reaction conditions, or purification methods can result in the introduction of new or higher amounts of impurities.[8] Lack of control of impurities may lead to rejection of batches, recall of products, or even regulatory penalties. Furthermore, unidentified or unqualified impurities compromise product consistency and can influence the therapeutic effectiveness of the drug.[9] Besides being necessitated by law, impurity control is a key aspect of quality-by-design (QBD) and good manufacturing practices (GMP) To ensure batch-to-batch consistency and product stability over a long period, drug manufacturers can specify tolerable levels of impurities and measure them with

validated analytical methods.[10] At the end of the day, strict control of impurities protects the integrity of the drug development process and builds confidence among regulators, healthcare professionals, and patients.[11]

1.3. Impact on Drug Safety, Efficacy, and Regulatory Compliance:

Drug impurities in pharmaceutical products can have a considerable impact on the efficacy, safety, and regulatory status of a drug. Even at very low levels, certain impurities—particularly genotoxic and carcinogenic impurities—present severe health threats from a safety perspective.[12] The International Council for Harmonisation (ICH) has put forth a number of guidelines (e.g., Q3A–Q3D, M7) that govern the way impurities should be handled during the drug life cycle. From an efficacy perspective impurities are able to change the pharmacokinetic and pharmacodynamic attributes of the drug substance, impacting the way it is absorbed, distributed, metabolized, and excreted.[13] Non-reproducible impurity profiles also impact the reproducibility and therapeutic response of the drug.[14]

1.4. Objectives and Scope

Impurity profiling includes identification, characterization, quantitation, and control of impurities in drug substances, excipients, and finished pharmaceutical products. It is widely and importantly applicable in all aspects of drug development and manufacture.[15] Impurity profiling has a critical role in ensuring pharmaceutical product safety, efficacy, and quality. It is an important factor in regulatory filings, process validation, stability, and product shelf-life determination.[16]

1. **Organic impurities**: starting materials, intermediates, by-products, and degradation products.
2. **Inorganic impurities**: catalysts, reagents, and heavy metals.
3. **Residual solvents and genotoxic impurities**: Each class demands unique analytical methods for detection and quantification.[17]

In addition, impurity profiling facilitates the optimization of synthesis and purification processes, ensuring batch uniformity, and reducing the potential toxicity. Impurity profiling also facilitates formulation development and packaging decisions through the assessment of degradation pathways.[18] The role of impurity profiling is, therefore, not restricted to quality control but encompasses regulatory compliance, patient safety, and lifecycle management in the pharmaceutical industry worldwide.[6]

2. Classification of impurities:

2.1. Organic Impurities:

Organic impurities are the most frequent type to be encountered at pharmaceutical development and production. They can occur at different stages of synthesis and consist of unreacted starting materials, intermediates, by-products, and degradation products [(19)]. The impurities are often structurally similar to the active pharmaceutical ingredient (API) and can be expected according to the chemical route applied to synthesis.[20]

Regulatory standards like ICH Q3A and Q3B mandate the identification and quantification of organic impurities beyond certain levels. Sensitive analytical tools like high-performance liquid chromatography (HPLC), ultra-performance liquid chromatography (UPLC), and mass spectrometry (MS) are employed for sensitive and accurate detection of such impurities.[21] Identification and control of organic impurities should be done appropriately to guarantee the safety and consistency of pharmaceutical products[1] Inorganic impurities derive from the synthesis raw materials, e.g., reagents, catalysts, ligands, or even equipment used for manufacturing.[22] These impurities are generally independent of the active pharmaceutical ingredient (API) structure and range from residual metals, inorganic salts, filter aids, and leached products of equipment used in processing.[5] Ionic residues can also result from reagents, such as acids, bases, or neutralization agents, if they are not removed effectively during purification. Whereas inorganic impurities cannot be distinguished from organic impurities by common chromatographic methods, they are not necessarily detectable.[23]

Specialized techniques such as Inductively Coupled Plasma Mass Spectrometry (ICP-MS), Atomic Absorption Spectroscopy (AAS), and X-ray fluorescence are used to determine elemental impurities.[24] The ICH Q3D guideline makes specific recommendations regarding limits of allowable elemental impurities based on toxicological studies.[25] Monitoring and control of inorganic impurities are essential not only for the safety of the product but also for ensuring that GMP compliance is sustained. Proper handling of their management ensures the chemical purity of the drug and minimizes variability and risk in drug substance and product.[26]

2.2. Residual Solvents:

Residual solvents are volatile organic compounds employed at the synthesis, purification, or formulation of drug products and excipients.[27] Though these solvents are very important in manufacturing operations—e.g., to favour chemical reaction, recrystallization, or extraction—they are not designed to be delivered with the final product.[28] When not fully eliminated, they may persist as trace-level impurities and cause potential health hazards.[29]

Residual solvents are divided by the International Council for Harmonisation (ICH) Q3C guideline into three classes:

Class 1: Solvents to be eliminated due to toxicity. e.g., benzene

Class 2: Solvents to be avoided due to inherent toxicity. e.g., methanol

Class 3: Solvents of low toxic potential. e.g., ethanol.[13]

Their permissible daily intake levels and concentration limits are well established in regulatory guidelines.[30] Analytical methods like Headspace GC and Gas Chromatography (GC) are the most used for detecting and measuring residual solvents because of their specificity and sensitivity towards volatile compounds.[31] Selection of solvents during process development, as well as proper purification techniques, is essential to keep residual solvents under acceptable limits.[32] Control of the residual solvents is crucial to ensure patient safety, product quality, and international regulatory compliance.[33]

2.3. Genotoxic Impurities

Genotoxic impurities (GTIs) are a unique class of impurities that have a direct interaction with DNA and can cause mutations, cancer, or heritable genetic damage.[34] These impurities, even in very trace amounts, are viewed as highly dangerous and are thus regulated very strictly.[35]

The most frequent sources of GTIs are reagents, intermediates, and by-products of the synthetic route of APIs.[36] Nitrosamines, alkylating agents, and epoxides are some examples. Guideline recommendations, especially ICH M7, present the estimation, control, and qualification of genotoxic impurities.[37] ICH M7 recommends a risk-based strategy encompassing structure–activity relationship (SAR) evaluations, experimental information, and toxicological evaluation.[38] It also sets acceptable intakes (AI) for genotoxic compounds, commonly on the basis of the Threshold of Toxicological Concern (TTC), usually 1.5 µg/day for lifetime exposure.[39] Analytical techniques like LC-MS/MS and GC-MS are most often utilized due to their exquisite sensitivity and selectivity, since GTIs are required to be at trace levels (ppm or even ppb).[21] Resolution of genotoxic impurities serves to maintain the health of patients, especially in chronic therapy where prolonged exposure could elevate the risk of cancer.[40] Their detection and control demonstrate the pharmaceutical industry's concern for achieving international safety standards and regulatory acceptance.[41]

3. International Regulatory Expectations

In order to restrict the number of contaminants, international regulatory agencies have established stringent requirements to ensure the quality, safety, and effectiveness of pharmaceutical products.[42] The expectations are grounded on risk assessment, scientific data, and toxicological information. Impurities—be it in the synthesis step, degradation, or packaging materials—need to be evaluated, tracked, and kept within acceptable limits throughout the product life cycle.[16] Regulatory requirements stress complete documentation of impurity profiles in drug development and submission for approval to market.[43] This encompasses method validation, impurity characterization, toxicological qualification, and justification beyond the threshold level.[44] Pharmaceutical companies are mandated by regulatory agencies such as the International Council for Harmonisation (ICH), United States Food and Drug Administration (USFDA), European Medicines Agency (EMA), and World Health Organization (WHO) to adopt harmonized standards in the control of impurities.[23] The industry is supposed to detect, quantify, and control impurities in both drug products and active pharmaceutical ingredients (APIs) using validated analytical techniques.[2] These worldwide standards allow uniformity in drug quality across markets and facilitate regulatory filing in several jurisdictions.[45] Non-compliance can cause warning letters, delayed approval, or product recall. Thus, know-how and compliance with worldwide regulatory requirements are the basis for pharmaceutical development and public health protection.[46]

3.1. ICH Guidelines: Q3A, Q3B, Q3C, Q3D and M7.

The International Council for Harmonisation (ICH) has made it important to put forward significant guidelines for the control of impurities in drug products and substances, which are practised extensively by regulatory authorities across the globe.[47]

ICH Q3A: ICH Q3A (R2) covers impurities in new drug substances, ranging from classification to identification thresholds and control strategies. It mandates identification and qualification of impurities above 0.1% or certain toxicological thresholds.[48]

ICH Q3B: ICH Q3B (R2) addresses impurities in new drug products, including degradation products which can occur on formulation and storage.[49]

ICH Q3C: ICH Q3C (R8) gives limits for residual solvents in drugs. It categorizes solvents into three groups on the basis of their toxicity and determines acceptable daily exposure limits.

ICH Q3D: ICH Q3D (R2) imposes limits on elemental impurities like heavy metals. It applies a risk-based model to establish acceptable levels based on toxicity and route of administration.[13].

ICH M7: ICH M7 offers a framework for the evaluation and control of DNA-reactive (mutagenic) impurities with potential carcinogenic activity. It includes toxicological evaluation, control limits based on lifetime exposure, and analytical approaches.[21]

3.2. USFDA, EMA, WHO Guidelines:

Aside from ICH guidelines, national and global regulatory agencies like the USFDA, EMA, and WHO have laid down their own regulatory guidelines regarding the detection and control of pharmaceutical impurities.[5] These are usually supplementary to ICH standards and include region-specific requirements and expectations.[50]

USFDA: The United States Food and Drug Administration (USFDA) issues comprehensive guidance documents, such as the "Guidance for Industry" series, that describe procedures for evaluating impurities in drug substances and drug products.[51] The FDA also places strong emphasis on risk-based methods and has issued particular guidelines on elemental impurities, residual solvents, and genotoxic impurities.[52]

EMA: The European Medicines Agency (EMA) closely follows ICH guidelines but adds clarification in its Quality Guidelines and Questions & Answers publications. The EMA especially focuses on environmental and manufacturing controls to avoid contamination (53).

WHO: WHO releases guidelines for application mostly in low- and middle-income countries and within prequalification programs. Guidelines are critical for maintaining uniform drug quality in global health programs and provide a pragmatic method for assessing impurities.[11]

3.3. Thresholds for Reporting, Identification, and Qualification

Regulatory guidelines set particular thresholds for reporting impurities, identification, and qualification in order to guarantee that potentially toxic impurities are properly evaluated and controlled.[54] Those thresholds are mostly defined as a function of the maximum daily dose of the drug and aim to balance patient safety with reasonable limits of analytical capability.[55]

Reporting thresholds: Reporting threshold is the amount at and above which an impurity needs to be reported in the product's impurity profile.[56]

Identification threshold: The identification threshold is the level at which or above which the chemical structure of the impurity must be determined.[57]

Qualification threshold: The qualification threshold is the level at or above which toxicological data are required to be presented to show that the impurity is safe at that level. For instance, for drug substances with a daily maximum dose of ≤2 g, ICH Q3A and Q3B guidelines commonly place the reporting threshold at 0.05%, the identification threshold at 0.10%, and the qualification threshold at 0.15%. The values depend on the dose and on the type of impurity.[58]

3.4. Mutagenic and Genotoxic Impurity Regulations:

Mutagenic and genotoxic impurities (MGIs) are a topic of significant concern in pharmaceutical impurity profiling due to their ability to harm DNA and induce mutations or cancer even at very low levels. MGIs may arise from reagents, intermediates, or degradation routes.[59] To manage such risks, ICH M7 offers a science- and risk-based method for the identification, evaluation, and control of MGIs in pharma products. ICH M7 classifies mutagenic impurities according to their mode of action and safety data available. In case an impurity is found to be mutagenic and has insufficient safety information, a threshold of toxicological concern (TTC) is used.[60] The default TTC for lifetime exposure is 1.5 µg/day,

which translates to a hypothetical excess lifetime cancer risk of 1 in 1,00,000. Increased values might be appropriate on the basis of compound-specific information. Control measures for MGIs involve process optimization to reduce formation, purification methodologies, and analytical methods of high sensitivity such as LC-MS/MS for trace-level detection.[61] In silico methodologies (e.g., QSAR) are frequently employed for initial mutagenicity evaluation.[62] Regulation of MGIs is increasingly a normal part of drug development and regulatory assessment, with failure to comply potentially causing delays in approval or market removal, as in the nitrosamine-related recalls over the past few years.[63]

4. Sources of impurities

4.1. Synthesis and Manufacturing process

Impurities that are introduced during synthesis and manufacturing are a primary source of contamination in pharmaceutical products and substances.[64] These impurities can result from unreacted starting materials, intermediates, by-products, and degradation products that are generated through chemical reactions.[65] Side reactions and rearrangements as well as incomplete reactions can produce unwanted and unexpected substances. Reagents, catalysts, and solvents could also introduce chemical residues if purification steps are poor.[66] Process-related impurities are frequently route-specific and depend on the synthetic pathway used, so selection and optimization of the route will be important for impurity control.[67] As an example, if halogenation is part of the synthesis, residual halides are left behind unless they are removed.[68] Likewise, heavy metal catalysts palladium or platinum, used in hydrogenation or coupling reactions, can remain as trace-level impurities. Manufacturing conditions also play a role in impurity generation through cross-contamination, equipment malfunction, or operator error.[69] Good manufacturing facility cleanliness, cleaning procedure validation, and Good Manufacturing Practices (GMP) are necessary to prevent these risks.[70]

4.2. Storage and Degradation process

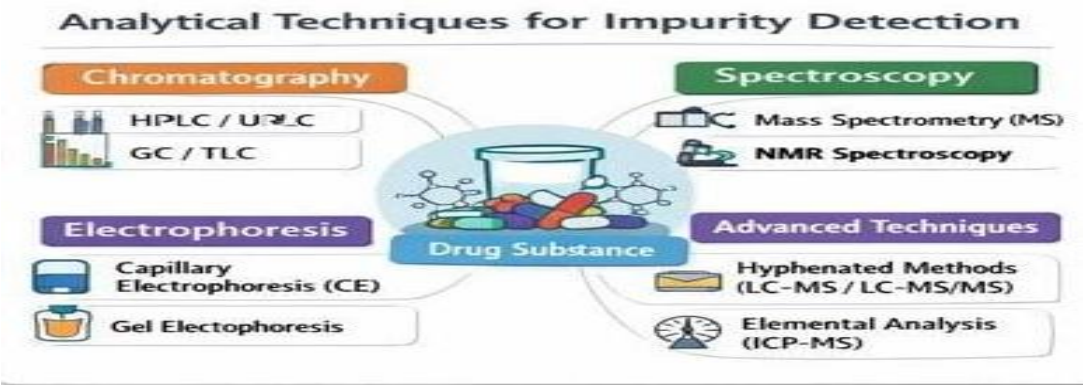
Storage conditions have a great impact on the stability of drug products and can result in the formation of degradation impurities during their long-term storage.[71] Exposure to light, heat, moisture, and oxygen may trigger chemical transformations, which degrade the API or excipients.[72] The resulting degradation products not only decrease the drug's potency but also lead to toxic effects.[73] For instance, hydrolysis is prevalent in ester, amide, or lactam-containing compounds, particularly in moist settings.[74] Oxidation is another degradation

process that frequently occurs in drugs with phenolic or amine functional groups, promoted by light and air exposure.[75] Photolytic degradation is a possible occurrence when drugs are subjected to UV or visible light, resulting in bond cleavage and intramolecular rearrangements.[76] Adequate packaging materials and storage conditions (such as temperature and humidity control) are critical to reducing such risks.[77] Stability studies, according to ICH guidelines (e.g., Q1A), assist in identifying degradation pathways and shelf-life and storage guidance determination.[78] Degradation product monitoring is an important part of impurity profiling. Forced degradation experiments mimic vigorous storage conditions to anticipate impurity development.[79] The resultant degradation products should be characterized, quantified, and regulated in accordance with allowable limits within the regulatory agency to guarantee product quality and patient safety over the shelf life of the drug.[80]

4.3. Packaging and Environmental Contamination process

Packaging materials and environmental conditions can be unforeseen yet considerable sources of contaminants in pharmaceuticals (81). The interaction between the drug product and container-closure system can cause chemical substances to migrate into the formulation.[82] Examples include plasticizers, adhesives, ink ingredients, or other extractables and leachables (E&L) from packaging materials like plastic, rubber, glass, or metal.[83] For instance, phthalates in plastic packaging or stoppers, and alkali metals in glass packaging, are able to migrate into liquid products, particularly under heat or stress.[84] Inadequately chosen or untested packaging materials therefore lower the purity and stability of the product. Environmental pollution may happen during storage, packaging, or manufacture.[85] Sources are airborne particulate matter, microbial contaminants, cleaning chemicals, or human contact-related impurities (e.g., hair, skin cells, fibers).[86] Facilities that do not follow cleanroom practices are particularly susceptible to these problems. In parenteral preparations or in sterile preparations, their presence is unacceptable and can be of serious health consequence.[87]

5. Analytical methods for impurity profiling:



5.1. Chromatographic Methods (HPLC, UPLC, GC):

Chromatographic methods are the pillar of impurity profiling because of their great resolution, sensitivity, and flexibility.[88]

Technique	Examples of Use	Common Problems	Troubleshooting
HPLC (High-Performance Liquid Chromatography)	Separation of degradation products, process-related impurities, quantification of organic impurities	Poor peak resolution, baseline noise and drift, tailing peaks, pump leakage	Use fresh mobile phase, degas solvents, replace column, optimize gradient/flow rate, check pump seals
	Quantification of degradation products in paracetamol	Broad/ overlapping peaks	Optimize mobile phase composition and gradient; change stationary phase (C18-C8);adjust PH

HPLC: High-Performance Liquid Chromatography (HPLC) is the most popular method for separation and quantitation of organic impurities, such as degradation products and process-related impurities. It provides good reproducibility and can be interfaced with UV, fluorescence, or mass detectors.[89]

UPLC: Ultra-Performance Liquid Chromatography (UPLC) is a recent evolution of HPLC based on the utilization of smaller particle sizes (<2 µm) for improved speed and resolution of separations, especially beneficial for intricate mixtures of impurities.[90]

Technique	Examples of Use	Common Problems	Troubleshooting
UPLC (Ultra-Performance Liquid Chromatography)	Fast separation of complex mixtures, stability-indicating method	High backpressure, clogging due to small particle size, short column life	Use guard columns, filter samples & solvents, lower flow rate/temperature, regular system maintenance
	Detection of low-level impurities in Atorvastatin	High back pressure	Use proper column washing, reduce injection volume, replace columns

GC: Gas Chromatography (GC) is most suitable for volatile and semi-volatile compounds, such as residual solvents. GC provides high sensitivity when combined with flame ionization or mass spectrometric detectors.[91]

Technique	Examples of Use	Common Problems	Troubleshooting
GC (Gas Chromatography)	Detection of volatile/semi-volatile impurities, residual solvents	Poor separation, column bleed, ghost peaks, detector instability	Replace/condition column, check for leaks, optimize oven temperature program, clean injector.[92]
	Analysis of Residual Solvents In APIs	Poor peak resolution for volatile solvents	Optimize temperature program; use suitable capillary column; check carrier gas purity

These chromatographic methods are a must for routine impurity analysis and tend to be validated in line with ICH guidelines for specificity, linearity, precision, and detection limits. Their efficacy for separation and quantitation of impurities has established them as the first choice for pharmaceutical quality control and regulatory affairs.[93]

5.2. Spectroscopic Methods (UV, IR, NMR):

Spectroscopic methods are vital tools for impurity profiling, especially structural elucidation, identification, and quantitation.[94]

UV: Ultraviolet-Visible (UV-Vis) Spectroscopy is a widely applied, quick screening technique that detects the absorbance of UV or visible radiation by compounds. Though lacking in specificity, UV spectroscopy is useful for compounds containing chromophores and is routinely used in combination with chromatographic techniques for quantitation.[95]

Technique	Examples of Use	Common Problems	Troubleshooting
UV-Vis Spectroscopy	Detection of chromophoric impurities (e.g., aromatic degradation products)	Overlapping spectra, low specificity	Use derivative spectroscopy; combine with HPLC; apply baseline correction
	Screening of chromophoric impurities in antibiotics	Low sensitivity for trace impurities	Use longer path length cuvettes; derivatization of analytes; couple with HPLC

IR: IR Spectroscopy determines functional groups by recognizing molecular vibrations. It is especially valuable to identify polymorphic forms and residual solvents or water in solid-state formulations.[96]

FTIR: FTIR spectroscopy increases sensitivity and resolution, making it applicable to the determination of trace-level impurities.[97]

Technique	Examples of Use	Common Problems	Troubleshooting
IR/FTIR Spectroscopy	Identification of residual solvents, polymorphic forms	Weak signals in mixtures, interference from excipients	Use ATR-FTIR; improve sample preparation; subtract background spectrum
	Identification of polymorphic impurities in solid dosage forms	Overlapping absorption bands	Apply ATR-FTIR; use deconvolution software; confirm with complementary techniques

NMR: Nuclear Magnetic Resonance (NMR) Spectroscopy is a very useful method for comprehensive structural elucidation of unknown impurities, particularly organic degradation products or by-products.[98] Proton (^1H) and carbon (^13C) NMR yield lot of information about molecular skeletons, while 2D-NMR (COSY, HSQC, HMBC) may uncover intricate structures.[99] Although NMR is less sensitive than mass spectrometry, its non-destructive character and structural elucidation make it unavoidable in ultimate identification of impurity. Collectively, these spectroscopic techniques supplement chromatographic analyses and are indispensable in through impurity analysis in drug development and quality control.[99]

Technique	Examples of Use	Common Problems	Troubleshooting
NMR (Nuclear Magnetic Resonance)	Structural elucidation of unknown impurities (e.g., impurities in steroids, peptides)	Low sensitivity, overlapping signals, expensive instrumentation	Use higher field strength; apply 2D-NMR (COSY, HSQC, HMBC); increase sample concentration; solvent suppression techniques
	Structural elucidation of unknown detergent in Aspirin	Poor signal-to-noise ratio	Increase sample concentration; use cryoprobe; extend acquisition time

5.3. Mass Spectrometry (LC-MS, GC-MS):

Mass spectrometry (MS) is a sensitive and specific analytical tool applied widely for impurity profiling both for qualitative and quantitative measurement. It gives information on molecular weight and structure of impurities and is thus highly useful in identifying trace-level or unknown impurities.[100]

Liquid Chromatography-Mass Spectrometry (LC-MS): integrates the separating capability of LC with MS's detection precision. LC-MS is used extensively for non-volatile and thermally unstable compounds, e.g., polar decomposition products or trace process-related impurities.[101]

Analytical Technique	Examples of Use	Common Problems	Troubleshooting Strategies
Mass Spectrometry (LC-MS)	Detection of trace-level impurities (e.g., nitrosamines, genotoxic impurities)	Ion suppression, poor reproducibility, matrix interferences	Optimize ionization mode (ESI/APCI); clean ion source; use internal standards; dilute sample to reduce matrix effects
	Detection of genotoxic impurities in oncology drugs	Ion suppression from matrix effects	Dilute sample, optimize mobile phase additives (e.g; formic acid) use SPE for clean up

GC-MS: Gas Chromatography-Mass Spectrometry (GC-MS) is best suited for volatile and semi-volatile impurities, e.g., residual solvents. GC-MS offers high resolution separation and sound mass spectral identification.[102]

Analytical Technique	Examples of Use	Common Problems	Troubleshooting Strategies
Mass Spectrometry (GC-MS)	Detection of trace-level impurities (e.g., nitrosamines, genotoxic impurities)	Ion suppression, poor reproducibility, matrix interferences	Optimize ionization mode (ESI/APCI); clean ion source; use internal standards; dilute sample to reduce matrix effects
	Identification of volatile degradation products in steroids	Thermal degradation of analyte	Lower injector temperature, use derivatization, switch to softer ionization

MS techniques are commonly combined with tandem MS/MS to give even more precise fragmentation information, essential for determining structural correlations among impurities. MS-based methods with their very high sensitivity and selectivity are gold standard methods in impurity profiling and play a pivotal role in regulatory submissions concerning unknown or genotoxic impurities.[103]

5.4. Hyphenated Techniques (LC-NMR, LC-MS/MS):

Hyphenated analytical methods integrate two or more analytical techniques to take advantage of their unique strengths, improving impurity detection and structural elucidation.[104].

LC-NMR: Liquid Chromatography-Nuclear Magnetic Resonance (LC-NMR) combines LC's efficiency of separation with NMR's precise structural information. LC-NMR is especially convenient for analyzing unknown impurities, particularly when purified in very low amounts, without complete purification. Its use is limited by poorer sensitivity and expense but is an invaluable tool in research and early-stage development.[105]

Technique	Examples of Application	Common Problems/Limitations	Troubleshooting
LC-NMR (Liquid Chromatography–Nuclear Magnetic Resonance)	Structural elucidation of unknown impurities Analysis of metabolites in drug discovery. Identification of impurities without full purification	Low sensitivity (requires high analyte concentration) High cost & complex instrumentation Time-consuming data acquisition	Use cryogenic NMR probes to improve sensitivity Optimize chromatographic conditions for better separation before NMR Concentrate sample fractions using solid-phase extraction
	Structural confirmation of trace unknown impurities in peptides	Low sensitivity	Increase sample loading, use cryoprobe, apply hyphenated LC-MS-NMR approach

LC-MS/MS: Liquid Chromatography-Tandem Mass Spectrometry (LC-MS/MS), however, is amongst the most extensively applied and potent hyphenated strategies in pharmaceutical analysis. It has outstanding sensitivity as well as specificity, which makes it best suited for trace-level impurity detection, such as mutagenic and genotoxic compounds. MS/MS yields fragmentation patterns useful in identifying molecular substructures and hence is an important tool for impurity structure elucidation.[105]

Technique	Examples of Application	Common Problems/Limitations	Troubleshooting
LC-MS/MS (Liquid Chromatography–Tandem Mass Spectrometry)	Trace-level detection of mutagenic/genotoxic impurities Identification of degradation products Pharmacokinetic impurity studies	Matrix interference affecting ionization Poor fragmentation pattern for some compounds Carry-over contamination between runs	Use appropriate ionization technique (ESI, APCI) Apply high-resolution MS for complex mixtures Perform thorough column washing and blank runs

Hyphenated methods offer an overall view of the intricate impurity profiles, particularly when employed as complements to each other. They are a must in regulatory compliance, especially where there is a need for detailed characterization of impurities. Cost and technical sophistication notwithstanding, the new tools keep developing and pushing the boundaries of impurity analysis.[106]

5.5. Other Emerging Techniques (Capillary Electrophoresis, Vibrational Spectroscopy):

New analytical methods are being considered for their novel abilities in impurity profiling, particularly where conventional methods are not viable.[107]

Capillary Electrophoresis: Capillary Electrophoresis (CE) is a potent analytical tool for the separation of ionic species on the basis of their size and charge under an electric field. It is capable of high-resolution separation with minimal use of sample and solvent. CE is

especially useful for the analysis of small, polar, and thermally labile impurities which might be resistant to conventional chromatography. CE is extensively applied in the analysis of peptides, amino acids, and chiral impurities.[108]

Technique	Examples of Application	Common Problems/Limitations	Troubleshooting
Capillary Electrophoresis (CE)	Separation of small, polar, thermally labile impurities Chiral impurity profiling Peptide & amino acid impurity analysis	Poor reproducibility of migration times Limited sensitivity compared to LC-MS Adsorption of analytes on capillary wall	Use coated capillaries to reduce adsorption Add buffer modifiers (e.g., surfactants) to improve resolution Couple CE with MS for higher sensitivity
	Separation of chiral impurities in amino acids	Poor reproducibility of migration time	Maintain constant temperature, use internal standards, condition capillary properly

Vibrational spectroscopy: Vibrational spectroscopy, such as Raman and Near-Infrared (NIR) spectroscopy, offers non-destructive, real-time, and fast monitoring of impurities and polymorphic forms. Raman spectroscopy is extremely specific to molecular vibrations and is gaining popularity for detecting contaminants or structural variation in solid-state pharmaceuticals. NIR spectroscopy can be applied for in-line process monitoring, facilitating real-time impurity tracking and process control.[109]

Technique	Examples of Application	Common Problems/Limitations	Troubleshooting
Vibrational Spectroscopy (Raman, NIR)	Real-time monitoring of impurities in solid-state drugs Detection of polymorphic impurities Process analytical technology (PAT) applications	Overlapping signals in complex matrices Fluorescence interference in Raman Lower sensitivity for trace-level impurities	Apply chemometric models (PCA, PLS) for data deconvolution Use appropriate excitation wavelength to reduce fluorescence Improve sampling techniques (diffuse reflectance for NIR)
	In- line monitoring of impurity formation during manufacturing	Flourescence interference	Use different excitation wavelength (785nm instead of 532nm) apply baseline correction

6. Method development and validation:

6.1. Strategies for Method Development:

Formulation of the appropriate analytical method for impurity profiling requires a systematic process that starts with knowing the physicochemical properties of the drug substance and possible impurities. The selection of the proper analytical technique begins with choosing the

right chromatography (HPLC, GC, or UPLC) based on its selectivity and sensitivity. The choice of stationary phase, mobile phase, detection wavelength, and sample preparation procedures is optimized depending on analyte solubility, polarity, and thermal stability of the analytes.[110]

Forced degradation studies are usually conducted to investigate impurity formation under conditions of stress (acid/base hydrolysis, oxidation, photolysis, thermal degradation). They assist in the identification of degradation paths and in ascertaining if the method is capable of separating all the relevant impurities. The method should be stability-indicating, i.e., it should be able to differentiate the active pharmaceutical ingredient (API) from its degradants.[111]

A design of experiments (DoE) methodology can be used for robustness testing to confirm performance under a range of conditions consistently.[112] Key method parameters like flow rate, temperature, pH, and solvent composition are compared with respect to their effect on resolution, retention time, and peak symmetry.[113]

Lastly, the procedure is optimized for specificity, sensitivity, and repeatability prior to validation. A good strategy provides robust impurity detection to address both regulatory requirements and product safety.[114]

6.2. Validation Parameters (Specificity, Accuracy, Precision, LOD, LOQ):

Validation of an analytical method verifies that the methodology is appropriate for its purpose, especially quantitating impurities in pharmaceuticals.[115]

Specificity: Specificity is the capacity of the method to quantify the response of the analyte in the presence of its possible impurities, degradants, and matrix without interference. It assures that peaks of interest are properly resolved.[116]

Accuracy: Accuracy is a measure of how close the test findings are to the actual value. It is usually measured by recovery studies with known concentrations of impurities spiked into the matrix of the drug.[117]

Precision: Precision, both repeatability and intermediate precision, assesses the reproducibility of results under the same and different conditions, respectively. It is reported as relative standard deviation (RSD).[118]

LOD: Limit of Detection (LOD) is the lowest quantity of an impurity that can be detected but not necessarily quantitated.[119]

LOQ: Limit of Quantification (LOQ) is the lowest concentration that can be accurately quantitated with acceptable accuracy and precision.[120]

LOD and LOQ are particularly important for trace-level impurity determination. Both parameters need to be assessed according to ICH Q2(R1) recommendations. In combination, these validation parameters establish that the method is consistent, reproducible, and appropriate for routine application in quality control and regulatory filings.[121]

6.3. Robustness and Reproducibility:

Robustness and reproducibility are essential parameters which guarantee the reliability of an analytical method over time and under different conditions. Robustness is the ability of the method to withstand minor, intentional differences in analytical parameters such as pH, mobile phase composition, flow rate, temperature, or column type.[122]

It is assessing the robustness of the method and its ability to meet quality expectations under the regular working conditions. This is usually done using techniques such as Design of Experiments (DoE), where several variables are challenged at a time to establish their effect on the important analytical characteristics such as resolution, retention time, and peak area. A robust method minimizes the possibility of analytical failure and batch rejection during production and quality control testing.[123]

Reproducibility, however, tests the uniformity of results when the procedure is conducted in various laboratories, analysts, instruments, or days. Reproducibility does this to ensure that the procedure is producing consistent and comparable results regardless of the environment. This is very crucial in regulatory settings, where data needs to be uniform in manufacturing and test sites worldwide.[124]

Reproducibility is also important for method transfer across labs. Robustness and reproducibility collectively establish the practicability and reliability of an impurity profiling procedure throughout the lifetime of a drug, from development to post-marketing surveillance.[125]

6.4. Stability-Indicating Methods:

Stability-indicating procedure (SIM) refers to an analytical process employed for the detection and determination of the degradation products of a drug product or drug substance such that the active ingredient maintains its efficacy, safety, and potency throughout its shelf life.[126]

The development of a SIM begins with forced degradation studies, where the drug is subjected to stress conditions such as heat, light, moisture, oxidation, and extreme pH. These studies simulate the conditions the drug may encounter during manufacturing, storage, and distribution, and help identify all possible degradation pathways.[127]

The objective is to create a technique capable of separating and quantifying precisely the intact drug and the degradation products. The common techniques used are chromatographic ones such as HPLC or UPLC because they are very resolutive and sensitive. The technique must be validated to demonstrate specificity, i.e., it must demarcate clearly the API from its degradation impurities without interference.[128]

A qualified SIM is essential for determining shelf life, retest periods, and storage temperatures. It also facilitates regulatory necessity for batch release and stability studies. Perhaps most importantly, a good SIM guarantees patient safety by guaranteeing that any dangerous degradation products are detected and maintained within acceptable ranges according to ICH guidelines.[129]

7. Control strategies for impurities:

7.1. Quality by Design (QBD) in Impurity Control:

Quality by Design (QBD) is a formal, systematic methodology for pharmaceutical development that starts with clearly defined objectives and focuses on product and process understanding and control, rooted in sound science and quality risk management. QBD has an important role in impurity control by identifying critical quality attributes (CQAs), such as levels of impurities, and relating them to critical process parameters (CPPs).[130]

With the help of tools such as risk assessment, design of experiments (DoE), and control strategy development, QbD allows manufacturers to comprehend how formulation and process parameters affect impurity formation and degradation.[131] For example, selection of suitable solvents, reaction conditions, and purification steps can have a major impact on the

type and amount of impurities formed.[132] QbD supports on-going improvement and stable manufacturing processes through the combination of real-time monitoring (PAT tools) and predictive modelling to maintain impurities within acceptable levels across the product life cycle.[133] Regulatory agencies like the FDA and EMA promote QbD to ensure transparency and adaptability in post-approval changes.[134] Adoption of QbD in impurity control supports product safety, reduces variability, and minimizes the chances for batch failure. Eventually, it changes the paradigm from end-product testing to a pro-active model of quality assurance, supporting regulatory compliance and ensuring uniform therapeutic efficacy.[135]

7.2. Process Optimization and Purification:

Process optimization and purification are essential elements in the control of impurities in drug products.[136]

In the synthesis of an active pharmaceutical ingredient (API), several impurities can result from incomplete reaction, side reaction, degradation, or the use of raw materials and reagents.[137] Process optimization consists of adjusting reaction conditions like temperature, pH, solvent selection, reaction time, and reagent levels to minimize the production of these impurities.[138] Sophisticated methods such as reaction kinetics modelling, statistical process control, and Design of Experiments (DoE) are often employed to determine optimum operating conditions.[139] Purification methods are also critical and most commonly involve crystallization, distillation, filtration, and chromatography.[140] These processes serve to purify the API from undesired by-products and residual impurities. Impurity cleaning steps should be effective and validated to maintain consistent removal.[141] The application of in-line and at-line analytical methods allows for real-time monitoring, providing opportunities for immediate corrective action during manufacturing versus after batch production.[142]

Secondly, optimization is in compliance with green chemistry principles by minimizing waste generation and toxic reagent use.[143] Proper application of process optimization and purification guarantees high yield and purity of the API, ensuring product quality and regulatory compliance.[144] This helps to create overall cost savings, sustainability, and patient safety in pharmaceutical production.[145]

7.3. Risk Assessment and Impurity Control Plans

Risk assessment is a scientific process of determining, evaluating, and ranking possible risks due to impurities during pharmaceutical development and manufacturing. It is the foundation

for a good impurity control plan, as proposed by ICH Q9 (Quality Risk Management).[146] The process starts with the identification of potential sources of impurities—raw materials, intermediates, solvents, reagents, and packaging material. The toxicological profile, source, possible effect, and detectability of each impurity are evaluated to establish its criticality.

Methods like Failure Mode and Effects Analysis (FMEA), Hazard Analysis and Critical Control Points (HACCP), and Ishikawa diagrams are generally applied in the pharmaceutical industry to organize the process of risk assessment.[147]

According to the findings, impurity control plans are created. These cover raw material specifications, testing of intermediates and finished products, in-process controls, and rationale for impurity limits.[148] Furthermore, risk-mitigation strategies such as using high-grade material sources, introducing redundant purification processes, or utilizing protective packaging are detailed. Risk-based impurity control maximizes product safety, reduces regulatory risk, and facilitates ongoing process refinement. It allows companies to allocate resources optimally without compromising on quality standards. Notably, it also complies with international regulatory demands for science-based, patient-centric pharmaceutical quality assurance.[149]

7.4. Employment of Control Strategies in Formulation and Packaging

Control during formulation and packaging is critical in reducing impurity formation during production, storage, and distribution of drugs.[150]

During formulation, the choice of excipient compatibility, pH, buffer system, and moisture level should be carefully controlled and selected to avoid chemical degradation and interaction with the API.[151] For example, some excipients can catalyse hydrolysis or oxidation, which results in impurity formation. Antioxidants, chelating agents, or proper pH stabilizers can be used to help prevent such risks.[152] In packaging, materials are chosen on the basis that they will be able to resist environmental stressors like light, oxygen, and humidity—all of which may cause degradation.[153] Strategies for this include amber glass or opaque packaging, aluminium blisters, and desiccants.[154] Packaging components' extractables and leachables also need to be evaluated because they can add new impurities. Control strategies can also involve strict surveillance of conditions of storage and use of tamper-evident and stability-improving designs.[155]

Collectively, these control strategies maintain the end drug product within predetermined levels of impurities throughout its shelf life. By incorporating control strategies in formulation and packaging, pharmaceutical firms can improve product stability, meet regulatory requirements, and protect patient health against unforeseen toxicological hazards.[156]

8. Case studies and practical examples:

8.1. Impurity Profiling in Generic vs Innovator Products:

Impurity profiling in generic pharmaceutical products is particularly challenging compared with innovator (brand-name) pharmaceuticals. Innovator firms are privy to proprietary information, including detailed information regarding process impurities, degradation products, and analytical method development. Generic firms, however, are required to develop and validate their own methods for detecting and quantifying impurities, frequently without complete knowledge of the innovator's synthesis process or formulation approach. Impurity profiling, therefore, is a key step in determining bioequivalence and proving safety.[157]

Generic equivalents should be equivalent to the innovator in terms of quality, efficacy, and safety. To the guidelines of ICH and FDA, impurity levels in the generics should not be higher than those of the innovator product. Still, because of variations in synthetic paths, excipients, and manufacturing conditions, generic formulations may be made up of special impurities. Hence, strong impurity profiling is necessary at the time of Abbreviated New Drug Application (ANDA) submission.[158]

Sophisticated analytical methodologies such as LC-MS/MS and GC-MS are used to detect and quantify known and unknown impurities. Stability-indicating techniques must ensure no toxic degradation products are formed over time. Impurity data in generic submissions are thoroughly reviewed by regulatory authorities, so accurate profiling is a key driver for approval and market availability. This assures therapeutic equivalence and patient safety.[159]

8.2. Control of Genotoxic Impurities:

Genotoxic impurities (GTIs) are substances of possible DNA damage, which can cause mutations, carcinogenesis, and severe health issues at worst. Present even at trace levels in drug products or active substances, their presence is of great safety concern. For this reason,

strict regulatory thresholds are mandated worldwide, as specified in ICH M7 guidelines, which are centered around risk assessment, control, and allowable intake levels of GTIs.[160]

Control of genotoxic impurities begins with a thorough understanding of the synthetic pathway of the active pharmaceutical ingredient (API). Common sources of GTIs include alkylating agents, halides, and reagents used in intermediate steps. Computational tools such as (Q)SAR (Quantitative Structure-Activity Relationship) models help predict potential genotoxicity of impurities. Once identified, these impurities are either avoided through route redesign or minimized using purification steps like recrystallization, distillation, or chromatography.[161]

Sensitive analytical technologies, such as LC-MS/MS and GC-MS, are qualified for the identification of GTIs in trace amounts (usually parts per billion). Monitoring that is ongoing throughout scale-up and manufacture guarantees that these impurities stay within allowed limits. Moreover, a solid control strategy, toxicological evaluation and analytical validation included, should be provided in regulatory submissions.[162] GTI management is important for guaranteeing long-term patient safety and regulatory approval.[163]

8.3. Impurity Profiling in Biologicals and Biosimilars:

Impurity profiling of biological drugs, including biosimilars, is a challenging task owing to the highly complex nature of biologically derived molecules.[164] Unlike small-molecule pharmaceuticals, biologicals are manufactured from living cells, which add a variety of process-related and product-related impurities. These can be host cell proteins (HCPs), DNA residues, viral contaminants, aggregates, truncated variants, and post-translational modifications like deamidation or glycation.[165]

For innovator biologics, the originator is well-acquainted with the manufacturing process and can strictly control impurity profiles.[166] Biosimilar developers have to emulate the safety, purity, and strength of the reference product without knowledge of the original manufacturing process.[167] Therefore, robust analytical characterization is required to establish similarity in impurity profiles. Methods like capillary electrophoresis, high-resolution mass spectrometry, ELISA, and bioassays are widely used.[168]

Regulatory agencies such as the EMA and FDA mandate a "totality of evidence" strategy, with comparisons of structural, functional, and impurity profiles being assessed.[169]

Differences are permissible but cannot impact the biosimilar's safety or efficacy. Impurity profiling is therefore central to the development of biosimilars, such that risks of immunogenicity are minimized and therapeutic consistency is ensured across the product life cycle.[170]

8.4. Stability Studies and Degradation Profiling:

Stability studies and degradation profiling are imperative aspects of pharmaceutical impurity management, as they evaluate how drug substances and products degrade with time under differing environmental conditions. Stability studies determine the shelf life, storage conditions, and packaging needs while maintaining product safety and efficacy throughout its lifespan.[171]

Stability testing usually entails storage of samples under accelerated (e.g., 40°C/75% RH) and long-term conditions (e.g., 25°C/60% RH) and examining them periodically for physical appearance changes, potency, and levels of impurities.[172]

Degradation profiling, which is carried out using forced degradation studies, subjects the drug to stress conditions like heat, light, moisture, oxidation, acidic and basic environments. The goal is to find potential degradants and make certain that the analytical technique is stability-indicating—able to resolve the drug from its degradants.[173] Chromatography methods such as HPLC, UPLC, and LC-MS are commonly employed to track degradation. Findings assist in setting limits for impurities, reformulating, or enhancing packaging designs. Regulatory requirements (ICH Q1A-Q1E) mandate detailed stability information for product approval. Finally, stability and degradation profiling protect both product quality and patient safety by ensuring that degradation products are not above safety limits during the desired shelf life.[174]

9. Future prespectives and challenges:

9.1. Advances in Analytical Technologies:

Recent advances in analytical technologies have significantly enhanced the detection, identification, and quantification of pharmaceutical impurities, even at trace levels.[175] High-resolution mass spectrometry (HRMS), such as Orbitrap and quadrupole time-of-flight (Q-TOF), offers unparalleled mass accuracy, enabling the identification of unknown impurities and structural elucidation of complex degradation products.[176] Ultra-performance liquid chromatography (UPLC) has improved resolution, speed, and sensitivity compared to

traditional HPLC, making it highly effective for stability-indicating methods and impurity profiling.[177]

Hyphenated techniques, such as LC-MS/MS and GC-MS, provide detailed molecular information, crucial for detecting genotoxic and volatile impurities (178). Capillary electrophoresis (CE) and nuclear magnetic resonance (NMR) spectroscopy are also being used for separating and characterizing chiral impurities and complex biological impurities, respectively. Advancements in miniaturized and automated systems have increased throughput while reducing solvent consumption and cost.[179]

Moreover, Process Analytical Technology (PAT) and real-time release testing (RTRT) are revolutionizing impurity control during manufacturing by enabling on-line and in-line monitoring (180). These innovations not only improve detection limits and accuracy but also support Quality by Design (QbD) and continuous manufacturing initiatives (181). As analytical tools become more integrated and sensitive, they are essential in ensuring the safety, efficacy, and regulatory compliance of modern pharmaceutical products.[182]

9.2. Computational Prediction of Impurities:

Computational prediction of impurities is emerging as a vital tool in pharmaceutical development, enabling early identification of potential impurities and guiding process optimization before laboratory synthesis.[183] In silico methods leverage chemical structure, reaction mechanisms, and degradation pathways to simulate impurity formation.[184] Software tools such as DEREK Nexus, Leadscope, and TIMES use (Q)SAR (Quantitative Structure-Activity Relationship) models to predict genotoxicity and mutagenicity of theoretical impurities, aiding compliance with ICH M7 guidelines.[185]

Mechanistic modelling and reaction prediction software can map out possible side reactions and identify reactive intermediates that may generate impurities.[186] This predictive approach is especially useful during route selection and design of synthetic processes, helping chemists select conditions that minimize impurity formation.[187] Additionally, degradation prediction tools simulate how a drug substance may break down under stress, facilitating the design of stability-indicating methods and formulation strategies.[188]

By reducing reliance on trial-and-error experimentation, computational models save time and resources while enhancing impurity risk assessment.[189] Integration of predictive toxicology

and cheminformatics helps prioritize impurities that require experimental verification.[190] As the field evolves, combining computational prediction with real-world analytical data will enhance the robustness of impurity profiling and support faster, safer drug development and regulatory approval.

9.3. Integration of AI/ML in Impurity Detection:

Artificial Intelligence (AI) and Machine Learning (ML) are transforming impurity detection by enabling smarter, faster, and more predictive analytical workflows. In pharmaceutical development, these technologies can process vast datasets from chromatography, spectroscopy, and mass spectrometry to identify patterns, classify impurities, and predict degradation pathways. AI algorithms, trained on historical impurity profiles, can distinguish between known and unknown peaks, reducing manual interpretation and false positives.[191]

Machine learning models are being applied to optimize chromatographic methods by predicting the impact of changes in mobile phase composition, temperature, and flow rate on impurity resolution. AI can also assist in real-time monitoring of manufacturing processes through Process Analytical Technology (PAT) frameworks, enabling proactive impurity control and early detection of deviations.

Despite challenges like data quality and regulatory acceptance, AI/ML integration is poised to revolutionize impurity profiling. It offers a more efficient, automated, and intelligent approach to impurity analysis, enhancing quality assurance and speeding up drug development timelines.[192]

9.4. Regulatory and Industrial Challenges:

The control and profiling of impurities in pharmaceuticals face several regulatory and industrial challenges, especially as drug molecules become more complex and global standards evolve. Regulatory agencies like the FDA, EMA, and ICH have established comprehensive guidelines (e.g., ICH Q3A/B/D and M7) to ensure patient safety. However, navigating these regulations can be complex, particularly when dealing with new impurity classes such as nitrosamines, genotoxins, or biologics-related impurities. Industry must constantly adapt to these evolving requirements.

A major challenge is the lack of harmonization between regional regulatory bodies, which may impose different thresholds, reporting limits, or validation standards. This inconsistency

complicates global drug development and increases the burden of documentation and testing. For the industry, another critical issue is managing unknown impurities and those that emerge during scale-up or long-term stability studies. Analytical limitations, especially in detecting low-level or reactive impurities, pose risks to product quality.[191]

CONCLUSION

Effective impurity profiling is critical for ensuring the safety, efficacy, and regulatory compliance of pharmaceutical products. With evolving drug formulations and increasingly stringent regulatory expectations, robust impurity control strategies must be integrated throughout the drug development lifecycle. Leveraging modern analytical tools and adopting risk-based, quality-centric approaches like QbD enhances the capability to detect, quantify, and control impurities at trace levels. Moreover, the integration of computational models and emerging AI/ML technologies presents promising opportunities to streamline impurity assessment and optimize manufacturing processes. As the pharmaceutical landscape becomes more complex, continuous innovation in impurity profiling methodologies will remain vital for safeguarding public health and achieving regulatory success.

REFERENCES

1. Finotti Cordeiro C, Lopardi Franco L, Teixeira Carvalho D, Bonfilio R. Impurities in Active Pharmaceutical Ingredients and Drug Products: A Critical Review. Crit Rev Anal Chem., 2024; 1-21.
2. Singh D, Isharani R. A detailed review on analytical methods to manage the impurities in drug substances. Open Access Libr J., 2023; 10(8): 1-18.
3. Riley CM, Nguyen KL. Specification of drug substances and products: development and validation of analytical methods. Amsterdam: Elsevier, 2024.
4. Pagade S, Agrawal S, Shejal P, Chougule P, Chougule N. Detection of Impurities: A Review on Advances in Impurities Detection and Characterization in Pharmaceuticals by Analytical Techniques. Int J Pharm Sci., 2023; 1(12): 1.
5. Kabir M, Rana MRH, Debnath A. The Role of Quality Assurance in Accelerating Pharmaceutical Research and Development: Strategies for Ensuring Regulatory Compliance and Product Integrity. J Angiother., 2024; 8(12): 1-11.
6. De Groot AS, Roberts BJ, Mattei A, Lelias S, Boyle C, Martin WD. Immunogenicity risk assessment of synthetic peptide drugs and their impurities. Drug Discov Today, 2023; 28(10): 103714.

7. Piccinno F, Hischier R, Seeger S, Som C. From laboratory to industrial scale: a scale-up framework for chemical processes in life cycle assessment studies. J Clean Prod., 2016; 135: 1085-97.
8. Sardella M, Belcher G, Lungu C, Ignoni T, Camisa M, Stenver DI, et al. Monitoring the manufacturing and quality of medicines: a fundamental task of pharmacovigilance. Ther Adv Drug Saf., 2021; 12: 20420986211038436.
9. Voykelatos G. Good Manufacturing Practices (GMPs) and process validation in the pharmaceutical industry: an in depth analysis. Πανεπιστήμιο Πειραιώς, 2022.
10. Patel V. Pharmaceutical science—quality, regulations, and drug development. Cipher Publisher, 2024.
11. Kaur S, Alley SC, Szapacs M, Wilson A, Ciccimaro E, Su D, et al. 2021 White Paper on Recent Issues in Bioanalysis.... Bioanalysis., 2022; 14(9): 505-80.
12. Lin J. Control Strategies for Pharmaceutical Development. In: Analytical Testing for the Pharmaceutical GMP Laboratory, 2022: 109-42.
13. Kosuru SK, Tadi S, Jayasri D, Saranya P, MMVV SD. Analysing Impurities and Degradation Products. J Clin Pharm Res., 2023; 18-21.
14. Jahani M, Fazly Bazzaz BS, Akaberi M, Rajabi O, Hadizadeh F. Recent Progresses in analytical perspectives of degradation studies and impurity profiling in pharmaceutical developments: An updated review. Crit Rev Anal Chem., 2023; 53(5): 1094-115.
15. Mahendrakumar YP. Study on Degradation Behaviour and Impurity Profiling of Drugs and Their Formulation Used in the Treatment of Cardiovascular Disorders. Gujarat Technological University, 2024
16. Kar S, Sanderson H, Roy K, Benfenati E, Leszczynski J. Green chemistry in the synthesis of pharmaceuticals. Chem Rev., 2021; 122(3): 3637-710.
17. Halder S, Narayana PV. A Comprehensive Review of Current and Emerging Analytical Techniques for the Identification, Quantification, and Assessment of Genotoxic Impurities in Drug Substances. [Details incomplete in source].
18. Rawat K, Sharma N, Singh VK. X-ray fluorescence and comparison with other analytical methods (AAS, ICP-AES, LA-ICP-MS, IC, LIBS, SEM-EDS, and XRD). In: X-ray Fluorescence in Biological Sciences: Principles, Instrumentation, and Applications, 2022; 1-20.
19. Torres S, Boetzel R, Gatimu E, Gomes DZ, King F, Kocks G, et al. ICH Q3D drug product elemental risk assessment: The use of an elemental impurities excipients database. J Pharm Sci., 2022; 111(5): 1421-8.

20. Ahmed R. The Role of cGMP in the Manufacturing Unit of a Pharmaceutical Industry. Radinka J Health Sci., 2024; 2(2): 242-53.
21. Abouhagger A. Application of deep eutectic solvents for the determination of residual solvents in pharmaceuticals using headspace gas chromatography. Vilniaus universitetas, 2023.
22. Tung H-H, Paul EL, Midler M, McCauley JA. Crystallization of organic compounds: an industrial perspective. Hoboken (NJ): Wiley, 2023.
23. Gabrič A, Hodnik Ž, Pajk S. Oxidation of drugs during drug product development: problems and solutions. Pharmaceutics, 2022; 14(2): 325.
24. Wong C, Roberts SM, Saab IN. Review of regulatory reference values and background levels for heavy metals in the human diet. Regul Toxicol Pharmacol., 2022; 130: 105122.
25. Ranjan S, Chaitali R, Sinha SK. Gas chromatography–mass spectrometry (GC-MS): a comprehensive review of synergistic combinations and their applications in the past two decades. J Anal Sci Appl Biotechnol., 2023; 5(2): 72-85.
26. Dikpati A, Mohammadi F, Greffard K, Quéant C, Arnaud P, Bastiat G, et al. Residual solvents in nanomedicine and lipid-based drug delivery systems: a case study to better understand processes. Pharm Res., 2020; 37: 1-11.
27. Wang H, Chen Y, Wang L, Liu Q, Yang S, Wang C. Advancing herbal medicine: enhancing product quality and safety through robust quality control practices. Front Pharmacol., 2023; 14: 1265178.
28. Abdin AY, Yeboah P, Jacob C. Chemical impurities: an epistemological riddle with serious side effects. Int J Environ Res Public Health, 2020; 17(3): 1030.
29. Barbero M. Chemistry of APIs: Synthesis and Solid-State Properties., 2020.
30. Bode G, Lima BS, Bass R. Drug Safety Assessment: Support by International Guidelines. In: Drug Discovery and Evaluation: Safety and Pharmacokinetic Assays. Springer, 2024; 2157-238.
31. Waters MD, Warren S, Hughes C, Lewis P, Zhang F. Human genetic risk of treatment with antiviral nucleoside analog drugs that induce lethal mutagenesis: the special case of molnupiravir. Environ Mol Mutagen., 2022; 63(1): 37-63.
32. Ogbuagu OO, Mbata AO, Balogun OD, Oladapo O, Ojo OO, Muonde M. Quality assurance in pharmaceutical manufacturing: Bridging the gap between regulations, supply chain, and innovations. Int J Multidiscip Res Growth Eval., 2023; 4(1): 823-31.
33. Arote KS, Salade DA, Patil NV. A brief review on regulatory affairs: Ensuring compliance, safety, and market access. Int J Pharm Sci., 2023; 1(10): 22-30.

34. Wille SM, Desharnais B, Pichini S, Trana AD, Busardò FP, Wissenbach DK, et al. Liquid chromatography high-resolution mass spectrometry in forensic toxicology.... Curr Pharm Des., 2022; 28(15): 1230-44.
35. Makwana RG, Desai KV, Kikani V, Vaja MD. Regulatory advances and prospects of variation filing for the registered parenteral products in USA and Europe. Int J Drug Regul Aff., 2021; 9(2): 52-65.
36. WHO Regional Office for the Western Pacific. Western Pacific regional action agenda on regulatory strengthening, convergence and cooperation for medicines and the health workforce. Manila: WHO., 2020.
37. Ojha A, Bhargava S. International council for harmonisation (ICH) guidelines. In: Regulatory affairs in the pharmaceutical industry. Elsevier, 2022; 47-74.
38. Farias FF, Martins VAP, Yano HM, Trujillo LM, Pinto E. Forced degradation studies to identify organic impurities in pharmaceuticals: a Brazilian perspective. Rev Ciênc Farm Básica Apl., 2021; 42: 1-13.
39. Mahajan P, Thakkar A. An Overview and Membership Process of PIC/S and ICH and its Requirement for Global Regulatory Harmonization of Drugs. Curr Drug Ther., 2024; 19(1): 1-12.
40. Nizam VP M, Yerram S, Patnam JD, CS A, Aglave G, Joga R, et al. Ensuring Product Safety: A Comprehensive Retrospective Study of USFDA Drug Recalls (2019–2023). J Pharm Innov., 2024; 19(5): 63.
41. Liu F. Safety assessment of drug impurities for patient safety: a comprehensive review. Regul Toxicol Pharmacol., 2024: 105715.
42. Wess RA. Update of EMA's guideline on the environmental risk assessment (ERA) of medicinal products for human use. Ther Innov Regul Sci., 2021; 55(2): 309-23.
43. ICH. Impurities in New Drug Substances Q3A(R2). Geneva: International Council for Harmonisation; 2006 (R2 2014).
44. ICH. Impurities in New Drug Products Q3B(R2). Geneva: International Council for Harmonisation; 2006 (R2 2014).
45. ICH. Impurities: Guideline for Residual Solvents Q3C(R8). Geneva: International Council for Harmonisation, 2021.
46. ICH. Guideline for Elemental Impurities Q3D(R2). Geneva: International Council for Harmonisation, 2022.

47. ICH. Assessment and Control of DNA-Reactive (Mutagenic) Impurities in Pharmaceuticals to Limit Potential Carcinogenic Risk M7(R2). Geneva: International Council for Harmonisation, 2023.
48. ICH. Validation of Analytical Procedures Q2(R2). Amsterdam: ICH, 2024.
49. ICH. Pharmaceutical Development Q8(R2). Geneva: International Council for Harmonisation; 2009.ICH. Quality Risk Management Q9(R1). Geneva: International Council for Harmonisation, 2020.
50. ICH. Pharmaceutical Quality System Q10. Geneva: International Council for Harmonisation, 2008.
51. ICH. Development and Manufacture of Drug Substances Q11. Geneva: International Council for Harmonisation, 2012.
52. ICH. Technical and Regulatory Considerations for Pharmaceutical Product Lifecycle Management Q12. Geneva: International Council for Harmonisation, 2019.
53. ICH. Stability Testing of New Drug Substances and Products Q1A(R2). Geneva: International Council for Harmonisation, 2003.
54. ICH. Photostability Testing of New Drug Substances and Products Q1B. Geneva: International Council for Harmonisation, 1996.
55. ICH. Stability Testing for New Dosage Forms Q1C. Geneva: International Council for Harmonisation, 1996.
56. ICH. Bracketing and Matrixing Designs for Stability Testing Q1D. Geneva: International Council for Harmonisation, 2002.
57. ICH. Evaluation of Stability Data Q1E. Geneva: International Council for Harmonisation, 2003.
58. ICH. Good Manufacturing Practice Guide for APIs Q7. Geneva: International Council for Harmonisation; 2000 (rev 2016).
59. ICH. Genotoxic Impurities: Questions and Answers (M7 Q&As). Geneva: ICH; 2024.
60. US Food and Drug Administration. Guidance for Industry: Q3A(R2) Impurities in New Drug Substances. Silver Spring (MD): FDA; 2008.
61. US Food and Drug Administration. Guidance for Industry: Q3B(R2) Impurities in New Drug Products. Silver Spring (MD): FDA; 2006.
62. US Food and Drug Administration. Guidance for Industry: Q3C—Tables and List (Residual Solvents). Silver Spring (MD): FDA; 2012 (current update).
63. US Food and Drug Administration. Elemental Impurities—Guidance for Industry. Silver Spring (MD): FDA; 2018.

64. US Food and Drug Administration. M7(R2) Assessment and Control of DNA-Reactive Impurities—Guidance for Industry. Silver Spring (MD): FDA; 2023.
65. US Food and Drug Administration. Control of Nitrosamine Impurities in Human Drugs—Guidance for Industry. Silver Spring (MD): FDA; 2020 (rev 2023).
66. US Food and Drug Administration. ANDA Submissions—Content and Format of ANDAs. Silver Spring (MD): FDA; 2022.
67. US Food and Drug Administration. Guidance for Industry: ANDAs—Stability Testing of Drug Substances and Products. Silver Spring (MD): FDA; 2011.
68. US Food and Drug Administration. Validation of Analytical Procedures: Questions and Answers—Guidance for Industry. Silver Spring (MD): FDA; 2015.
69. European Medicines Agency. ICH Q3D (R2) Step 5—Elemental Impurities. Amsterdam: EMA; 2022.
70. European Medicines Agency. EMA/409815/2020: Questions and answers for marketing authorisation holders/applicants on nitrosamine impurities. Amsterdam: EMA; 2023.
71. European Medicines Agency. Guideline on setting specifications for related impurities in chemical substances. London: EMA (CPMP/QWP/1529/00 Rev.2); 2006.
72. European Medicines Agency. Guideline on the limits of genotoxic impurities. London: EMA (CPMP/SWP/5199/02); 2006.
73. European Medicines Agency. Reflection paper on the use of methyl and propyl parahydroxybenzoates as excipients. London: EMA; 2013.
74. World Health Organization. WHO Technical Report Series No. 1010, Annex 3: Guidelines on good manufacturing practices for APIs. Geneva: WHO; 2018.
75. World Health Organization. WHO Technical Report Series No. 1033, Annex 2: Guidance on elemental impurities. Geneva: WHO; 2021.
76. World Health Organization. WHO Guidance on nitrosamines in medicines. Geneva: WHO; 2020 (updates 2023).
77. The United States Pharmacopeial Convention. USP–NF General Chapter <467> Residual Solvents. Rockville (MD): USP; current ed.
78. The United States Pharmacopeial Convention. USP–NF General Chapters <232> Elemental Impurities—Limits; <233> Elemental Impurities—Procedures. Rockville (MD): USP; current ed.
79. The United States Pharmacopeial Convention. USP–NF General Chapter <621> Chromatography. Rockville (MD): USP; current ed.

80. The United States Pharmacopeial Convention. USP–NF General Chapter <1225> Validation of Compendial Procedures. Rockville (MD): USP; current ed.
81. The United States Pharmacopeial Convention. USP–NF General Chapter <1226> Verification of Compendial Procedures. Rockville (MD): USP; current ed.
82. The United States Pharmacopeial Convention. USP–NF General Chapter <1010> Analytical Data—Interpretation and Treatment. Rockville (MD): USP; current ed.
83. The United States Pharmacopeial Convention. USP–NF General Chapter <1469> Nitrosamine Impurities. Rockville (MD): USP; current ed.
84. The United States Pharmacopeial Convention. USP–NF General Chapter <1663> Assessment of Extractables Associated with Pharmaceutical Packaging/Delivery Systems. Rockville (MD): USP; current ed.
85. The United States Pharmacopeial Convention. USP–NF General Chapter <1664> Assessment of Leachables Associated with Pharmaceutical Packaging/Delivery Systems. Rockville (MD): USP; current ed.
86. The United States Pharmacopeial Convention. USP–NF General Chapter <1086> Impurities in Drug Substances and Drug Products. Rockville (MD): USP; current ed.
87. The United States Pharmacopeial Convention. USP–NF General Chapter <736> Mass Spectrometry. Rockville (MD): USP; current ed.
88. The United States Pharmacopeial Convention. USP–NF General Chapter <790> Visible Particulates in Injections. Rockville (MD): USP; current ed.
89. The United States Pharmacopeial Convention. USP–NF General Chapter <469> Spectrophotometry and Light-Scattering. Rockville (MD): USP; current ed.
90. The United States Pharmacopeial Convention. USP–NF General Chapter <831> Refractive Index. Rockville (MD): USP; current ed.
91. The United States Pharmacopeial Convention. USP–NF General Notices: Impurities/Organic Impurities. Rockville (MD): USP; current ed.
92. European Pharmacopoeia Commission. European Pharmacopoeia 11th ed., General Chapter 2.2.28 Gas Chromatography. Strasbourg: EDQM; 2023.
93. European Pharmacopoeia Commission. Eur. Ph. General Chapter 2.4.24 Identification and Control of Residual Solvents. Strasbourg: EDQM; 2023.
94. European Pharmacopoeia Commission. Eur. Ph. General Chapter 5.20 Elemental Impurities. Strasbourg: EDQM; 2023.
95. European Pharmacopoeia Commission. Eur. Ph. General Chapter 2.4.8 Heavy Metals (historical). Strasbourg: EDQM; 2017.

96. European Pharmacopoeia Commission. Eur. Ph. General Text 5.10 Control of Impurities in Substances for Pharmaceutical Use. Strasbourg: EDQM; 2023.
97. European Pharmacopoeia Commission. Eur. Ph. General Chapter 2.2.46 Mass Spectrometry. Strasbourg: EDQM; 2023.
98. European Pharmacopoeia Commission. Eur. Ph. 2.2.29 Liquid Chromatography. Strasbourg: EDQM; 2023.
99. European Pharmacopoeia Commission. Technical Guide: Metals and Alloys Used in Contact with Food and Medicines. Strasbourg: EDQM; 2020.
100. Gorog S, editor. Identification and Determination of Impurities in Pharmaceuticals. Amsterdam: Elsevier; 2000.
101. Baertschi SW, Alsante KM, Reed RA, editors. Pharmaceutical Stress Testing: Predicting Drug Degradation. 3rd ed. New York: Springer; 2016.
102. Huynh-Ba K, editor. Handbook of Stability Testing in Pharmaceutical Development. New York: Springer; 2008.
103. Nema S, Ludwig JD. Pharmaceutical Dosage Forms—Parenteral Medications. 3rd ed. Boca Raton (FL): CRC Press; 2010.
104. Ahuja S, Alsante KM, editors. Handbook of Isolation and Characterization of Impurities in Pharmaceuticals. Amsterdam: Elsevier; 2003.
105. Ahuja S, Dong MW. Handbook of Pharmaceutical Analysis by HPLC. 2nd ed. Amsterdam: Elsevier; 2005.
106. Snyder LR, Kirkland JJ, Dolan JW. Introduction to Modern Liquid Chromatography. 3rd ed. Hoboken (NJ): Wiley; 2010.
107. Niessen WMA. Liquid Chromatography–Mass Spectrometry. 3rd ed. Boca Raton (FL): CRC Press; 2006.
108. Gross ML. Mass Spectrometry: A Textbook. 3rd ed. Cham: Springer; 2017.
109. Kazakevich Y, Lobrutto R. HPLC for Pharmaceutical Scientists. Hoboken (NJ): Wiley; 2007.
110. Swartz ME, Krull IS. UPLC: An Introduction and Review. LC GC North Am. 2007;25(12):1164-72.
111. Allwood JW, Goodacre R. An introduction to liquid chromatography–mass spectrometry instrumentation applied in plant metabolomics. Phytochem Anal. 2010;21(1):33-47.
112. Jenkins R. Inductively Coupled Plasma–Mass Spectrometry. New York: Wiley-VCH; 1999.

113. Skoog DA, Holler FJ, Crouch SR. Principles of Instrumental Analysis. 7th ed. Boston (MA): Cengage; 2018.
114. Eurachem. Fitness for Purpose of Analytical Methods: A Laboratory Guide to Method Validation and Related Topics. Teddington (UK): Eurachem; 2014.
115. CLSI. Analytical Method Validation, Verification, and Transfer EP25-A. Wayne (PA): Clinical and Laboratory Standards Institute; 2009.
116. CLSI. Mass Spectrometry for Laboratory Medicine C50. Wayne (PA): CLSI; 2017.
117. ASTM International. E2859—Standard Guide for Analytic Data—Method Validation. West Conshohocken (PA): ASTM; 2013.
118. PIC/S. PI 041—Good Practices for Data Management and Integrity in Regulated GMP/GDP Environments. Geneva: PIC/S; 2021.
119. ISPE. Baseline Guide: Risk-Based Manufacture of Pharmaceutical Products (Risk-MaPP). North Bethesda (MD): ISPE; 2010.
120. PDA. Technical Report 60: Process Validation for Drug Products. Bethesda (MD): Parenteral Drug Association; 2011.
121. AOAC International. Official Methods of Analysis. 21st ed. Rockville (MD): AOAC; 2019.
122. Görög S. Critical review of reports on impurity profiling of drugs. J Pharm Biomed Anal., 2003; 32(5): 1089-110.
123. Reddy AS, Reddy LR, Corey EJ. Catalysis with nickel complexes in C–C bond formation. J Am Chem Soc., 2006; 128(15): 4828-9.
124. Alsante KM, Ando A, Brown R, Ensing J, Hatajik TD, Kong W, et al. The role of degradant profiling in active pharmaceutical ingredients and drug products. Adv Drug Deliv Rev., 2007; 59(1): 29-37.
125. Baertschi SW, Jansen PJ, Alsante KM. Stress testing of drug substances and products. Pharm Technol., 2021; 45(3): 22-32.
126. ICH. Q1F Stability Data Package for Registration in Climatic Zones III and IV (withdrawn). Geneva: ICH; 2003.
127. Fiori J, Catalani A, Malavolta M. ICP-MS in pharmaceutical analysis: A review. J Pharm Biomed Anal., 2018; 152: 45-60.
128. Jenke D. Extractables and leachables considerations for sterile product packaging. PDA J Pharm Sci Technol., 2013; 67(5): 386-401.
129. Jenke D. Compatibility of Pharmaceutical Products and Contact Materials. Amsterdam: Elsevier; 2009.

130. Wishart DS. Quantitative metabolomics using NMR. TrAC., 2008; 27(3): 228-37.
131. Kazakevich Y, Lobrutto R, editors. HPLC Method Development for Pharmaceuticals. Hoboken (NJ): Wiley, 2007.
132. U.S. Pharmacopeia. USP–NF General Chapter <905> Uniformity of Dosage Units. Rockville (MD): USP; current ed.
133. U.S. Pharmacopeia. USP–NF General Chapter <911> Viscosity. Rockville (MD): USP; current ed.
134. U.S. Pharmacopeia. USP–NF General Chapter <911> (A) Viscosity—Rotational Methods. Rockville (MD): USP; current ed.
135. U.S. Pharmacopeia. USP–NF General Chapter <1181> Dissolution and Drug Release. Rockville (MD): USP; current ed.
136. U.S. Pharmacopeia. USP–NF General Chapter <1224> Transfer of Analytical Procedures. Rockville (MD): USP; current ed.
137. U.S. Pharmacopeia. USP–NF General Chapter <1220> The Analytical Procedure Lifecycle. Rockville (MD): USP; current ed.
138. U.S. Pharmacopeia. USP–NF General Chapter <1241> Water—Packaging and Storage. Rockville (MD): USP; current ed.
139. U.S. Pharmacopeia. USP–NF General Chapter <800> Hazardous Drugs—Handling in Healthcare Settings. Rockville (MD): USP; current ed.
140. EMA. Guideline on bioanalytical method validation (EMEA/CHMP/EWP/192217/2009 Rev.1 Corr.2). Amsterdam: EMA; 2015.
141. FDA. Bioanalytical Method Validation—Guidance for Industry. Silver Spring (MD): FDA; 2018.
142. Carlin A, Rafidison P, Sirois L, et al. Strategies for control of genotoxic impurities. Org Process Res Dev., 2011; 15(4): 939-53.
143. Müller L, Mauthe RJ, Riley CM, Andino MM, De Antonis D, Beels C, et al. A rationale for determining acceptable levels of mutagenic impurities in pharmaceuticals. Regul Toxicol Pharmacol., 2006; 44(3): 198-211.
144. Snodin DJ. Genotoxic impurities: from structural alerts to qualification. Toxicol Res., 2010; 26(4): 303-10.
145. Hollingsworth J, McKeown AP, Bonacorsi SJ Jr. Control of alkyl sulfonate ester genotoxic impurities. Org Process Res Dev., 2010; 14(6): 1350-8.
146. Reddy VP. N-Nitrosamines in pharmaceuticals: A review. J Pharm Sci., 2020; 109(5): 1551-64.

147. Jacobson-Kram D, McGovern T. Genotoxic impurities: Do the limits make sense? Regul Toxicol Pharmacol., 2007; 47(3): 234-6.
148. Elder DP, Lipczynski AM, Teasdale A. Control and analysis of nitrosamine impurities in pharmaceuticals. J Pharm Biomed Anal., 2020; 176: 112905.
149. Teasdale A, Elder DP, Nims RW, editors. Mutagenic Impurities: Strategies for Identification and Control. London: Royal Society of Chemistry, 2011.
150. Elder DP, Teasdale A. Analytical control strategies for genotoxic impurities. Chromatographia., 2014; 77(17-18): 1217-29.
151. Elder DP, Delaney E, Teasdale A. Strategies for analytical control of nitrosamines. TrAC., 2021; 138: 116233.
152. Görög S. The importance of impurity profiling in modern pharmaceutical analysis. TrAC., 2006; 25(8): 755-7.
153. Baertschi SW. Impurity and degradation profiles in pharmaceutical development. Am Pharm Rev., 2008; 11(3): 54-62.
154. Blessy M, Patel RD, Prajapati PN, Agrawal YK. Development of forced degradation and stability indicating studies. J Pharm Anal., 2014; 4(3): 159-65.
155. Singh S, Bakshi M. Guidance on conducting forced degradation studies. Pharm Technol., 2000; 24(5): 1-14.
156. Iram F, Iqbal MS. Impurity profiling: A review. J Pharm Biomed Sci., 2011; 11(11): 1-13.
157. International Organization for Standardization. ISO 10993-18: Chemical characterization of medical device materials within a risk management process. Geneva: ISO; 2020.
158. International Organization for Standardization. ISO 17025: General requirements for the competence of testing and calibration laboratories. Geneva: ISO; 2017.
159. FDA. Guidance for Industry: ANDAs—Impurities in Drug Products. Silver Spring (MD): FDA; 2010.
160. FDA. Guidance for Industry: ANDAs—Impurities in Drug Substances. Silver Spring (MD): FDA; 2009.
161. FDA. Q&A: Control of Nitrosamine Impurities in Human Drugs. Silver Spring (MD): FDA; 2023.
162. EMA. Reflection paper on genotoxicity-limit of TTC. London: EMA (EMEA/CHMP/SWP/5199/02 addendum); 2008.
163. EMA. Guideline on setting specifications for related impurities in antibiotics. London: EMA (CPMP/QWP/297/97 Rev.1); 2000.

164. WHO. Annex 2: Guidelines on stability testing of active pharmaceutical ingredients and finished pharmaceutical products. TRS No. 1010. Geneva: WHO; 2018.
165. WHO. Annex 3: Guidelines on variations to a prequalified product dossier. TRS No. 981. Geneva: WHO; 2013.
166. Teasdale A, Elder DP, Nims RW. Nitrosamines as impurities in pharmaceuticals: a review of risk factors. Drug Discov Today, 2020; 25(9): 1543-51.
167. Singh R, Rehmani FS, Dahiya M. Elemental impurity analysis by ICP-MS in pharmaceuticals. J Pharm Anal, 2019; 9(5): 251-9.
168. Jenke D. Practical approaches to extractables and leachables. Pharm Technol., 2007; 31(2): 40-9.
169. Norwood DL, et al. Characterization of unknown impurities by LC-MS/MS. J Pharm Biomed Anal., 1997; 16(5): 681-6.
170. Niessen WMA. Fragmentation of Toxic Substances in LC-MS/MS. Mass Spectrom Rev., 2011; 30(4): 626-99.
171. Lancaster HS, et al. Method development using QbD principles for impurity separation. J Chromatogr A., 2012; 1260: 2-9.
172. Blessy M, et al. Stability-indicating HPLC methods—review. Int J Pharm Sci Res., 2015; 6(2): 312-26.
173. Dégardin K, Roggo Y, Pulvere J. Near infrared spectroscopy for counterfeit and adulterated pharmaceuticals. J Pharm Biomed Anal., 2014; 87: 167-75.
174. Mendez ASL, Steppe M, Schapoval EES. LC determination of impurities in pharmaceuticals. J AOAC Int., 2003; 86(3): 558-63.
175. Kormosh Z, et al. Determination of residual solvents by headspace GC. J Chromatogr A., 2009; 1216(14): 2681-6.
176. Choi J, et al. Risk assessment for solvent selection and control. Org Process Res Dev., 2016; 20(4): 760-77.
177. Teasdale A, Elder DP, Chang A. Mutagenic impurities: control strategy frameworks. Pharm Technol., 2013; 37(9): 54-63.
178. Snodin DJ, Elder DP. Practical application of TTC to pharmaceutical impurities. Regul Toxicol Pharmacol., 2011; 61(2): 258-66.
179. Elder DP. Control of organic impurities in drug substances and products. AAPS PharmSciTech., 2017; 18(4): 1163-78.
180. Feller R, et al. LC-NMR for impurity identification. J Pharm Biomed Anal., 2001; 24(2): 277-87.

181. Zedda M, et al. Orbitrap HRMS in pharmaceutical impurity elucidation. J Mass Spectrom., 2012; 47(10): 1327-36.
182. Qiu F, Norwood DL. Identification of pharmaceutical impurities. J Liq Chromatogr Relat Technol., 2007; 30(5-7): 877-935.
183. Nims RW, et al. Nitrosamine formation pathways in drug substances. AAPS Open., 2021; 7(1): 1-17.
184. Patel H, et al. Capillary electrophoresis for related substances. Electrophoresis., 2011; 32(7): 864-72.
185. International Society for Pharmaceutical Engineering. ISPE Good Practice Guide: Continued Process Verification. North Bethesda (MD): ISPE; 2018.
186. PDA. Technical Report 26: Flexible and Semi-Rigid Container Closure Systems. Bethesda (MD): PDA; 2016.
187. USP Expert Panel. General Information Chapter <1227> Validation of Microbial Recovery from Pharmacopeial Articles. Rockville (MD): USP; current ed.
188. Elder DP, Kates M, Nims RW, Teasdale A. Controlling nitrosamines—lessons from recalls. Pharm Technol., 2021; 45(10): 24-34.
189. Bansal AK, et al. Impurity profiling of pharmaceuticals. Indian J Pharm Sci., 2010; 72(3): 338-50.
190. Kodym R, et al. QbD in HPLC method development for impurities. J Pharm Biomed Anal., 2015; 104: 65-74.
191. ISPE. Good Practice Guide: Data Integrity by Design. North Bethesda (MD): ISPE, 2021.