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Perspective

Method-at-a-Glance (MAG) as a Graphical Approach for Method Development in Reversed-Phase Liquid Chromatography of Pharmaceuticals

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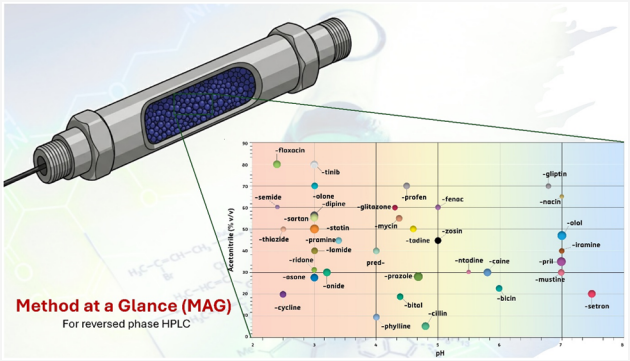
ABSTRACT: Reversed-phase high-performance liquid chromatography (RP-HPLC) remains the dominant separation mode in pharmaceutical and biomedical analysis, yet method development is still largely driven by trial-and-error practices, fragmented literature knowledge, or data-intensive chemometric tools. Despite decades of accumulated chromatographic experience, this knowledge is rarely transferred in a form that enables rapid, rational, and reproducible decision-making at the early stages of method development. This Perspective introduces Method-at-a-Glance (MAG), a conceptual, literature-encoded graphical framework that reimagines RP-HPLC method development as a visually guided process grounded in collective chromatographic knowledge. By abstracting and mapping reported chromatographic conditions for structurally related pharmaceutical classes, MAG enables the rapid identification of rational starting conditions, significantly reducing the scope of preliminary experimentation. Rather than proposing a new analytical method, this Perspective synthesizes the existing literature into a transferable visual intelligence tool and discusses its implications for accelerating method development, improving reproducibility, and reducing experimental burden. The broader opportunities of MAG are explored, including its potential role in AI-assisted chromatography and sustainability-oriented analytical design. MAG exemplifies how analytical chemistry can move beyond the isolated method reporting toward knowledge-integrated, concept-driven frameworks that support the evolving needs of modern laboratories.



INTRODUCTION

HPLC dominates pharmaceutical and biomedical analysis due to its ease of use, precision, and selectivity.[1] Various HPLC modes have been applied in drug analysis, including ion exchange, ion exclusion,[2] size exclusion, hydrophilic interaction,[3] normal phase, and reversed phase. Among these, reversed-phase HPLC (RP-HPLC) is the most prevalent, owing to its superior efficiency in separating nonpolar analytes using aqueous−organic mobile phases.

RP-HPLC offers high flexibility, as its parameters such as the stationary phase and mobile phase composition can be tailored to specific analytical requirements. The development of a robust RP-HPLC method demands careful optimization and validation to ensure reproducibility, sensitivity, and accuracy. As such, method development is a crucial phase in achieving high-quality data for applications in pharmaceuticals, biotechnology, and environmental monitoring.

In chemical analysis, method development is the most critical step, and can be time-consuming and complex.[4,5] Traditional development relies on trial-and-error, informed by the analyst's expertise and preliminary knowledge about the analyte. A common initial strategy involves using an isocratic mobile phase composed of 50:50 v/v organic to aqueous solvent, with subsequent adjustments to mobile phase strength based on the observed separation. An alternative method employs gradient elution (e.g., 5−100% methanol at pH 2.5) followed by fine-tuning of % organic content and buffer pH to achieve isocratic conditions yielding retention factors between 0.5 and 20.[6]

Although trial-and-error approaches can be useful, particularly when combined with expertise from previous failures, they are often inefficient, costly, and time-intensive. Additionally, such approaches can be frustrating and may lead to unintended experimental outcomes.

Chemometric techniques have also been used for method optimization, offering the ability to evaluate multiple variables simultaneously and predict optimal conditions from a limited

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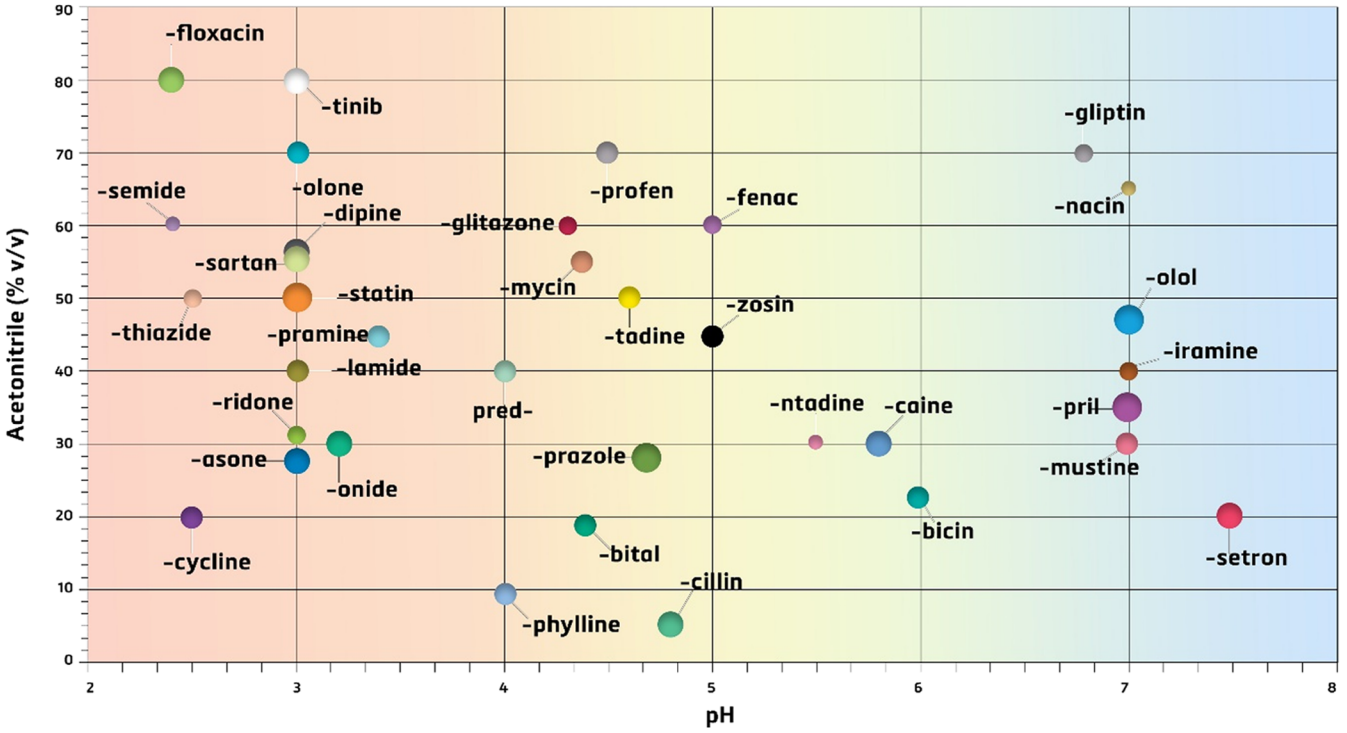

Figure 1. Graphical representation of recommended chromatographic conditions for different drug classes using RP-HPLC.

set of experiments.[7] However, this approach requires expertise in modeling software and statistical analysis. Moreover, the requirement for a large number of experimental data points can be a limitation. Chemometric tools also do not inherently incorporate the extensive empirical knowledge available in chromatographic literature.

The present study aims to introduce a novel graphical approach to RP-HPLC method development based on a comprehensive review of published methods. This approach facilitates method development and minimizes experimental workload. It also assists in selecting appropriate gradient programs when separating chemically diverse drug classes and helps in choosing internal standards for biomedical analyses. This Method-at-a-Glance (MAG) approach can be a useful tool in quality control laboratories.

EXPERIMENTAL SECTION

Chemicals and Materials

Valsartan (99.80%), irbesartan (99.80%), telmisartan (99.80%), atorvastatin (99.80%), amoxicillin (99.80%), and diclofenac sodium (99.90%) were kindly provided by Sigma Pharmaceutical Industries (Quesna, Egypt). HPLC-grade methanol and acetonitrile (ACN), as well as potassium dihydrogen phosphate, were purchased from Merck (Darmstadt, Germany). Sodium hydroxide and phosphoric acid were obtained from Piochem (sixth of October City, Egypt). Deionized water was used for the preparation of all aqueous solutions.

Instrumentation

All chromatographic analyses were performed using a Dionex UltiMate 3000 HPLC system (Thermo Scientific Dionex, Sunnyvale, CA, USA), equipped with an LPG-3400SD quaternary pump, a WPS-3000TSL autosampler, a TCC-3000SD column thermostat, and a DAD-3000 diode array detector. Data acquisition and processing were carried out using Chromeleon 7 software. Weighing was conducted using an AXIS balance (Model ALN220, Gdańsk, Poland). A Jenway 3510 pH meter (Cole-Parmer, Saint Neots, UK) was used for pH measurements. An ultrasonic cleaning unit (Elmasonic S 60 H) was employed for mobile phase degassing, and a SUEZ filtration system (Paris, France) was used for water deionization.

Chromatographic Conditions

The stationary phase used was a Thermo Hypersil ODS C18 column (250 mm × 4.6 mm, 5 μm particle size). For gradient method development, a mobile phase consisting of ACN and phosphate buffer (50 mM, pH 4.80) in a ratio of 5:95 (v/v) was delivered at a flow rate of 1.50 mL/min. The injection volume was 20 μL, and detection was performed at 210 nm using a UV detector. For internal standard selection, a mobile phase composed of ACN and phosphate buffer (50 mM, pH 3.00) in a 50:50 (v/v) ratio was used, delivered at a flow rate of 1.00 mL/min. The injection volume was also 20 μL, and the UV detector was set at 230 nm.

Standard and Working Solutions

Stock standard solutions of valsartan, irbesartan, telmisartan, and atorvastatin were individually prepared at a concentration of 1 mg/mL in acetonitrile. Appropriate aliquots of each were combined to prepare an aqueous working solution containing 10 μg/mL of each drug. A stock solution of amoxicillin was prepared at 1 mg/mL in deionized water, while diclofenac sodium was dissolved at 1 mg/mL in methanol. Aliquots from these stock solutions were mixed to prepare an aqueous working solution containing 10 μg/mL of each compound.

RESULTS AND DISCUSSION

MAG aims to provide a user-friendly framework that simplifies and accelerates method development. This approach was based on a graph that summarizes the chromatographic conditions suitable as a starting point for a new HPLC method, based on extensive literature survey. The inclusion criteria included RP-HPLC methods that employ modified silica column as a stationary phase, ACN as a mobile phase modifier running in an isocratic mode, and phosphate buffer as the aqueous phase. Methods that employed gradient elution, chemical derivatization, methanol or organic modifiers other than acetonitrile, or buffer systems other than phosphate were excluded. The

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methods included have to be applied for the separation of pharmaceutical compounds from the same pharmaceutical class. If more than one method was found in literature, priority would be given to the method that could separate more compounds from the same class in one single run. Each method is presented on the graph as a single point, whose diameter varies according to the number of papers/compounds supporting the specified chromatographic conditions.

Developing a robust and efficient HPLC method is essential for the accurate and reproducible analysis of pharmaceutical compounds. This study presents a novel graphical approach to HPLC method development, grounded in an extensive literature review encompassing various drug classes. The proposed MAG approach is based on two assumptions; first, compounds within a pharmaceutical class often share core structural motifs (e.g., the '-sartan' scaffold) which frequently leads to similar chromatographic behavior, making class-based predictions a valid hypothesis, though not infallible, as discussed below in the Limitations section. The second assumption is that structurally similar compounds are separated using the same chromatographic mode, under similar conditions, as supported by literature review and the data in Table S1.

MAG's Graph Development

To develop the graph used in MAG, a literature review was conducted, yielding 68 relevant research articles that met the inclusion criteria. We specifically selected isocratic methods as they provide a single, defined mobile phase composition for a given separation, which serves as an ideal coordinate (%ACN, pH) for mapping a compound class. All selected studies employed RP-HPLC using a C18 column and an ACN/phosphate buffer or ACN/water mobile phase.

The MAG framework uses these literature-derived isocratic conditions as foundational coordinates. For a single analyte or a closely related mixture, the corresponding point on the graph (Figure 1) provides a direct recommendation for isocratic method development. For mixtures of analytes from distinct classes that reside in different regions of the map, these coordinates provide the critical boundary conditions for designing a gradient elution program. The starting and ending %ACN can be rationally selected based on the positions of the target analytes, transforming the map from a lookup table for isocratic conditions into a design space for gradient methods. The suggested starting mobile phase for any condition consists of a mixture of ACN and either phosphate buffer or water. The exact ratio of aqueous to organic components, along with the buffer pH, can be determined directly from Figure 1. The data from which the graph was extracted are shown in Table S1.

The structural similarity among compounds within a drug class can be used to predict retention behavior and elution order based on physicochemical properties such as hydrophobicity, polarity, and molecular interactions with the stationary phase. This similarity also aids in optimizing RP-HPLC parameters including gradient profile, flow rate, column temperature, and buffer pH by using a model compound or class representative.

Although derivatization is commonly employed in liquid chromatography to enhance detection sensitivity or chromatographic behavior,[8] methods involving precolumn derivatization were excluded from our study. These methods often introduce additional complexity, such as the need for specific reagents and equipment, increased sample preparation time, and potential sources of error or interference.[9]

For detection, we recommend the use of UV detectors operating at 210 nm a wavelength at which many pharmaceutical compounds exhibit strong absorbance. This wavelength is also compatible with a broad range of solvents and mobile-phase additives, making it a practical choice for general-purpose RP-HPLC analysis.

Chromatographic Mode of Separation. RP-HPLC is widely recognized as the most commonly employed chromatographic mode, particularly in pharmaceutical and biomedical analysis. Aguilar et al.[10] attributed its exceptional popularity to several advantages, including its capacity to achieve high-resolution separations of structurally diverse and closely related compounds under a wide range of chromatographic conditions. RP-HPLC also offers flexible selectivity, as adjustments in mobile phase composition can fine-tune separation efficiency. Moreover, it allows for high recovery even at ultramicroanalytical levels and supports repeated analyses with excellent reproducibility due to the stability of modern stationary phases under diverse mobile phase conditions. In addition to its robust performance, RP-HPLC offers excellent cost-efficiency and the capability to characterize physicochemical interactions between solutes, eluents, and hydrophobic stationary phases. This provides insights into molecular interactions that may affect retention and selectivity, contributing to better method optimization. Another significant advantage of RP-HPLC is its compatibility with a wide variety of detection systems, including UV, fluorescence, electrochemical, and mass spectrometry detectors. According to a report by Žuvela et al., RP-HPLC accounts for over 90% of all HPLC separations worldwide.[11] Additionally, RP-HPLC is user-friendly and does not require toxic, corrosive, or expensive solvents or buffers.[12] Its columns offer long service life and stability, as they are generally resistant to mobile phase-induced degradation.[13] Given these advantages, RP-HPLC was selected as the optimal chromatographic mode for the MAG approach.

Mobile Phase Organic Modifier. In HPLC, the mobile phase is one of the most critical parameters influencing chromatographic performance. In RP-HPLC, the mobile phase typically consists of a mixture of a polar solvent commonly water and a nonpolar organic modifier such as ACN, methanol, or tetrahydrofuran.[14,15]

The primary requirement for a mobile phase is its ability to dissolve and elute the analytes at concentrations compatible with the detection method. Among commonly used organic modifiers, ACN offers several physicochemical advantages that make it highly suitable for RP-HPLC. One of the key advantages of ACN is its lower UV cutoff compared to methanol, making it more suitable for detection at low UV wavelengths. Furthermore, ACN-water mixtures exhibit lower viscosity than methanol−water mixtures, resulting in reduced backpressure during chromatographic runs. This is advantageous for preserving column integrity and prolonging instrument lifespan, while also allowing for higher flow rates and shorter analysis times.

Another important factor is the elution strength of ACN. When mixed with water in equal proportions, ACN generally provides stronger elution power than methanol, which enables more efficient separation of hydrophobic compounds with similar polarity.[16] As such, ACN contributes to improved resolution and faster elution. For all these reasons, ACN was

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selected as the optimal organic modifier for mobile phase development in MAG.

Mobile Phase Aqueous Component. While water is commonly used as the aqueous component of the mobile phase in RP-HPLC, it is often combined with organic solvents such as ACN to modulate polarity and improve separation. However, for analytes with ionizable functional groups, buffers are typically employed to control the pH and stabilize the ionization state of the analytes, thereby improving retention time reproducibility and peak shape.[17]

A variety of buffering agents are available for use in RP-HPLC, including phosphate, citrate, acetate, tris-(hydroxymethyl)aminomethane (TRIS), and borate. The choice of buffer depends on the pK_a of the analytes and the desired pH range. It is generally advised to avoid buffers composed of weak acids or bases with pH values near their pK_a, as such conditions result in partial ionization, leading to poor peak shapes and inconsistent retention times. For optimal reproducibility, the mobile phase pH should be adjusted to at least two units above or below the analyte's pK_a.[18]

Phosphate buffer is particularly advantageous due to its three dissociation constants, which allow for effective buffering across a broad pH range. This versatility makes it suitable for a wide variety of compounds.[19−25] Additionally, phosphate buffer exhibits a UV cutoff around 210 nm at pH ranges of 2.0−3.0 and 6.0−8.5, meaning it does not interfere with UV detection at wavelengths above 210 nm. These characteristics along with its solubility in water and compatibility with organic solvents like ACN and methanol make phosphate buffer a widely adopted choice in RP-HPLC. It ensures stable pH conditions, supports consistent analyte ionization, enhances peak symmetry, and improves separation reproducibility. Given its comprehensive advantages, phosphate buffer was selected as the universal aqueous component of the mobile phase in this study.

Interpreting the MAG Graph. In the MAG representation, the horizontal axis corresponds to the percentage of acetonitrile (%ACN) in the mobile phase under isocratic conditions. This axis reflects mobile-phase elution strength in reversed-phase chromatography, with lower %ACN values corresponding to more polar mobile phases and stronger retention of hydrophobic analytes, and higher %ACN values indicating increased elution strength and reduced retention times. Movement along this axis therefore represents adjustments in solvent strength as a primary driver of analyte elution.

The vertical axis represents the pH of the aqueous component of the mobile phase, typically a phosphate buffer. This axis captures the influence of analyte ionization on retention and selectivity, particularly for compounds containing ionizable functional groups. Vertical positioning on the graph reflects the pH range most commonly reported in the literature for achieving stable retention, acceptable peak shape, and reproducible separation within a given pharmaceutical class. The background color gradient transitions from red to blue as pH increases. This color coding is designed to provide an intuitive visual cue for pH selection.

Each data point on the MAG graph corresponds to one or more literature-reported RP-HPLC methods applied to one or more compounds within a specific pharmaceutical class. The position of the point indicates the %ACN and buffer pH employed in that method, while the bubble size conveys the relative breadth of applicability of the reported conditions. Larger bubbles represent more consensus or methods that successfully separated a greater number of structurally related compounds within a single chromatographic run or more evidence, suggesting higher robustness and transferability of the conditions. Smaller bubbles reflect methods optimized for fewer analytes or narrower applications.

To extract a starting condition using MAG, the analyst first identifies the pharmaceutical or chemical class of interest in the graph. The cluster of bubbles associated with that class defines a region of high probability for successful separation. A practical starting point can be selected by choosing conditions near the center of this cluster, where literature consensus is strongest. When multiple clusters overlap or lie in close proximity, this spatial relationship can inform decisions regarding internal standard selection or gradient design for multicomponent separations.

Importantly, the MAG graph should be interpreted as a guidance tool rather than a predictive model. The recommended conditions serve as rational entry points that reduce uncertainty and experimental burden, but they do not eliminate the need for subsequent fine-tuning to account for matrix effects, column-specific selectivity, or detector considerations. By translating dispersed literature knowledge into an intuitive visual format, MAG enables analysts to move from empirical guessing toward informed decision-making at the outset of RP-HPLC method development, especially at the very beginning.

Beyond its practical utility in guiding initial conditions, the MAG framework inherently embodies the principles of Green Analytical Chemistry (GAC).[26] By consolidating decades of published knowledge into a single, visual starting point, MAG significantly decreases the reliance on extensive trial-and-error experimentation during the early stages of method development. This systematic reduction in the number of preliminary runs translates directly into tangible sustainability benefits, including lower consumption of organic solvents, reduced energy expenditure from instrument operation, and minimized generation of hazardous chemical waste.[27] In this context, MAG contributes to broader sustainability goals, most notably the United Nations Sustainable Development Goal (SDG) 12: Responsible Consumption and Production, by promoting resource-efficient laboratory practices and reducing the environmental footprint of pharmaceutical analysis.[28] It also applies the principles of circular economy by exploiting the massive data in literature to save time, effort and chemicals in method development.[29]

Applications for MAG

Utilizing MAG is highly recommended for the development of RP-HPLC methods for several compelling reasons. First, it significantly reduces time and effort by allowing researchers to directly consult the recommended chromatographic conditions specific to each drug class, serving as an efficient starting point for method development. Moreover, the suggested conditions pose minimal risk to both analytical instruments and users.

In the analysis of drugs within biological matrices, the use of an internal standard is essential for ensuring the accurate quantification of analytes. Typically, the internal standard is introduced during the early stages of sample preparation and extraction, allowing it to undergo the entire analytical process alongside the analyte. This approach helps compensate for potential variability during both preparation and analysis. The overview presented in Figure 1 offers a strategic advantage by identifying drug classes with similar chromatographic con-

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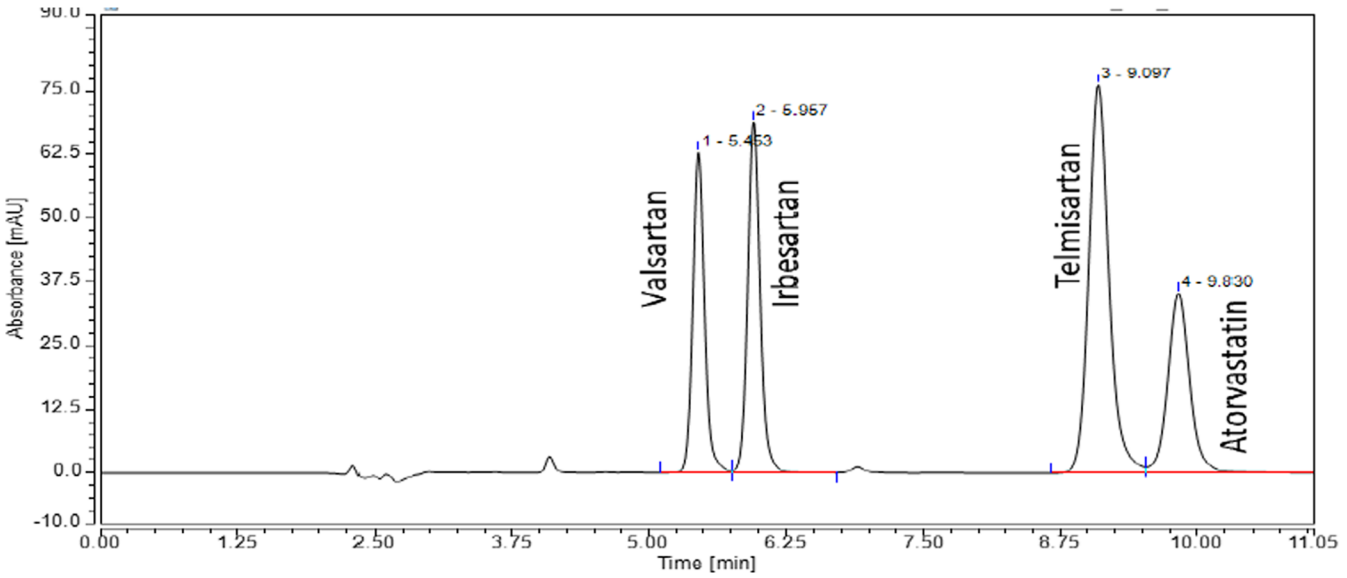

Figure 2. Application of the valsartan separation method to telmisartan, irbesartan and atorvastatin. Column: Thermo Hypersil ODS C18 (250 mm × 4.6 mm, 5 μm); Mobile phase: phosphate buffer (50 mM, pH 3.00):ACN (50:50, v/v); flow rate: 1.00 mL/min; injection volume: 20 μL; UV detection: 230 nm.

ditions, thereby facilitating the selection of appropriate internal standards.

To illustrate this concept, we considered the analysis of valsartan a widely used antihypertensive agent as a case study. Based on Figure 1, sartans appear in close proximity to statins, suggesting compatible chromatographic behavior. Consequently, atorvastatin was tested using the chromatographic conditions established for valsartan[30] and was found suitable for use as an internal standard. Furthermore, the same method was applied, with minor adjustments, to other sartan drugs such as telmisartan and irbesartan. It demonstrated effective separation and resolution for both drugs within the same class and for a structurally distinct compound located within the same chromatographic region (Figure 2). This application shows that the MAG approach provides an evidence-based starting point for method development and that further fine optimization may be necessary to enhance resolution, selectivity, and separation efficiency depending on the specific analytical context.

The MAG graph also reveals regions where pharmaceutical classes do not share similar chromatographic space, as well as areas where multiple classes cluster together. In situations where no pharmaceutical class elutes under conditions sufficiently close to the target analyte, alternative strategies must be employed. Users may consider selecting a structural analog of the target compound, even if it belongs to a different therapeutic class, provided it shares similar physicochemical properties. Alternatively, minor adjustments to the mobile phase composition (e.g., ± 5% ACN or ± 0.5 pH units) can be explored to shift the retention of a potential internal standard candidate into the desired window without compromising separation integrity. In cases where multiple classes overlap within the same chromatographic region, secondary selection criteria become essential. These may include comparing UV spectral similarity using diode array detection, evaluating chemical structure relatedness, or assessing commercial availability and cost.

The work offers substantial value for the analysis of complex mixtures of pharmaceutical compounds. In cases where the mobile phase composition or polarity must vary during the chromatographic run to accommodate multiple analytes with differing chemical properties, gradient elution becomes necessary. Developing a gradient elution RP-HPLC method for separating two drugs with distinct retention behaviors requires careful optimization of the ACN gradient.

A gradient approach is particularly advantageous when the analytes elute at markedly different ACN concentrations. This ensures effective separation and improved peak resolution. Typically, the gradient begins with a lower ACN concentration to retain the more polar compound, followed by a gradual increase in ACN to elute the less polar analyte. This strategy enhances peak symmetry, shortens analysis time, and minimizes peak broadening. By fine-tuning the gradient parameters, the method can achieve reproducible, high-resolution separations suitable for pharmaceutical applications.

The visual reference provided in Figure 1 facilitates the selection of appropriate mobile phase composition and pH for each analyte. This allows analysts and researchers to rationally design a gradient method tailored to the separation of multicomponent mixtures. As a proof of concept, a gradient RP-HPLC method was developed for the simultaneous separation of two compounds from different drug classes. According to Figure 1, the -cillin and -fenac classes fall within a similar pH range but differ significantly in their required % ACN for elution. Specifically, -cillins elute at approximately 5% ACN,[31] while -fenacs require around 60% ACN.[32] Based on this information, a gradient method was developed using an ACN range from 5 to 60% for the separation of amoxicillin and diclofenac sodium, as outlined in Table S2. The method demonstrated successful separation, with amoxicillin eluting at 3.00 min and diclofenac sodium at 6.34 min, achieving acceptable resolution and minimal baseline drift (Figure S1). These proof-of-concept applications demonstrate the utility of MAG for internal standard selection and gradient design.

Limitations and Scope

While the MAG approach offers a structured and visually intuitive approach to initiating RP-HPLC method develop-

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ment, several limitations should be recognized to appropriately define its scope and applicability. MAG is designed as a knowledge-guided starting framework, not as a substitute for experimental optimization, regulatory validation, or application-specific fine-tuning.

First, MAG relies on the assumption that pharmaceutical compounds within the same therapeutic or chemical class exhibit broadly similar chromatographic behavior. Although this assumption is well supported by structure−retention relationships,[33,34] exceptions may arise due to subtle differences in functional groups, ionization behavior, or molecular conformation. Consequently, MAG should be viewed as a probabilistic guidance tool rather than a deterministic predictor of optimal conditions.

Second, the current implementation of MAG is intentionally restricted to conventional RP-HPLC conditions, specifically C18 stationary phases, acetonitrile-based mobile phases, and phosphate buffer systems. While this constraint enhances comparability and transferability, it limits direct applicability to alternative chromatographic modes, such as hydrophilic interaction chromatography, mixed-mode separations, or methods employing nonphosphate buffers or unconventional organic modifiers.

Third, the graphical presentation does not explicitly account for secondary variables that can influence chromatographic performance, including column brand-specific selectivity, particle morphology, temperature effects, detector-specific considerations, or matrix-induced interactions. These factors remain critical during later stages of method refinement and must be addressed through targeted experimentation.

Fourth, the predictive utility of MAG may diminish for certain complex chemical entities. The framework operates most reliably for compounds where retention is governed by a dominant, shared hydrophobic core (e.g., the steroid backbone of corticosteroids). It may provide suboptimal starting points for: (1) Zwitterions or molecules with multiple, competing ionization states (e.g., many peptide-based drugs), where retention exhibits a highly nonlinear and pH-sensitive relationship that a single (%ACN, pH) coordinate cannot capture; (2) Compounds within exceptionally broad mechanistic classes where the common suffix belies significant structural diversity, such as monoclonal antibodies (″-mab″); and (3) Molecules whose chromatography is dominated by specific secondary interactions (e.g., metal chelation, strong $\pi-\pi$ interactions) not primarily controlled by hydrophobic or simple ionization-suppression mechanisms. For such challenging analytes, analysts are advised to consider alternative chromatographic modes better suited to their physicochemical properties, such as ion-exchange chromatography, hydrophilic interaction liquid chromatography (HILIC), or size-exclusion chromatography; extending the MAG framework to encompass these modalities represents a key direction for future research.

When MAG's guidance is expected to be less reliable, or when initial screening based on MAG is not very successful, analysts should treat this not as an approach failure but as a diagnostic signal. It indicates that the analyte's behavior deviates from the generalized class pattern. Finally, MAG reflects the state of the published literature at the time of compilation and is therefore subject to inherent literature biases, including underrepresentation of unsuccessful methods or unpublished optimization attempts. As such, the framework benefits from periodic updating and expansion as new chromatographic data become available.

Recognizing these limitations is essential for the responsible application of MAG. When used within its intended scope; as an informed entry point into method development rather than an end point, the MAG approach provides meaningful value while preserving the rigor and adaptability required in modern analytical chemistry.

CONCLUSION

The MAG approach illustrates how decades of accumulated chromatographic knowledge can be transformed from fragmented, case-specific reports into a coherent, visually accessible decision-support model. By abstracting literature-derived RP-HPLC conditions into a graphical representation, MAG shifts the early stages of method development from empirical trial-and-error toward knowledge-guided initiation, offering a practical response to some aspects of inefficiencies in chromatographic practice. Looking forward, MAG highlights a broader opportunity for analytical chemistry: the transition from isolated method optimization toward knowledge-integrated analytical design. Graphical presentation enables analysts to rapidly identify rational starting conditions, reducing redundant experimentation, solvent consumption, and instrument burden. In this context, MAG aligns naturally with emerging priorities in sustainability, reproducibility, and operational efficiency, particularly in pharmaceutical and quality-control laboratories where RP-HPLC remains indispensable.

Beyond its immediate application, MAG may serve as a conceptual precursor to digital and AI-assisted chromatographic tools. The underlying principle, encoding expert knowledge into structured visual space, can be extended to computational platforms capable of continuously updating chromatographic maps as new literature becomes available. Such systems could support adaptive method development, and hybrid workflows that integrate human expertise with machine learning, rather than replacing either. To realize this vision, we propose transitioning the MAG framework from a static publication-based figure to an open-source, community-driven digital resource at bit.ly/mag2026. Such a platform would consist of an extensible database of chromatographic conditions, linked to a dynamically generated visualization that updates annually as new literature is curated. This would not only ensure the continued accuracy and relevance of the guidance but also invite contributions from the broader analytical community, transforming MAG into a living tool that evolves alongside the field.

Importantly, MAG is not intended to supplant experimental optimization, chemometrics, or regulatory validation frameworks. Instead, it complements these approaches by addressing a critical gap at the earliest decision-making stage of method development, where informed starting conditions exert a disproportionate influence on overall efficiency and success. As analytical challenges grow in complexity, approaches that prioritize knowledge reuse and visual cognition will become increasingly valuable. The broader implication of this Perspective is that analytical chemistry stands to benefit from reconsidering how knowledge is communicated and operationalized. By moving beyond static method descriptions toward dynamic, integrative representations, the field can accelerate innovation while preserving rigor. MAG represents one such step toward a future in which chromatographic

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expertise is not only accumulated, but systematically translated into accessible, transferable analytical intelligence.

■ ASSOCIATED CONTENT

Supporting Information

The Supporting Information is available free of charge at https://pubs.acs.org/doi/10.1021/acs.analchem.5c07982.

Acetonitrile percentage, phosphate buffer composition, and pH used for each drug (Table S1); gradient elution program specifically developed for the separation of amoxicillin and diclofenac sodium (Table S2); and an overlay of chromatograms for the separation of amoxicillin and diclofenac sodium (Figure S1) (PDF)

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Notes

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