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Demonstrating ophthalmic bioequivalence: A comprehensive PK/PD Study Framework under FDA and EMA guidance with MIDD as an enabling tool

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Abstract

Ophthalmic eye drops present one of the most demanding scenarios in bioequivalence (BE) science. Drug concentrations at the local site of action — the cornea, aqueous humor, and uveal tissues — are largely inaccessible through conventional plasma-based pharmacokinetic (PK) sampling. In response, the U.S. Food and Drug Administration (FDA) and the European Medicines Agency (EMA) have developed product-specific, graduated regulatory frameworks that deploy three classes of in vivo evidence for ophthalmic BE: aqueous humor PK studies, pharmacodynamic (PD) endpoint studies using intraocular pressure (IOP) or clinical outcome measures, and plasma PK studies for systemically absorbed drugs. These approaches are operationalized through the FDA's Product-Specific Guidance (PSG) system and the EMA's product-specific bioequivalence guideline (PSBGL) framework. This article presents a rigorous, guideline-anchored review of how PK and PD study methodologies are applied to demonstrate ophthalmic BE, using multiple drug class examples including prostaglandin analogues (latanoprost), alpha-2 agonists (brimonidine), corticosteroids (loteprednol etabonate, prednisolone acetate), and anti-infectives. We further describe how Model-Informed Drug Development (MIDD) — particularly physiologically-based ocular PK (PBOPK) modeling and population PK analysis — functions as a scientifically enabling tool within, not instead of, the existing regulatory BE framework. An optimal study design and regulatory submission strategy aligned with current FDA and EMA requirements is proposed, with explicit guidance on when PK versus PD endpoints are appropriate, how BE statistical criteria are applied in each context, and what MIDD can contribute at each stage.

Keywords: Ophthalmic Bioequivalence; Aqueous Humor PK; IOP Pharmacodynamic Endpoint; FDA Product-Specific Guidance; EMA PSBGL; PBOPK Modeling; Latanoprost; Loteprednol; Brimonidine; Generic Eye Drops; ANDA; MIDD

1. Introduction

Generic ophthalmic drug products — eye drops in particular — occupy a critical position in global healthcare, treating glaucoma, infections, inflammation, allergy, and dry eye disease in tens of millions of patients. The ophthalmic generic market exceeds USD 6 billion annually and is growing, driven by the expiry of foundational molecule patents such as those covering latanoprost, travoprost, bimatoprost, and the fluoroquinolone anti-infectives. Yet bringing a generic ophthalmic product to market through an Abbreviated New Drug Application (ANDA) or a generic Marketing Authorisation Application (MAA) remains disproportionately difficult compared to oral solid dosage forms [1].

The root cause is methodological: the site of pharmacological action for most ophthalmic drugs is not the systemic circulation but local ocular tissues. Plasma drug concentrations following topical ophthalmic dosing are either undetectable or reflect nasolacrimal absorption rather than ocular bioavailability and are demonstrably insensitive to formulation differences [2]. This renders standard plasma PK-based BE criteria — 90% confidence interval of the

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geometric mean T/R ratio within 80.00–125.00% for AUC and Cmax — inapplicable as a primary BE measure for locally acting ophthalmic products.

Both FDA and EMA have responded with drug- and formulation-specific guidance that prescribes which combination of in vitro physicochemical tests, in vivo PK studies (aqueous humor or plasma), PD endpoint studies (IOP, clinical outcomes), and quality equivalence approaches are required for each reference product. The FDA operationalizes this through its Product-Specific Guidance (PSG) database, which currently covers over 80 ophthalmic reference products. The EMA uses its product-specific bioequivalence guideline (PSBGL) framework alongside the general Guideline on Investigation of Bioequivalence (CPMP/EWP/QWP/1401/98 Rev. 1). It should be noted that the EMA Guideline on Quality and Equivalence of Locally Applied, Locally Acting Cutaneous Products (EMA/CHMP/QWP/708282/2018, adopted October 2024, in effect April 2025), which replaced the 2018 draft guideline on topical products, applies primarily to cutaneous (skin) preparations. Its principles may be considered relevant to ophthalmic products by analogy, but the guideline explicitly addresses cutaneous use and does not constitute mandatory guidance for eye drops.

The central thesis of this article is that understanding how PK and PD studies are used within these guideline frameworks — and where MIDD can support, optimize, and in limited circumstances supplement them — is the key competency required for successful ophthalmic generic development. We present this framework through the lens of real regulatory guidance documents and published FDA scientific papers, using specific drug class examples to illustrate study design and BE criteria.

2. Regulatory Framework for Ophthalmic BE: FDA and EMA

2.1. FDA Approach: The Product-Specific Guidance (PSG) System

The FDA's Office of Generic Drugs (OGD) publishes Product-Specific Guidances for ANDA submissions that prescribe, on a drug-by-drug basis, the recommended BE study design and acceptance criteria. For ophthalmic products, PSGs have been available since 2015 and now cover most commercially significant reference products. The fundamental principle underlying FDA's PSG framework for ophthalmic products, as articulated by Choi and Lionberger (2016) from FDA's Office of Research and Standards, is that 'the type of study that can be used to demonstrate bioequivalence depends on the drug product's active pharmaceutical ingredient(s), dosage form, indication, site of action, mechanism of action, and scientific understanding of drug release/drug availability' [1].

For ophthalmic solutions with full Q1 (qualitative) and Q2 (quantitative) sameness — meaning identical inactive ingredients at concentrations within ±5% of the reference listed drug (RLD) — FDA may grant a waiver of the in vivo BE study requirement. Any deviation from Q1/Q2 sameness triggers the requirement for in vivo BE evidence, which must be selected from a hierarchy of approaches defined in the applicable PSG.

Table 1 General overview of FDA regulatory positions on ophthalmic bioequivalence

FDA Core Position on Ophthalmic BE (PSGs, 2015–2023)
For ophthalmic solutions: Q1/Q2 sameness + in vitro physicochemical equivalence = BE waiver. Any Q1/Q2 deviation requires in vivo BE study. For non-solutions (suspensions, emulsions, gels): BE must be demonstrated by one or more of the following — (1) clinical endpoint study; (2) PK study in aqueous humor; (3) microbial kill rate study [anti-infectives]; (4) in vitro comparative physicochemical characterization (Q3 approach). The choice is product-specific and prescribed in the PSG.

2.2. EMA Approach: CPMP/EWP/QWP/1401/98 Rev. 1 and PSBGLs

The EMA's primary BE guideline (CPMP/EWP/QWP/1401/98 Rev. 1, 2010) is principally designed for systemic products, but Appendix II acknowledges topical ophthalmic products and refers applicants to product-specific bioequivalence guidelines. The EMA's Pharmacokinetics Working Party (PKWP) has published PSBGLs for a growing number of ophthalmic products covering both solutions and suspensions [4].

For ophthalmic products, EMA guidance aligns closely with FDA in recognizing that systemic PK endpoints are inappropriate for locally acting drugs. An important regulatory update must be acknowledged here: the former draft Guideline on Quality and Equivalence of Topical Products (CHMP/QWP/708282/2018) has been superseded by the final EMA Guideline on Quality and Equivalence of Locally Applied, Locally Acting Cutaneous Products (EMA/CHMP/QWP/708282/2018 Corr.1), adopted by CHMP on 1 October 2024 and entering into force on 2 April 2025.

Critically, this final guideline explicitly narrows its mandatory scope to cutaneous (skin) products. While its text notes that the principles "may also be relevant for other topical medicines, e.g. preparations for auricular or ocular use," this is an advisory statement, not a binding requirement. For ophthalmic eye drops specifically, this guideline does not constitute applicable mandatory EMA guidance. The governing EMA instruments for ophthalmic BE remain: the Guideline on Investigation of Bioequivalence (CPMP/EWP/QWP/1401/98 Rev. 1), applicable EMA product-specific bioequivalence guidelines (PSBGLs) [5,6].

2.3. The BE Study Hierarchy for Ophthalmic Products

Synthesizing FDA PSG data and EMA guidelines, the current BE study hierarchy for topical ophthalmic products can be represented as a graduated tier model:

Table 2 Graduated bioequivalence study tier model for topical ophthalmic products: from biowaiver through aqueous humor pharmacokinetic study to pharmacodynamic clinical endpoint study, with regulatory basis and applicable dosage forms

Tier	BE Approach	Applicable Dosage Forms	Regulatory Basis
Tier 1 — Biowaiver	Q1/Q2 sameness + in vitro physicochemical tests (pH, viscosity, osmolality, surface tension, droplet size)	Aqueous solutions (Q1/Q2 compliant)	FDA PSGs; EMA PSBGL; EMA CPMP/EWP/QWP/1401/98; EMA/CHMP/QWP/708282/2018
Tier 2 — Aqueous Humor PK Study	In vivo PK in aqueous humor (AUC, Cmax); study design: single dose, cataract surgery patients, parallel group, destructive sampling	Suspensions, gels, emulsions (Q1/Q2 compliant but non-solution)	FDA PSGs for corticosteroid & anti-infective suspensions; EMA PSBGLs
Tier 3 — Pharmacodynamic Endpoint Study	IOP reduction (glaucoma drugs) or clinical endpoint (anti-inflammatories, anti-allergics); crossover or parallel group in patients; 90% or 95% CI criteria	Solutions or suspensions where Q1/Q2 sameness is absent or AH PK is not sensitive	FDA PSGs: brimonidine, brinzolamide, timolol; EMA PSBGLs for glaucoma drugs
Tier 4 — Clinical Endpoint Study	Pivotal efficacy endpoint (e.g., visual acuity, fluorescein staining in dry eye; bacterial eradication in infection); non-inferiority design	Complex products, novel delivery systems, Q1/Q2 non-compliant products	FDA PSGs: nepafenac suspension; cyclosporine emulsion (Restasis); EMA guidelines
Tier 5 — Plasma PK Study	Conventional plasma PK (AUC, Cmax); 90% CI within 80–125%	Drugs with meaningful systemic absorption (e.g., carbonic anhydrase inhibitors with systemic exposure)	FDA: applicable only where plasma concentrations are measurable and sensitive to formulation differences

3. Aqueous Humor PK Studies: Design, Methodology and MIDD Support

3.1. Scientific Rationale

For ophthalmic suspensions and other non-solution dosage forms where Q1/Q2 sameness is achieved, FDA PSGs for several corticosteroid suspensions (prednisolone acetate, loteprednol etabonate, dexamethasone) and anti-infective suspensions recommend demonstrating BE through an in vivo PK study measuring drug concentrations in aqueous

humor (AH). AH drug concentrations represent the most directly accessible pharmacokinetic measure of anterior segment drug exposure, and unlike plasma concentrations, they are expected to be sensitive to formulation-specific differences in particle size distribution, dissolution rate, and corneal permeability [7].

The scientific justification for AH PK as a BE endpoint rests on the assumption that AH drug exposure correlates with pharmacological effect at the intended target tissues (uvea, trabecular meshwork, ciliary body for glaucoma drugs; cornea and anterior uvea for anti-inflammatories). For corticosteroids, where the PD endpoint — anterior chamber flare — is difficult to quantify reproducibly, AH PK provides a more objective, quantifiable, and statistically efficient alternative [8].

3.2. Study Design: FDA PSG Requirements

The FDA PSG for loteprednol etabonate ophthalmic gel 0.38% (Lotemax® Gel, NDA 208219) provides a fully detailed AH PK study design template that is representative of the FDA's approach across corticosteroid products. The key design elements are:

Table 3 Aqueous humor pharmacokinetic study design template for ophthalmic corticosteroids under FDA Product-Specific Guidance: key design elements, PSG specifications, and regulatory rationale (reference product: loteprednol etabonate ophthalmic gel 0.38%, NDA 208219)

Design Element	FDA PSG Specification (Loteprednol Etabonate Ophthalmic Gel)	Regulatory Rationale
Study population	Patients undergoing indicated cataract surgery scheduled to receive ophthalmic corticosteroids pre-operatively	Provides ethical justification for destructive AH sampling; represents the indicated patient population
Dosing regimen	Single dose instilled into inferior cul-de-sac of the eye prior to cataract extraction	Single-dose design isolates formulation PK differences; minimises cumulative exposure risk
Sampling scheme	Single AH sample from one eye per patient, at one pre-assigned time point (destructive sampling)	AH sampling by paracentesis is invasive; only one sample feasible per patient per study
Sample size	Multiple cohorts, each assigned to a different sampling time point; minimum 12–15 patients per time point to construct composite PK profiles	Constructing T½, AUC, Cmax requires sufficient data per time point across cohorts
PK parameters	AUCt (AUC from zero to last sampling time), Cmax	Both rate (Cmax) and extent (AUCt) of absorption must be demonstrated equivalent
Statistical criterion	90% CI of the mean AUCt ratio Rt and Cmax within (0.80, 1.25); CI derived by bootstrap or parametric method applied to population-level mean concentrations at each time point	PSG specifies a population mean-based ratio (Rt) with 90% CI by bootstrap or parametric method — NOT the standard individual log-ANOVA ABE used for oral products; 80–125% limits apply to the mean ratio Rt, not individual GMR
Formulation	Parallel design (default) OR crossover design in patients undergoing cataract surgery in both eyes; washout period for crossover must not exceed 35 days	PSG allows crossover if cataract surgery is planned for both eyes (washout ≤35 days); parallel design is the default when surgery is in one eye only

Blinding	Double-masked (patient and assessor)	Minimises bias in AH sampling and analysis

As confirmed by Harigaya et al. (2018) from FDA's Division of Bioequivalence II, in their comprehensive review of AH PK studies submitted to FDA for ophthalmic corticosteroid suspensions, the AUC and Cmax in AH are significantly influenced by subject demographics including ethnicity and age group, while gender was not primarily associated with AH PK differences. This finding has important implications for study design: stratification or covariate control for age and ethnicity in the parallel group design is recommended to reduce bias from demographic imbalance between test and reference cohorts [8].

3.3. PK Parameters, Sample Handling, and Bioanalytical Validation

AH samples (volume approximately 50–200 µL) are collected by limbal paracentesis during the surgical procedure and immediately frozen on dry ice. Bioanalytical quantification typically employs LC-MS/MS with validated lower limits of quantification (LLOQ) in the range of 0.1–1.0 ng/mL for corticosteroids. Method validation must comply with the ICH Guideline M10 on Bioanalytical Method Validation and Study Sample Analysis (adopted 24 May 2022; EMA reference EMA/CHMP/ICH/660315/2022, effective 23 January 2023; FDA effective 7 November 2022), which supersedes both the former EMA Guideline on Bioanalytical Method Validation (EMEA/CHMP/EWP/192217/2009 Rev. 1 Corr. 2) and the FDA Guidance for Industry: Bioanalytical Method Validation (2018). ICH M10 harmonises requirements across all ICH regions and adds dedicated sections on study sample analysis, cross-validation, and endogenous analyte bioanalysis. Validation must include demonstration of matrix-specific performance in pooled human AH — a matrix presenting unique challenges due to its very limited availability for validation purposes [9].

3.4. MIDD Contribution: PBOPK Model to Support AH PK Study Design

While the AH PK study itself is mandated by the PSG, MIDD tools can provide significant value in the study design phase, specifically in determining optimal sampling time points. Since each patient contributes only one AH sample, the allocation of patients across time points directly determines the resolution of the reconstructed PK profile. Physiologically based ocular PK (PBOPK) simulation, using verified models for the drug class, can identify the time points most informative for capturing Cmax and the terminal elimination phase, thereby minimizing the total number of patients required and maximizing statistical power [2].

Additionally, PBOPK models parameterized with in vitro particle size distribution and dissolution data for both test and reference suspensions can prospectively simulate the predicted AH AUC T/R ratio, providing a feasibility assessment before committing to the expensive and ethically demanding clinical study. If simulations predict T/R ratios near the acceptance boundary, formulation adjustments can be made proactively — a high-value application of MIDD in ophthalmic generic development.

Table 4 Regulatory positioning of Model-Informed Drug Development (MIDD) within aqueous humor pharmacokinetic bioequivalence studies: permitted contributions of PBOPK modelling and population PK analysis versus required in vivo evidence

MIDD Role in AH PK Studies — Regulatory Positioning
MIDD (PBOPK simulation) does NOT replace the AH PK study required by the FDA PSG. It supports: (1) optimal sampling time point selection to maximize statistical power; (2) pre-study simulation of T/R ratios to confirm feasibility; (3) post-study PopPK model fitting to derive individual parameter estimates and support sensitivity analysis. MIDD contributions should be documented in Module 5.3.1.2 of the eCTD dossier as supplementary clinical pharmacology data.

4. Pharmacodynamic Endpoint Studies for Ophthalmic BE

4.1. When Are PD Endpoint Studies Required?

PD endpoint studies for ophthalmic BE are required by FDA PSGs when: (1) the drug product is a non-solution (suspension, emulsion, gel) that is NOT Q1/Q2 same; (2) AH PK is not feasible or is not considered sensitive enough to detect formulation differences for the specific drug class; or (3) the clinical endpoint more directly measures the pharmacodynamic effect relevant to the indication. The most prominent therapeutic category requiring IOP-based PD

endpoint studies under FDA PSGs is the anti-glaucoma class: alpha-2 adrenergic agonists (brimonidine tartrate), beta-adrenergic blockers (timolol maleate, betaxolol), carbonic anhydrase inhibitors (brinzolamide, dorzolamide), and their fixed-dose combinations [1,12].

4.2. IOP as a Pharmacodynamic BE Endpoint: Scientific and Regulatory Basis

Intraocular pressure (IOP) is both the primary modifiable risk factor in glaucoma and a validated pharmacodynamic biomarker of drug effect for anti-glaucoma medications. IOP is measured non-invasively by Goldmann applanation tonometry (GAT) in clinical practice and research settings, making it an ethically and logistically feasible PD endpoint for BE studies. The IOP-lowering response is well-characterized, dose-dependent, and reproducible within subjects across repeated measurements on the same day, although it exhibits significant diurnal variation and inter-subject variability [12].

The FDA's regulatory acceptance of IOP as a PD BE endpoint is longstanding and is explicitly operationalized in multiple PSGs. The EMA similarly accepts IOP as the primary PD endpoint for glaucoma products in its PSBGLs, consistent with its general guidance that 'quantifiable pharmacodynamic effects may be used if the PK/PD relationship is well-established and the PD endpoint is directly linked to the clinical outcome'

4.3. FDA PSG Design for Brimonidine Tartrate: A Detailed Case Study

Brimonidine tartrate ophthalmic solution 0.1% (Alphagan® P, NDA 021770, AbbVie) and 0.15% (NDA 021262) are among the most frequently cited examples of FDA PSG-mandated IOP-based PD endpoint BE studies. Two separate but structurally parallel PSGs govern these strengths; both were originally recommended in September 2008 and most recently revised in October 2025 (PSG_021770 and PSG_021262, respectively). The October 2025 revisions introduced a dual-pathway structure: Option 1 permits a biowaiver based on demonstrated Q1/Q2 sameness under 21 CFR 320.22(b)(1), while Option 2 requires a clinical endpoint BE study for products that differ from the RLD. Both revisions also formalised the BE statistical criterion as a dual standard — the 95% CI of the difference in mean IOP change from baseline must be within ±1.5 mmHg at all four timepoints and within ±1.0 mmHg for the majority (three of four) of the timepoints. Device constituent part evaluation and user interface assessment were added as required sections, consistent with FDA's evolving combination product framework for ophthalmic products. For products triggering Option 2, the FDA PSG specifies the following clinical BE study design:

Table 5 Clinical endpoint bioequivalence study design specifications for brimonidine tartrate ophthalmic solution under FDA Product-Specific Guidance (PSG_021770 / PSG_021262, October 2025): patient selection, IOP measurement timepoints, per-protocol population definition, and bioequivalence acceptance criteria

Study Parameter	PSG Specification	Notes
Study design	Double-masked, randomized, parallel, two-arm BE study	Crossover not feasible due to long washout periods and ethical concerns in glaucoma patients
Patient population	Male or non-pregnant females ≥18 years with chronic open-angle glaucoma or ocular hypertension in both eyes; baseline mean IOP ≥22 and ≤34 mmHg in each eye at two pre-treatment visits	Ensures adequate IOP at baseline to detect drug-induced reduction; bilateral disease ensures within-patient eye averaging
Dosing	One drop in both eyes three times daily at approximately 08:00, 16:00, and 22:00 for 42 days (6 weeks)	6-week treatment period reflects steady-state IOP reduction; mimics clinical practice dosing schedule
Primary endpoint	Mean IOP of both eyes at pre-defined time points: pre-dose on Day 1 (baseline), and at 2 hours post-dose on Day 1, at 2 weeks, and at 6 weeks	Multiple time points capture onset (Day 1), steady-state (2 weeks), and maintained effect (6 weeks); both eyes averaged to reduce measurement variability
Statistical method	95% confidence interval of the difference in mean IOP between test and reference must fall within −1.5 mmHg equivalence margin at each time point	Note: PD BE uses a fixed difference margin (in mmHg), not a ratio criterion; the ±1.5 mmHg margin is clinically justified as the minimum clinically important difference for IOP

Washout	Prior IOP-lowering agents discontinued with appropriate washout (minimum 28 days for prostaglandins; 14 days for beta-blockers)	Inadequate washout artificially suppresses baseline IOP, reducing sensitivity to test vs. reference differences
Rescue medication	Parasympathomimetics or carbonic anhydrase inhibitors may substitute excluded drugs during washout if medically necessary	Patient safety provision; approved substitute classes do not interfere with the study endpoints
Sample size	Not pre-specified in PSG; sponsor must power study to achieve adequate precision at each time point	Power calculation must account for standard deviation of IOP, expected mean difference, and the ±1.5 mmHg BE margin

A critical distinction from oral product BE studies must be emphasized here: the IOP-based PD endpoint BE study uses a 95% confidence interval of the mean IOP difference (not a ratio), with a fixed equivalence margin of ±1.5 mmHg. This is fundamentally different from the 90% CI of the geometric mean ratio (80–125%) criterion used for PK BE. The ±1.5 mmHg margin was established based on clinical judgment about the minimum IOP difference of clinical significance and requires specific justification in the study protocol and statistical analysis plan [11].

In May 2025, FDA issued a new Product-Specific Guidance (PSG_218424) for brimonidine tartrate ophthalmic solution/drops at the 0.025% strength (NDA 218424; LUMIFY® Preservative Free, Bausch & Lomb), an OTC product indicated for the relief of ocular redness due to minor eye irritations. This PSG is structurally distinct from the prescription IOP-lowering PSGs: it recommends a bioequivalence waiver on the basis of Q1/Q2 sameness under 21 CFR 320.22(b)(1), rather than a clinical endpoint IOP study. The 0.025% concentration is approximately four- to eight-fold lower than the IOP-lowering strengths and acts via a purely vasoconstrictive mechanism at the conjunctival level, without measurable aqueous humour pharmacokinetics. The BE pathway for the OTC product therefore relies on formulation sameness and physicochemical characterisation, with a comparative analysis of three exhibit batches for pH, specific gravity, osmolality, buffer capacity, and viscosity. In addition, FDA recommends a comparative user interface assessment for the vial-with-dropper-tip device constituent part (PSG_218424, Recommended May 2025) [11c]. The prescription PSGs for the 0.1% and 0.15% IOP-lowering products were revised in October 2025 (PSG_021770 and PSG_021262), introducing the dual-pathway biowaiver option and updated BE statistical criteria, while preserving the IOP-based PD clinical endpoint study design as the default pathway for non-Q1/Q2-same products.

4.4. Clinical Endpoint BE Studies: Nepafenac, Cyclosporine, and Beyond

When neither AH PK nor IOP provides a suitable endpoint — either because the drug's mechanism does not modulate IOP, or because the formulation difference is too complex for a PK surrogate — FDA PSGs require a full clinical endpoint BE study. Examples include:

Table 6 Drug class-specific clinical endpoint bioequivalence study frameworks under FDA Product-Specific Guidances: examples across anti-inflammatory, immunomodulatory and anti-allergic ophthalmic product categories

Drug Product	**Indication**	**FDA PSG Clinical Endpoint**	**Study Design**
Nepafenac 0.1% suspension (Nevanac®)	Post-operative inflammation & pain	Pain score (VAS 0–100) at 14 days post-cataract surgery; proportion of patients with no pain	Double-masked, parallel, placebo-controlled; patients undergoing unilateral cataract surgery; non-inferiority vs. reference
Cyclosporine 0.05% emulsion (Restasis®)	Dry eye disease	Conjunctival goblet cell density (impression cytology); Schirmer test score at 6 months	Double-masked, parallel, randomized in chronic dry eye patients; non-inferiority design; very large N (≥90/arm) required due to high variability
Olopatadine HCl 0.2% solution (Pataday®)	Allergic conjunctivitis	Ocular itching score at 3, 5, 7 minutes post-challenge in Conjunctival Allergen Challenge (CAC) model	Double-masked, crossover in allergen-sensitized subjects; CAC model provides controlled allergen exposure for reproducible PD response

Brimonidine tartrate 0.025% (Lumify®, OTC)	Ocular redness	Conjunctival redness score (validated photographic scale) at pre-defined time points post-dose	Double-masked, crossover in subjects with mild-to-moderate conjunctival redness; rapid onset allows within-session crossover

The statistical approach for clinical endpoint BE studies uses a non-inferiority margin rather than an equivalence interval, reflecting the practical impossibility of demonstrating exact equality on ordinal or continuous clinical scales with inherent measurement variability. FDA typically requires a non-inferiority margin of one unit on a validated scoring scale, justified by minimum clinically important difference data from the literature [1].

4.5. MIDD Contribution to PD Endpoint Study Design

Model-Informed Drug Development can contribute significantly to the design of IOP-based PD endpoint BE studies through exposure-response (E-R) modeling. For well-characterized glaucoma drugs with known IOP-lowering pharmacology, an Emax or indirect response PK/PD model linking AH drug concentrations to IOP reduction can be parameterized from published clinical data. This model enables:

- Sample size simulation: Modeling the expected distribution of IOP responses at each time point based on PK predictions, allowing power calculations under realistic variability assumptions
- Dosing schedule optimization: Confirming that the PSG-specified dosing times capture the peak and trough IOP effects relevant to demonstrating BE
- Sensitivity analysis: Identifying patient subgroups (e.g., by baseline IOP stratum, iris pigmentation, or concurrent medications) that drive variability in the PD endpoint, supporting stratification decisions
- Covariate identification: E-R modeling using data from prior studies can identify demographic covariates that should be balanced between treatment arms through stratified randomization.

Table 7 Regulatory positioning of Model-Informed Drug Development (MIDD) within intraocular pressure-based pharmacodynamic bioequivalence studies: permitted contributions of exposure-response modelling versus required in vivo clinical evidence

MIDD Role in PD Endpoint Studies — Regulatory Positioning
An IOP PD endpoint BE study mandated by the FDA PSG cannot be replaced by MIDD/PBOPK simulations. MIDD contributes to study design optimization (sample size, time point selection, stratification), protocol review by regulatory agencies, and interpretation of results. E-R model outputs should be included as supplementary data in Module 2.7.2 (Summary of Clinical Pharmacology) and Module 5.3.1.2 of the eCTD. For EMA Scientific Advice, E-R model-based justification of the IOP BE margin (±1.5 mmHg) may be presented to support protocol discussion.

5. Drug Class–Specific BE Study Frameworks Under FDA PSGs

5.1. Prostaglandin Analogues (Latanoprost 0.005%): Q1/Q2 Biowaiver Framework

Latanoprost 0.005% ophthalmic solution (Xalatan®, Pfizer) is the most widely prescribed prostaglandin analogue for glaucoma, and is now widely generic globally. The FDA PSG for latanoprost 0.005% solution recommends a Q1/Q2 sameness approach: if the generic product contains identical inactive ingredients at concentrations within ±5% of the RLD, a biowaiver of the in vivo BE study requirement is granted, supported by in vitro physicochemical equivalence data (pH ±0.5 units, osmolality ±20 mOsm/kg, viscosity ±20%, surface tension ±5 mN/m, and droplet size D50 ±10%) [3].

If Q1/Q2 sameness cannot be achieved, the PSG requires a full clinical BE study with an IOP endpoint (parallel group, 42 days, glaucoma patients), mirroring the brimonidine design. This creates a strong commercial and formulation incentive for generic developers to achieve Q1/Q2 sameness. MIDD (PBOPK simulation) can be used to prospectively evaluate the PK consequences of minor excipient deviations — for example, substitution of benzalkonium chloride (BAK) with an alternative preservative, or changes in viscosity-enhancing agent concentration — and may support a scientific argument to FDA that the deviation is not expected to alter ocular bioavailability, thereby informing a Type B pre-ANDA meeting discussion [10].

5.2. Corticosteroid Suspensions (Loteprednol Etabonate 0.38% Gel / 0.5% Suspension): AH PK Framework

The FDA PSG for loteprednol etabonate ophthalmic gel 0.38% (Lotemax® Gel, NDA 208219) is the most complete published example of an AH PK-based BE study design. The PSG requires: (1) Q1/Q2 sameness for the inactive ingredients; (2) comparative in vitro dissolution (particle size, dissolution in simulated tear fluid); and (3) a single-dose, in vivo PK study (parallel or crossover design, per PSG_208219) measuring loteprednol etabonate concentrations in AH in cataract surgery patients [14]. The PSG permits a crossover design when surgery is planned for both eyes, with a washout period not exceeding 35 days; a parallel design is used when only one eye undergoes surgery.

The AH PK study uses a sparse, parallel design: each patient contributes a single AH sample at one pre-assigned time point (0.25, 0.5, 0.75, 1, 1.5, 2, 3, 4, 6, and 8 hours post-dose, with approximately 12–15 patients per time point). The composite PK profiles for test and reference are then constructed from the pooled cross-sectional data, and AUCt and Cmax are estimated by non-compartmental analysis (NCA) or PopPK modeling. BE is declared when the 90% CI of the T/R geometric mean ratio falls within 80.00–125.00% for both AUCt and Cmax.

Research from FDA's own scientists (Harigaya et al., 2018) demonstrated that in AH PK studies submitted for corticosteroid suspensions, ethnicity and age significantly affected AH AUC and Cmax, while gender did not. Specifically, younger subjects and certain ethnic groups showed higher AH drug exposures, likely due to differences in corneal thickness, aqueous humor turnover rate, and nasolacrimal drainage anatomy. This evidence supports the regulatory recommendation that AH PK studies should stratify or balance subjects by age and ethnicity between test and reference arms [8].

5.3. Alpha-2 Agonists (Brimonidine Tartrate 0.1%/0.2%): IOP PD Endpoint Framework

As detailed in Section 4.3, brimonidine tartrate ophthalmic solutions require IOP-based PD endpoint clinical BE studies when Q1/Q2 sameness cannot be achieved. Two product-specific PSGs govern these products: PSG_021770 for brimonidine tartrate ophthalmic solution 0.1% (NDA 021770, revised October 2025) and PSG_021262 for the 0.15% strength (NDA 021262, revised October 2025). Both specify an identical study design for the clinical endpoint option: randomised, double-masked, parallel, 6-week treatment in open-angle glaucoma or ocular hypertension patients, with IOP measured at pre-dose (Hour 0) and 2 hours post-dose (Hour 2) on Day 14 and Day 42. The October 2025 revisions formalised the dual BE criterion: 95% CI of the difference in mean IOP change from baseline within ±1.5 mmHg at all four timepoints and within ±1.0 mmHg for the majority (three of four) of the timepoints, to be applied to the per-protocol population [11].

The parallel design is driven by the impossibility of a proper washout in glaucoma patients, for whom IOP control is medically necessary. Substituting the study drug with a non-interfering agent during washout (parasympathomimetics or CAIs, as specified by the PSG) allows for an ethical parallel design without exposing patients to sustained uncontrolled IOP elevation.

Population sizes for IOP-based PD BE studies are typically substantially larger than for AH PK studies: based on published power calculations using observed IOP standard deviations ($\sigma \approx 3–5$ mmHg in glaucoma populations) and the ±1.5 mmHg equivalence margin, sample sizes of 100–300 patients per arm are commonly required, making these studies significantly more expensive than AH PK studies [1].

5.4. Anti-Infective Suspensions (Besifloxacin 0.6%): Microbial Kill Rate and PBOPK

For topical ophthalmic anti-infective suspensions, FDA PSGs may recommend a microbial kill rate study as an alternative to clinical endpoint or AH PK studies. This in vitro approach uses standardized bacterial challenge testing to demonstrate that test and reference products achieve equivalent microbicidal activity in relevant pathogens. The Minimum Inhibitory Concentration (MIC) distribution of the drug against target organisms serves as the microbiological PD endpoint.

Besifloxacin 0.6% ophthalmic suspension (Besivance®) is a particularly interesting case study: a 2024 publication by FDA researchers (Le Merdy et al., Pharmaceutics 2024) described the application of PBOPK modeling using GastroPlus® to simulate ocular tissue exposures for this product across multiple dosage forms and formulations. The PBOPK model was parameterized with preclinical rabbit ocular PK data and then extrapolated to humans using species-specific physiological parameters, providing quantitative predictions of AH and conjunctival drug concentrations that would be prohibitively difficult to measure clinically. This represents a landmark example of MIDD being used within a regulatory context — supported by FDA's own research program — to characterize ocular drug exposure for a complex ophthalmic suspension [10].

6. Statistical Framework for Ophthalmic PK and PD BE Studies

6.1. AH PK BE: Standard ABE Criteria

For AH PK-based BE studies, the standard average bioequivalence (ABE) criterion applies: the 90% CI of the geometric mean T/R ratio for both AUCt and Cmax must fall within 80.00–125.00%. The statistical analysis follows the same general principles as for oral solid dosage forms: log-transformation of PK parameters, ANOVA with fixed effects of formulation (test, reference), time point, and potential interactions, and CI construction using the ANOVA residual mean square error (MSE) [4;15].

Unique challenges in AH PK statistical analysis include: (1) the parallel (not crossover) design eliminates the within-subject period effect cancellation that improves power in crossover studies; (2) the sparse, destructive sampling scheme requires NCA or PopPK-based AUC estimation from cross-sectional composite profiles; and (3) the very small sample volume (~100–200 μL) and high variability in AH drug concentrations due to individual differences in corneal anatomy and pre-corneal drug dynamics may yield higher residual variability than plasma PK studies. These factors collectively drive the need for larger patient cohorts per time point than naïve power calculations suggest.

6.2. IOP PD BE: Fixed Equivalence Margin with 95% CI

For IOP-based PD endpoint BE studies, the statistical framework differs fundamentally from PK BE. Rather than a ratio-based criterion, an additive equivalence margin is used: BE is declared when the 95% CI of the mean IOP difference (test minus reference) at each pre-specified time point falls entirely within the interval [−1.5, +1.5] mmHg. This 95% (not 90%) CI criterion reflects the greater clinical conservatism appropriate for a PD surrogate compared to a direct PK measure [11].

The analysis uses a linear mixed model for repeated measures (MMRM) with fixed effects of treatment (test, reference), visit (time point), treatment-by-visit interaction, and baseline IOP as a covariate, with site as a random effect for multi-center studies. Both eyes are averaged per patient at each visit to reduce measurement variability. A pre-specified hierarchical testing procedure may be used when multiple time points are included in the primary endpoint definition.

6.3. Clinical Endpoint BE: Non-Inferiority with Clinical Margin

Table 8 Statistical framework for ophthalmic bioequivalence studies: comparison of hypothesis testing approaches, confidence interval levels, acceptance criteria, and margin justifications across aqueous humor PK, IOP PD, and clinical endpoint study types

BE Study Type	Statistical Test	CI Level	Criterion	Margin Basis
AH PK (solutions/suspensions)	Log-ANOVA, parallel group	90% CI	Geometric mean T/R ratio 80.00–125.00% for AUCt and Cmax	Standard ABE criterion; same as oral solid dosage forms
IOP PD (glaucoma drugs)	MMRM, repeated measures	95% CI	Mean IOP difference (T−R) within ±1.5 mmHg at each time point	MCID = 1.5 mmHg based on clinical judgment and IOP variability data
Clinical endpoint (anti-infective, anti-inflammatory)	ANCOVA / logistic regression	95% CI	Non-inferiority: lower bound of 95% CI > −Δ (1 scale unit or pre-specified %)	MCID established from published clinical trials of reference product
Plasma PK (systemic drugs)	Log-ANOVA, crossover	90% CI	Geometric mean T/R ratio 80.00–125.00% for AUC0-t, AUC0-∞, and Cmax	Standard ABE; applicable only where plasma PK is sensitive to formulation differences

Clinical endpoint BE studies use a non-inferiority design with a pre-specified non-inferiority margin (Δ) established based on the minimum clinically important difference (MCID) for the relevant outcome scale. The FDA's general

guidance on clinical endpoint BE studies requires that the 95% CI for the treatment difference (test minus reference) at the primary endpoint time point lies entirely above −Δ, where Δ is typically one unit on a validated 0–4 or 0–4 scale (e.g., pain score, itching score) or a pre-specified percentage difference for continuous outcomes such as Schirmer test score. The reference arm must demonstrate superiority to placebo (when included) as a study assay sensitivity check.

7. Optimal Regulatory Submission Framework

7.1. eCTD Dossier Structure for Ophthalmic BE

The regulatory dossier for an ophthalmic ANDA (FDA) or generic MAA (EMA) must integrate physicochemical characterization data, in vitro test results, and in vivo PK/PD study reports in a coherent, guideline-compliant structure. The critical CTD modules for ophthalmic BE evidence are:

Table 9 eCTD submission framework for ophthalmic bioequivalence dossiers: required content by CTD module for ANDA (FDA) and generic MAA (EMA) applications, covering biowaiver justification, pharmacokinetic and pharmacodynamic study reports, and MIDD evidence

eCTD Module	Section	Required Content for Ophthalmic BE
Module 2.7.1	Summary of Biopharmaceutic Studies & Associated Analytical Methods	Q1/Q2 assessment; BE strategy rationale (waiver, AH PK, or PD endpoint); cross-reference to PSG; summary of in vitro data package
Module 2.7.2	Summary of Clinical Pharmacology Studies	PopPK or PBOPK model summary (if MIDD was used); E-R model for PD endpoint justification; AH PK parameter summary; demographics and covariates analysis
Module 5.3.1.2	Bioavailability Study Reports (in vivo)	Full AH PK study report per FDA format; IOP PD endpoint study report; plasma PK study report (if applicable); all per GCP with statistical analysis plan
Module 5.3.1.1	BE Study Reports	In vitro physicochemical comparability reports (Q1/Q2, viscosity, osmolality, pH, droplet size, surface tension, particle size distribution for suspensions)
Module 5.3.4 / 5.3.5	Bioanalytical Method Validation	LC-MS/MS method validation in AH matrix; LLOQ, linearity, accuracy, precision, stability; per ICH M10 Bioanalytical Method Validation and Study Sample Analysis (2022), which supersedes the former FDA 2018 and EMA 2011 guidance
Module 3.2.P.2	Pharmaceutical Development	Formulation development history; Q1/Q2 compliance documentation; particle size specification justification for suspensions; IVIVC data if applicable
Module 2.5	Clinical Overview	Integrated benefit-risk statement incorporating BE evidence; label claim support; MIDD outputs (if used) contextualized within overall clinical pharmacology narrative

7.2. Pre-Submission Regulatory Interactions

Given the complexity and cost of ophthalmic PK/PD BE studies, proactive regulatory interaction before study initiation is strongly recommended and, in many cases, necessary to confirm the appropriate study design:

Table 10 Recommended proactive regulatory interaction strategy for ophthalmic bioequivalence program development: meeting types, agency contacts, recommended timing, and key discussion topics from pre-ANDA through post-study consultation

Regulatory Interaction	Agency	Recommended Timing	Key Topics
Type B Pre-ANDA Meeting (Special Protocol Assessment)	FDA/OGD	At least 12 months before ANDA filing	Confirm PSG applicability; Q1/Q2 deviation justification; AH PK vs. IOP PD endpoint selection; study protocol review; MIDD evidence relevance
MIDD Paired Meeting (if MIDD is central to strategy)	FDA/CDER	18–24 months before ANDA filing	PBOPK model qualification; simulation outputs to support study design; MIDD as primary vs. supportive evidence
EMA Scientific Advice (SA)	EMA/CHMP	18–24 months before MAA	PSBGL alignment; IOP margin justification; AH PK protocol; PBOPK qualification opinion under EMA/CHMP/458101/2016
EMA Bioequivalence Working Party (BEWP) Consultation	EMA/BEWP	12–18 months before MAA	BE study design details; statistical analysis plan; acceptability of non-standard BE approaches
Pre-submission meeting (Health Canada)	Health Canada	12 months before NDS/ANDS	Alignment with FDA/EMA approaches; confirmation of Canadian PSG if available

7.3. Decision Algorithm for BE Study Approach Selection

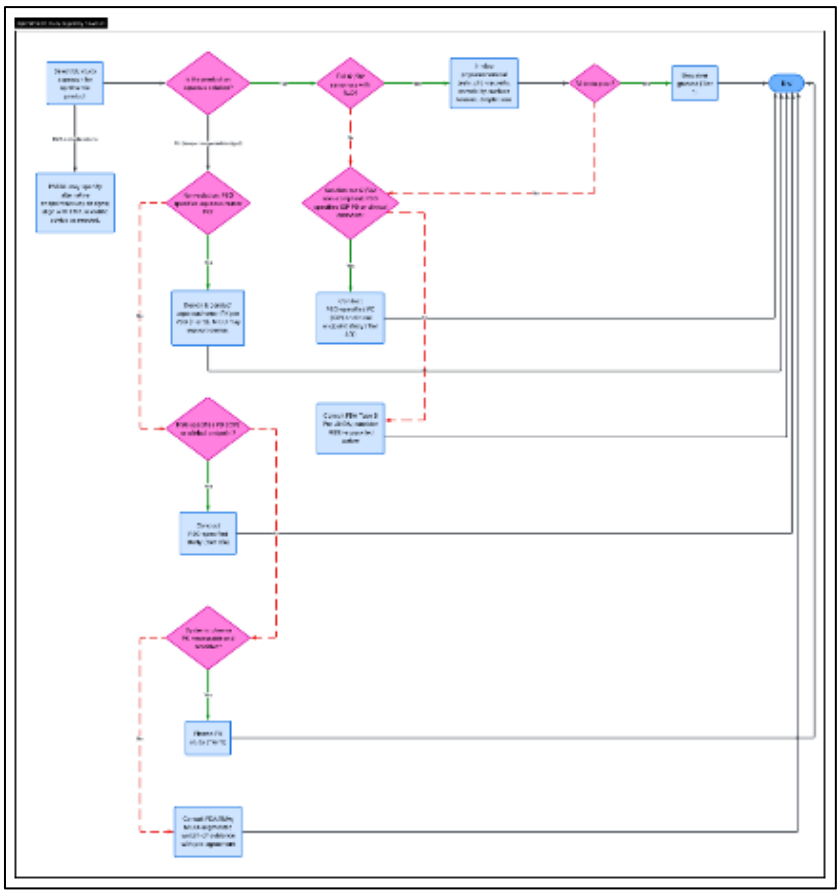

Figure 1 Decision algorithm for bioequivalence study approach selection in ophthalmic eye drop products. Adapted from FDA PSG requirements and EMA PSBGLs. MIDD = Model-Informed Drug Development; PSG = Product-Specific Guidance; RLD = Reference Listed Drug; Q1/Q2 = qualitative/quantitative sameness; AH PK = aqueous humor pharmacokinetics; IOP = intraocular pressure; T/R = test/reference

8. Remaining Challenges and Unresolved Questions

8.1. Sensitivity of AH PK Studies

A fundamental unresolved question in ophthalmic BE science is whether AH drug concentrations are sufficiently sensitive to detect formulation differences that are clinically meaningful but modest in magnitude. Published AH PK datasets for corticosteroid suspensions show very high inter-subject variability (CV frequently exceeding 50–80%), which substantially increases the sample size required to power a BE study at 80% for the standard 80–125% acceptance criterion. Harigaya et al. (2018) specifically noted that the 'relationships among AH PK, subject demographics, ocular anatomy, physiology and the compounds' physicochemical characteristics are not well understood,' a gap that PBOPK modeling is positioned to fill [8~~6~~].

8.2. Q1/Q2 Sameness as a Barrier to Generic Innovation

The Q1/Q2 sameness requirement, while scientifically motivated, has been criticized as an overly prescriptive formulation straitjacket that may prevent the development of improved generic formulations with superior tolerability (e.g., preservative-free alternatives). Regulatory evolution toward weight-of-evidence approaches — where PBOPK simulation combined with in vitro permeation data could justify minor excipient deviations — is an active area of FDA and EMA policy development. Several PSGs have already begun moving in this direction by incorporating Q3 microstructural characterization approaches [5,7].

8.3. Ethical Constraints on AH Sampling

The requirement to conduct AH PK studies exclusively in cataract surgery patients limits study feasibility and creates a potential disconnect between the study population (elderly cataract patients with altered ocular physiology) and the target patient population (e.g., glaucoma patients in their 50s with preserved ocular anatomy). PBOPK modeling can help bridge this gap by predicting AH PK in the target population from model parameters verified in the surgical population, but such extrapolations require explicit regulatory acceptance [10].

8.4. Harmonization of FDA and EMA Approaches

While FDA PSGs and EMA PSBGLs are broadly aligned in recommending AH PK or IOP PD endpoints for ophthalmic BE, differences persist in specific acceptance criteria, BE margin definitions, and the role of in vitro alternatives. For example, EMA places greater emphasis on Q3 microstructural characterization for suspensions, while FDA's PSGs tend to mandate in vivo studies more directly. International programs such as CIRS-UK and IGDRP promote harmonization, but regulatory differences remain commercially significant for generic sponsors pursuing simultaneous US-EU market access.

9. Conclusion

The demonstration of bioequivalence for ophthalmic eye drops is uniquely challenging but not scientifically intractable, provided that the regulatory framework is thoroughly understood, and the study design is precisely aligned with the applicable FDA PSG or EMA PSBGL for the reference product.

The core principle governing ophthalmic BE is that local drug exposure at the target tissue — not systemic plasma exposure — determines pharmacological equivalence. In response, regulators have built a graduated, product-specific framework that deploys aqueous humor PK studies (for corticosteroid and anti-infective suspensions), IOP-based pharmacodynamic studies (for anti-glaucoma solutions and suspensions), and clinical endpoint studies (for complex formulations such as cyclosporine emulsions) as appropriate surrogates for local ocular bioequivalence. These approaches are operationalized through product-specific guidances that prescribe study design, patient populations, endpoint definitions, and statistical criteria with increasing precision.

MIDD — particularly PBOPK modeling and E-R analysis — does not replace these regulatory-mandated studies but adds substantial value in study design optimization, formulation decision support, regulatory pre-submission discussions, and post-study data interpretation. The FDA's own research program (Harigaya et al., Le Merdy et al.) has demonstrated the utility of PBOPK modeling in characterizing ocular drug exposure and AH PK variability, establishing an important precedent for MIDD contributions to regulatory ophthalmic BE decisions.

For regulatory scientists at ophthalmic pharmaceutical companies, the optimal strategy is to (1) consult the applicable FDA PSG and EMA PSBGL at the earliest stage of generic development; (2) determine Q1/Q2 compliance early and

document deviations prospectively; (3) engage FDA via a Type B Pre-ANDA meeting or EMA via Scientific Advice before initiating any in vivo study; and (4) consider MIDD as a scientifically enabling tool to optimize and contextualize the mandated BE studies within the eCTD submission. This approach, combining rigorous adherence to regulatory frameworks with state-of-the-art quantitative pharmacology tools, represents the current best practice for ophthalmic generic drug development.

Compliance with ethical standards

Disclosure of conflict of interest

The author is an employee of Alcon Pharmaceuticals, a global leader in ophthalmic drug and medical device development. This article represents the author's independent scientific opinion and does not constitute official Alcon regulatory strategy, policy, or legal advice.

Data Availability

All regulatory citations are to publicly available FDA and EMA guidance documents, PSGs, and published peer-reviewed literature. Regulatory document URLs are provided in the reference list.

References

[1] Choi SH, Lionberger RA. Clinical, Pharmacokinetic, and In Vitro Studies to Support Bioequivalence of Ophthalmic Drug Products. AAPS J. 2016 Jul;18(4):1032-8. doi: 10.1208/s12248-016-9932-z.

[2] Le Merdy M, Tan ML, Babiskin A, Zhao L. Physiologically Based Pharmacokinetic Model to Support Ophthalmic Suspension Product Development. AAPS J. 2020 Jan 6;22(2):26. doi: 10.1208/s12248-019-0408-9.

[3] U.S. Food and Drug Administration. Product-Specific Guidance for Generic Drug Development — Latanoprost Ophthalmic Solution 0.005%. Silver Spring, MD: FDA Office of Generic Drugs; 2020.

[4] European Medicines Agency, Committee for Medicinal Products for Human Use (CHMP). Guideline on the Investigation of Bioequivalence. CPMP/EWP/QWP/1401/98 Rev. 1/Corr **. Amsterdam: EMA; 209. Available at: https://www.ema.europa.eu/en/documents/scientific-guideline/guideline-investigation-bioequivalence-rev1_en.pdf

[5] European Medicines Agency, Committee for Medicinal Products for Human Use (CHMP). Guideline on Quality and Equivalence of Locally Applied, Locally Acting Cutaneous Products. EMA/CHMP/QWP/708282/2018 Corr.1. Adopted 1 October 2024; in effect 2 April 2025. Amsterdam: EMA; 2024. Note: Primary scope is cutaneous products; stated as potentially relevant by analogy for ocular preparations, but not mandatorily applicable to eye drops. Available at: https://www.ema.europa.eu/en/documents/scientific-guideline/guideline-quality-equivalence-locally-applied-locally-acting-cutaneous-products_en.pdf

[6] European Medicines Agency, Committee for Medicinal Products for Human Use (CHMP). Draft Guideline on Quality and Equivalence of Topical Products. CHMP/QWP/708282/2018. Amsterdam: EMA; 2018. Available at: https://www.ema.europa.eu/en/documents/scientific-guideline/draft-guideline-quality-equivalence-topical-products_en.pdf

[7] Renukuntla J, Palakurthi SS, Bolla PK, et al. Advances in in-vitro bioequivalence testing methods for complex ophthalmic generic products. Int J Pharm. 2022 Nov 5:627:122209. doi: 10.1016/j.ijpharm.2022.122209.

[8] Harigaya Y, Jiang X, Zhang H, Chandaroy P, Stier EM, Pan Y. Bioequivalence Study Methods with Pharmacokinetic Endpoints for Topical Ophthalmic Corticosteroid Suspensions and Effects of Subject Demographics. Pharm Res. 2018 Nov 15;36(1):13. doi:10.1007/s11095-018-2537-8

[9] International Council for Harmonisation of Technical Requirements for Pharmaceuticals for Human Use (ICH). ICH Guideline M10 on Bioanalytical Method Validation and Study Sample Analysis. Step 4, adopted 24 May 2022. EMA reference: EMA/CHMP/ICH/660315/2022 (effective 23 January 2023); FDA effective 7 November 2022. Note: This guideline supersedes the EMA Guideline on Bioanalytical Method Validation (EMEA/CHMP/EWP/192217/2009 Rev. 1 Corr. 2) and the FDA Guidance for Industry: Bioanalytical Method Validation (2018). Available at: https://database.ich.org/sites/default/files/M10_Guideline_Step4_2022_0524.pdf

[10] Le Merdy M, Conner DP, Jiang X, et al. Clinical Ocular Exposure Extrapolation for a Complex Ophthalmic Suspension Using Physiologically Based Pharmacokinetic Modeling and Simulation. Pharmaceutics. 2024 Jul 9;16(7):914. doi: 10.3390/pharmaceutics16070914.

[11] U.S. Food and Drug Administration. Product-Specific Guidances for Brimonidine Tartrate: (a) Ophthalmic Solution 0.1%, IOP-lowering (NDA 021770). Recommended Sep 2008; Revised Feb 2014, Dec 2014, Mar 2015, Oct 2017, Oct 2025. Introduces dual-pathway structure (Q1/Q2 biowaiver or clinical endpoint study) and formalised dual BE criterion (±1.5 mmHg at all timepoints; ±1.0 mmHg for 3/4 timepoints). Available at: https://www.accessdata.fda.gov/drugsatfda_docs/psg/PSG_021770.pdf. (b) Ophthalmic Solution 0.15%, IOP-lowering (NDA 021262; PSG_021262). Recommended Sep 2008; Revised Feb 2014, Dec 2014, Mar 2015, Jul 2017, Oct 2025. Same structural revisions as (a). Available at: https://www.accessdata.fda.gov/drugsatfda_docs/psg/PSG_021262.pdf. (c) Ophthalmic Solution/Drops 0.025%, OTC ocular redness (NDA 218424; LUMIFY® Preservative Free; PSG_218424). Recommended May 2025. BE pathway: biowaiver based on Q1/Q2 sameness under 21 CFR 320.22(b)(1). Available at: https://www.accessdata.fda.gov/drugsatfda_docs/psg/PSG_218424.pdf. Silver Spring, MD: FDA

[12] Goldberg I. Relationship between intraocular pressure and preservation of visual field in glaucoma. Surv Ophthalmol. 2003 Apr;48 Suppl 1:S3-7. doi: 10.1016/s0039-6257(03)00006-7.

[13] U.S. Food and Drug Administration. MIDD Paired Meeting Program for Product Development Tools — Guidance for Industry. Rockville, MD: FDA; 2020. Available at: https://www.fda.gov/media/133563/download

[14] U.S. Food and Drug Administration. Product-Specific Guidance: Loteprednol Etabonate Ophthalmic Gel 0.38% (NDA 208219). Silver Spring, MD: FDA. Available at: https://www.accessdata.fda.gov/drugsatfda_docs/psg/PSG_208219.pdf

[15] U.S. Food and Drug Administration. Guidance for Industry: Bioequivalence Studies with Pharmacokinetic Endpoints for Drugs Submitted Under an ANDA. Rockville, MD: FDA; 2013.

[16] U.S. Food and Drug Administration. Draft Guidance for Industry: Quality Considerations for Topical Ophthalmic Drug Products. Docket FDA-2023-D-4177. Silver Spring, MD: FDA; December 2023.