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Navigating the complexity of biologics and biosimilar: structural, clinical, and regulatory insights

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ABSTRACT

Biologics are complex therapeutic agents derived from living cells, and biosimilar is their highly similar, lower-cost alternatives developed after patent expiration. These therapies have revolutionized treatment paradigms in oncology, immunology, and rare diseases by providing targeted and effective interventions. Despite their potential, challenges persist, including high production costs, complex biomanufacturing processes, regulatory inconsistencies, and concerns about environmental sustainability. Early research on biosimilar often focused narrowly on efficacy or cost-effectiveness, overlooking broader functional implications, immunogenicity risks, and sustainability of large-scale manufacturing. Limited comparative studies between originator biologics and biosimilar created significant knowledge gaps in defining critical quality attributes and establishing robust benefit–risk profiles. Furthermore, while recombinant protein-based therapeutics and monoclonal antibodies dominate the biopharmaceutical market, their development and clinical integration are hindered by multifaceted challenges, including regulatory heterogeneity, quality assurance, and long-term patient safety considerations. Early literature has also inadequately addressed process design innovations, sustainable manufacturing practices, and the complexities of clinical adoption in diverse healthcare systems. These lacunas necessitate a comprehensive review that integrates structural, functional, clinical, and regulatory perspectives, with a particular emphasis on immunogenicity and sustainability. This article aims to bridge these gaps by offering a critical appraisal of biosimilar development, adoption, and clinical integration, informing future strategies and policy frameworks.

1. INTRODUCTION

Biologics and biosimilar represent a significant leap in pharmaceutical innovation, offering targeted and efficient therapies for life-threatening and chronic diseases. Biologics are sophisticated treatments derived from living organisms, produced through advanced biotechnological processes, and extensively applied in oncology, immunology, and the treatment of rare genetic diseases. Biosimilar, which exhibits a high degree of similarity to authorized reference biologics with no clinically significant variations in effectiveness or safety, is intended to enhance affordability and access for a broader range of patient populations [1]. According to recent market analyses, biologics account for more than 40% of worldwide pharmaceutical Research and Development spending, largely due to their growing prominence in contemporary healthcare. As global demand grows, there is a pressing need to establish sustainable methods that reduce production costs, conserve resources, and provide equal access across different geographies. Although biosimilar are cost-effective, their production still requires extensive testing, specialized manufacturing facilities, and stringent regulatory requirements. Despite their potential, the production and use of biologics and biosimilar are hindered by issues such as high energy

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requirements, the application of animal-based materials, and stringent quality control measures [2]. These limitations underscore the need to leverage innovative technologies, including drug design and green bioprocessing, thereby enhancing the robustness of sustainability. This review undertakes a critical assessment of the sustainable methods currently employed in biologic and biosimilar development, identifying gaps in the traditional approach and proposing integrative solutions that incorporate technology, policy, and clinical research. Major areas of focus include structural complexity, production platforms, immunogenicity, regulatory policy, and market integration, everything viewed through the prism of sustainability.

2. METHODOLOGY

This review aimed to critically examine sustainable strategies in biologic and biosimilar development, with emphasis on manufacturing, regulatory environments, immunogenicity, and new technologies. An extensive literature search was conducted using databases such as PubMed, Scopus, ScienceDirect®, and Google Scholar, covering the period from 2015 to 2025. Keywords such as "biosimilar," "biologics," "green bioprocessing," "biopharmaceutical manufacturing," "sustainability," and "regulatory harmonization" were searched in different combinations. Articles were selected based on relevance, scientific validity, and emphasis on sustainability, excluding non-English publications and studies with methodological flaws. Moreover, regulatory guidelines were downloaded from official websites such as the United States Food and Drug Administration (U.S. FDA), European Medicines Agency (EMA), World Health Organization, and Central Drugs Standard Control Organization (CDSCO). Literature was critically reviewed, thematically arranged, and synthesized to provide a consolidated overview of existing developments and issues in the sustainable development of biologics and biosimilars.

2.1. Structural and Functional Complexity of Biologics

Biologics are therapeutic agents derived from living systems such as microbial, animal, or human cells and are characterized by large, structurally complex macromolecules, including proteins, nucleic acids, and polysaccharides. Unlike chemically synthesized small molecule drugs, which have well-defined and reproducible structures, biologics exhibit inherent heterogeneity due to their biosynthetic origin. This complexity necessitates stringent manufacturing controls to ensure product consistency and safety [3]. A critical aspect of biologics is their structural complexity, which often includes post-translational modifications (PTMs) such as glycosylation, phosphorylation, and folding factors essential for biological activity. For example, monoclonal antibodies (mAbs) possess multiple subunits, disulfide bonds, and highly specific antigen-binding sites that require exact structural configuration. Even minor variations in tertiary or quaternary structures can affect therapeutic efficacy and immunogenicity [4]. Maintaining batch-to-batch consistency remains a significant challenge. Advanced analytical techniques such as mass spectrometry, X-ray crystallography, and high-performance liquid chromatography are routinely used to assess critical quality attributes (CQAs) [5]. However, despite technological advancements, complete structural characterization remains difficult, and some degree of variability is typically accepted within defined regulatory thresholds.

2.2. Functional Implications

Biologics have highly specific mechanisms of action, achieved through receptor binding, immune modulation, or enzymatic activity. Such mechanisms frequently require the presence of fine structural detail and are thereby highly sensitive to manufacturing conditions [6]. Interactions with the immune system can result in immunogenicity, which requires preclinical and clinical testing to identify the formation of anti-drug antibodies. This combined structural and functional complexity calls for sophisticated manufacturing and regulatory management. From the point of sustainability, these operations need large amounts of energy, water, and raw material inputs. Fine-tuning these parameters through green bioprocessing, digital twins, and continuous monitoring has the potential to decrease environmental load without diminishing therapeutic integrity. Figure 1 illustrates a comparison between biologics and small-molecule drugs in molecular size, structural complexity, manufacturing process, and regulatory control. Biologics, such as mAbs, are complex, heterogeneous molecules produced in living systems, necessitating intricate purification and quality control. Small molecule pharmaceuticals are chemically produced, have well-specified structures, and relatively simple manufacturing processes. The figure highlights key differences pertinent to therapeutic design, scalability, and sustainability issues.

2.3. Recombinant Protein-based Therapeutics

Recombinant protein therapeutics have emerged as a cornerstone of biologic drug discovery, fundamentally transforming the management of chronic, infectious, and genetic disorders by enabling highly specific, mechanism-based interventions [7]. Unlike conventional small molecules, which often exert pleiotropic effects with variable selectivity, recombinant proteins are designed to replicate, enhance, or modulate endogenous bimolecular pathways, thereby conferring superior therapeutic precision [8]. Their development is enabled through genetically engineered expression systems, such as *Escherichia coli,* yeast (*Saccharomyces cerevisiae* and *Pichia pastoris*), Chinese hamster ovary (CHO) cells, and, more recently, plant-based and insect-cell platforms [9]. Each of these host systems presents distinct strengths and limitations that significantly shape therapeutic quality, cost, and scalability. *E. coli* remains the most widely used microbial expression system due to its rapid doubling time, well-characterized genetics, and cost-effectiveness; however, its lack of PTM machinery often results in mis-folded proteins or the need for refolding from inclusion bodies [10]. Yeast systems provide higher eukaryotic folding and secretion capabilities but are limited by non-human glycosylation patterns that may elicit immunogenic responses [11]. CHO cells, considered the "gold standard" for complex biologics, generate human-compatible glycosylation and high product fidelity. However, their extended culture duration and expensive media requirements make them less cost-efficient [12]. Plant-based systems have garnered attention

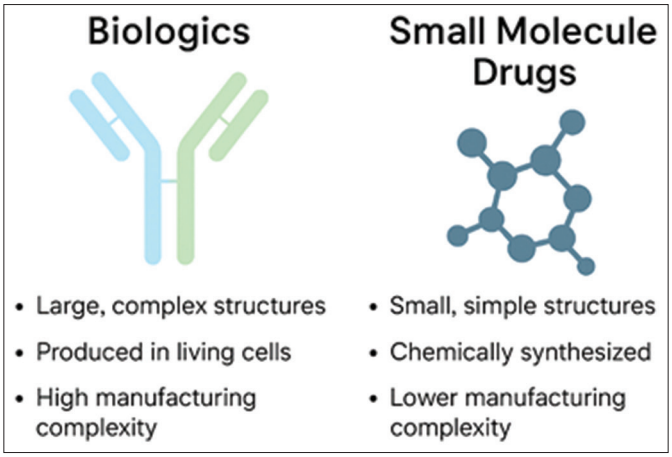

Figure 1: Comparison between biologics and small-molecule drugs.

for their scalability and biosafety profile; however, they face ongoing regulatory skepticism and batch-to-batch variability [13]. The choice of expression platform is therefore not merely technical, but strategic, influencing downstream purification complexity, regulatory approval, and ultimately patient accessibility. While microbial systems dominate in the production of relatively simple proteins such as insulin or growth hormones, mAbs and fusion proteins, which require precise PTMs, are almost exclusively produced in mammalian systems [14]. Increasingly, hybrid strategies such as glycoengineered yeast or plant lines are being investigated to combine the low cost advantages of non-mammalian hosts with the functional fidelity of mammalian cells [15]. This reflects a broader paradigm shift in recombinant protein engineering, aiming to balance clinical efficacy, economic sustainability, and global accessibility.

A landmark achievement in recombinant protein therapeutics was the approval of recombinant human insulin in 1982, which not only revolutionized diabetes care but also validated recombinant DNA technology as a viable pharmaceutical platform [16]. This breakthrough paved the way for mAbs and fusion proteins, which have since become the dominant segment of biologics, setting new standards for therapeutic specificity, clinical efficacy, and reduced off-target toxicity [17]. Despite this success and a forecasted global market exceeding USD 203 billion by 2029 [18], formidable challenges remain. Chief among these are immunogenicity risks arising from subtle glycosylation differences, the high cost and complexity of large-scale production, and sustainability concerns linked to resource-intensive upstream and downstream processes [19,20]. Moreover, regulatory heterogeneity across jurisdictions complicates biosimilar approvals, slowing equitable patient access. Thus, the field must reconcile clinical innovation with affordability, manufacturability, and global accessibility. Thus, while recombinant platforms have revolutionized biologics, their future trajectory depends on striking a balance between clinical efficacy, affordability, and sustainability. Figure 2 illustrates a flowchart of recombinant protein production, highlighting key sustainability checkpoints, including the reuse of media and energy efficient purification.

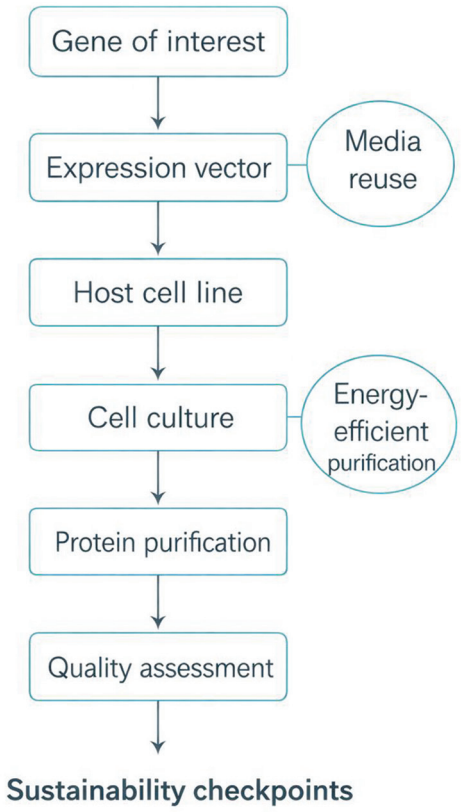

Figure 2: Pipeline of recombinant protein production with sustainability checkpoints.

2.4. Multifaceted Challenges in Biosimilar Development and Adoption

The development of biosimilar presents a unique set of scientific, regulatory, manufacturing, and market related challenges that distinguish them from traditional small-molecule generics. Biologics, due to their inherent structural complexity and sensitivity to manufacturing conditions, require a biosimilar to demonstrate a high degree of similarity, not identity, to the reference products in terms of structure, function, safety, and efficacy. This necessitates extensive analytical comparability studies using advanced techniques, such as liquid chromatography-tandem mass spectrometry (LC-MS), capillary electrophoresis, and bioassays, to assess CQAs such as glycosylation profiles, aggregation, and binding affinity. However, minor variations can still arise due to differences in cell lines, expression systems, or purification methods.

To ensure the reliability and safety of biosimilar, it is imperative to critically evaluate primary research studies that assess their comparability to reference biologics. These studies often employ advanced analytical techniques, such as LC-MS, capillary electrophoresis, and bioassays, to evaluate CQAs like glycosylation patterns, aggregation, and binding affinities. A study assessing the degradation profiles of biosimilar anti-VEGF mAbs under thermal stress conditions highlighted differences between biosimilar and their originators, which were sourced from different regions. Such findings stress the need for rigorous and standardized analytical methods to detect subtle variations that could impact therapeutic efficacy [21]. Moreover, the FDA's draft guidance on the development of therapeutic protein biosimilar emphasizes the necessity for comprehensive analytical assessments. However, critics suggest that the guidance may not fully address the complexities involved in demonstrating biosimilarity, particularly concerning the interchangeability of biosimilars with reference products.

Consequently, biosimilar manufacturers must invest heavily in process validation, scale-up optimization, and continuous quality monitoring, which significantly increase costs and development timelines [22]. Primary research studies in biologics manufacturing often present data that, while valuable, may lack the depth required for comprehensive process optimization. Studies on cell line development may report productivity metrics without addressing underlying factors such as metabolic profiles or PTMs, which are critical for ensuring consistent product quality [23]. Similarly, research on purification processes might focus on yield percentages but overlook the impact of host cell protein (HCP) co-elution or aggregate formation, both of which can affect the safety and efficacy of the final product.

Furthermore, many studies employ standard analytical techniques without considering the influence of process parameters, such as shear stress or pH fluctuations [24], which can significantly alter protein folding and bioactivity. Such oversights can lead to discrepancies between laboratory-scale findings and industrial-scale outcomes. Therefore, it is imperative to critically evaluate these studies, considering the broader context of process variability and its implications on product consistency and regulatory compliance. Integrating a more holistic approach in primary research can bridge the gap between experimental data and real-world manufacturing challenges, ultimately enhancing the reliability and quality of biologic therapeutics [25].

To strengthen the evidence base for biosimilar safety and efficacy, it is essential to critically dissect specific primary research studies that provide granular data on immunogenicity, pharmacokinetics (PKs), and therapeutic outcomes. A 2024 study comparing the immunogenicity of biosimilar natalizumab (biosim-NTZ) with its reference product (ref-NTZ) found that both induced similar anti-drug antibody (ADA) and neutralizing antibody (nAb) responses in healthy subjects and patients with relapsing-remitting multiple sclerosis [26]. This suggests comparable immunogenic profiles, which are crucial for assessing long-term safety and efficacy. PK equivalence was demonstrated in a 2023 trial of the proposed tocilizumab biosimilar, MSB11456, where the PK profiles, safety, and immune responses were similar to those of the US-licensed reference tocilizumab [27]. A 2023 study on MW031, a denosumab biosimilar, confirmed similar PKs, pharmacodynamics (PD), safety, and immunogenicity profiles to the reference product in healthy Chinese participants [28]. These studies highlight the importance of rigorous evaluation of primary research to ensure the safety, efficacy, and interchangeability of biosimilar, particularly when considering the extrapolation of indications. By carefully evaluating the design, methodology, and findings of these primary studies, researchers can draw nuanced conclusions regarding biosimilar interchangeability, indication extrapolation, and patient safety, thereby informing regulatory decisions and clinical practice with greater confidence.

Beyond technical and regulatory concerns, biosimilar also face significant market and perception challenges. Adoption may be hindered by skepticism among clinicians and patients regarding the equivalence of these biologics to originator biologics [29]. While policy and perception issues are important, the evidence base supporting biosimilar use relies heavily on primary research studies, which must be critically evaluated. Randomized controlled trials (RCTs) comparing biosimilar to their originator biologics have generally demonstrated comparable efficacy, safety, and immunogenicity profiles; however, the generalizability of these findings can be limited by factors such as sample size, patient selection, trial duration, and endpoint definitions [30]. Observational studies and real-world evidence provide complementary insights but are often subject to confounding and bias, highlighting the need for careful methodological evaluation. Comparative effectiveness studies in diverse patient populations can reveal differences in immunogenic responses or adverse events that may not be evident in controlled trial settings. Pharmacovigilance data are equally critical, but variations in reporting practices and follow-up periods can influence the interpretation of safety outcomes [31]. Therefore, a systematic and critical appraisal of both RCTs and observational studies is essential to ensure that clinical decisions regarding biosimilar adoption are informed by robust, transparent, and contextually relevant evidence. This approach will enhance confidence among clinicians and support the development of policies for biosimilar utilization. Transparent post-marketing surveillance data can reinforce trust by demonstrating long-term safety and efficacy. Moreover, strategic incentives such as reduced copayments, favorable formulary placement, and competitive pricing can further facilitate the uptake of biosimilar. A coordinated approach addressing scientific, regulatory, and perceptual obstacles is thus vital for the successful integration of biosimilar into routine clinical practice.

2.5. Integrated Biopharmaceutical Manufacturing: Processes and Sustainability

Biopharmaceutical manufacturing is a complex and highly regulated domain that encompasses the production of biologics, such as recombinant proteins, mAbs, and biosimilar. The manufacturing workflow encompasses both upstream and downstream processes, each of which is critical for ensuring product safety, efficacy, and consistency. Upstream processing encompasses cell line development, media preparation, and cell cultivation in bioreactors, typically utilizing expression systems such as CHO cells, *E. coli*, or yeast [32]. These processes are tightly regulated to maintain optimal parameters, including temperature, pH, dissolved oxygen, and nutrient concentrations, thereby maximizing protein expression while ensuring cell viability. Advances, such as continuous and perfusion-based systems, have enhanced productivity and scalability. Downstream processing, on the other hand, involves the recovery and purification of target proteins through steps like centrifugation, filtration, chromatography, and lyophilization. Maintaining protein integrity while minimizing contaminants and endotoxins is essential. Single-use technology (SUT) has gained prominence in both upstream and downstream operations, offering advantages in sterility, turnaround efficiency, and reduced cross-contamination [33-35] [Figure 3].

2.6. Process Design and Quality Assurance

The production of biosimilar, remarkably analogous although not identical replicas of permitted reference biologics, is based on the ability to reproduce key structural and functional attributes through highly controlled bioprocesses. In contrast to small-molecule generics, biosimilar cannot be precisely synthesized due to the nature of biologic expression systems. Production begins with cell line development, often utilizing mammalian systems such as CHO cells, which are conducive to human-like post-PTMs. These systems minimize water usage, eliminate the use of aggressive cleaning agents, and provide more flexibility for multiproduct facilities, but the environmental impact of disposable plastics is a cause for concern, which has driven continuous research into biodegradable products and closed-loop recycling systems [30,36-38].

Transitioning from traditional stainless steel bioreactors to SUTs in biosimilar production offers both operational and environmental advantages. Lifecycle assessment studies indicate that a 2000 L single-use process has a global warming potential (GWP) of 22.7 tons carbon dioxide (CO_2 equivalent per kilogram of drug substance, with 88% of the GWP arising during the use phase [39]. This is significantly lower than the GWP of stainless steel bioreactors, which can reach up to 50 tons CO_2 equivalent per kilogram of drug substance. In addition to reducing carbon emissions, SUTs also require less water and energy, leading to decreased operational costs and a more sustainable manufacturing process overall. The switch to SUTs not only benefits the environment but also improves efficiency and cost-effectiveness in biosimilar production. Compared with conventional stainless steel systems, SUTs can reduce GWP by ~34%, cumulative energy demand by ~32%, and energy consumption by up to 29%, primarily due to decreased water heating and steam requirements [40]. This shift toward more sustainable practices in biosimilar production is crucial in reducing the overall environmental impact of pharmaceutical manufacturing. By implementing SUTs, companies can not only meet their sustainability goals but also save money in the long run. Water consumption is also dramatically lowered by ~87%, due to the elimination of intensive cleaning processes. This significant reduction in water usage not only contributes to environmental conservation but also helps companies adhere to increasingly stringent regulations regarding water conservation and pollution. Despite these benefits, the end-of-life disposal of single-use plastics introduces environmental burdens, although this stage accounts for less than 1% of total lifecycle impacts. These quantitative data stress that while SUTs enhance sustainability and process efficiency, ongoing research into biodegradable materials and closed-loop recycling remains essential to

reduce environmental impacts in biosimilar manufacturing further. In general, the production of biosimilar is a technologically challenging but economically and therapeutically rewarding venture. It not only increases access to sophisticated biologic treatments at a lower cost but also drives innovation toward more sustainable, efficient, and high-quality bioproduction processes.

2.7. Immunogenicity Considerations in Biosimilar mAb Development

Immunogenicity refers to the ability of a biologic or biosimilar to elicit an immune response, resulting in the formation of ADAs. These immune responses can impair therapeutic efficacy, alter PKs, or result in adverse effects, making immunogenicity a critical concern in the development and regulatory evaluation of biologics and biosimilar [41]. Various factors influence the immunogenic potential, including product-related attributes such as structural complexity, glycosylation, protein aggregation, and HCP content. Manufacturing processes and storage conditions can introduce impurities that further elevate immunogenic risk. Patient-specific variables, such as genetic predisposition, immune competence, disease state, and concurrent medications, also contribute to immunogenic variability.

A critical examination of primary research studies is crucial for understanding the complexities of immunogenicity in biologic therapies. A study on the immunogenicity of biosimilar etanercept found that its safety, efficacy, and immunogenicity profiles were comparable to the reference biologic. However, the study's design and patient population may limit the generalizability of these findings to diverse clinical settings [42]. Similarly, research on ADAs in rheumatoid arthritis patients suggests that ADA formation is associated with reduced response to biologic treatments. The variability in ADA detection methods and patient characteristics necessitates cautious interpretation of these results [43]. The impact of HCPs as residual impurities in biopharmaceuticals has been linked to immunogenic responses. While HCPs are typically removed during production, their presence in trace amounts can still provoke immune reactions, highlighting the need for stringent purification processes. The processing and presentation of peptide antigens by antigen-presenting cells are influenced by the patient's HLA-human leukocyte antigen (HLA) profile, which plays a key role in triggering T-cell responses [44].

Biosimilar mAbs, in particular, represent a complex class of biologics that are increasingly utilized as cost-effective alternatives to originator products. They are employed in treating chronic and life-threatening diseases, including rheumatoid arthritis, breast cancer, and inflammatory bowel disease. Due to their structural and functional complexity, particularly regarding glycosylation and immunological properties, the development of biosimilar mAbs demands robust analytical, functional, and clinical comparability studies to ensure similarity in all CQAs [45,46].

2.8. Establishing Clinical Comparability and Benefit–risk Profile of Biosimilars

Biosimilars undergo a stringent stepwise comparability process to demonstrate their similarity to the reference biologic in terms of efficacy, safety, PKs, and immunogenicity. This approach typically begins with *in vitro* analytical characterization followed by non-clinical studies and confirmatory clinical trials. Analytical techniques, such as mass spectrometry, glycosylation profiling, and bioactivity assays, are employed to evaluate both structural and functional attributes. These findings are complemented by comparative PKs/PD analyses that assess absorption, distribution, metabolism, and elimination profiles. If the analytical and PK/PD data are robust, regulatory authorities may reduce the extent of clinical trials required [47]. *In silico* predictive modeling, including machine learning algorithms, is increasingly used to predict immunogenicity by analyzing molecular differences between the biosimilar and reference product, thereby optimizing candidate selection and trial design while reducing reliance on animal testing and associated costs.

Tools such as biosimilar registries, electronic health records, and spontaneous reporting systems support continuous monitoring, providing reassurance to both healthcare providers and patients [48,49]. By establishing clinical comparability and a favorable benefit–risk profile, biosimilar offer cost-effective alternatives that maintain clinical integrity, contributing significantly to healthcare sustainability. Primary research studies employing quantitative data are essential for rigorously assessing the safety, efficacy, and clinical comparability of biosimilars. These studies typically involve RCTs designed to demonstrate equivalence or non-inferiority to reference biologic products. For instance, the development of biosimilar often includes PK studies to establish bioequivalence, followed by clinical trials that assess PD and therapeutic efficacy. A notable example is the development of trastuzumab biosimilar, which has demonstrated comparable efficacy and safety profiles to the reference product trastuzumab [50]. Quantitative analyses in these studies typically involve statistical methods to compare treatment effects, adverse event rates, and immunogenicity profiles. Adaptive trial designs have been utilized to streamline the process of establishing both PK and efficacy equivalence, potentially reducing sample size and duration of studies [51]. Furthermore, Bayesian statistical approaches have been employed to integrate real-world data with clinical trial data, enhancing the robustness of efficacy estimates and supporting regulatory decision-making [52]. A stepwise outline of the biosimilar approval pathway is illustrated in Figure 4.

2.9. Clinical Integration of Biosimilars in Patient Care

The utilization of biosimilar in clinical practice represents a significant advancement toward individualized and sustainable therapeutic strategies [53]. Following a diagnosis, the choice of an appropriate

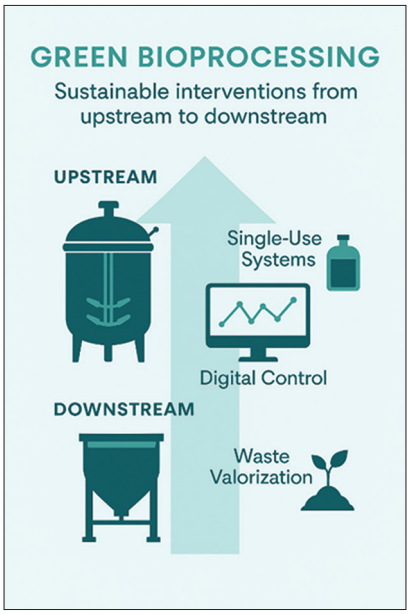

Figure 3: Green interventions in upstream and downstream bioprocessing, including single-use systems and digital twins.

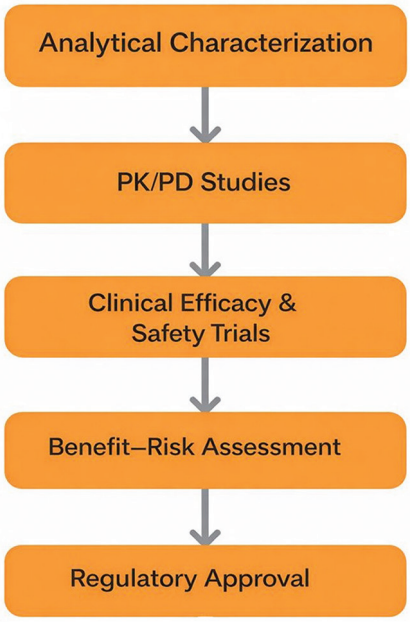

Figure 4: Biosimilar comparability assessment.

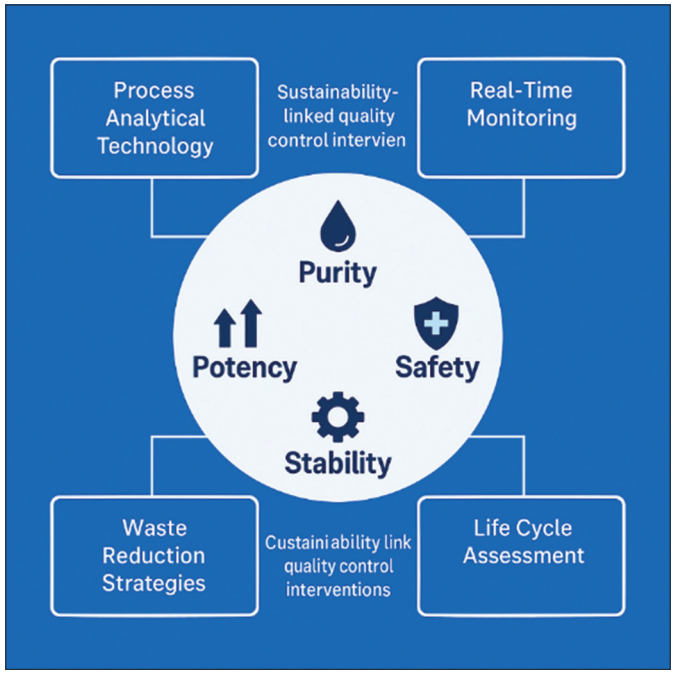

Figure 5: Target quality attributes and sustainability interventions in biosimilar development.

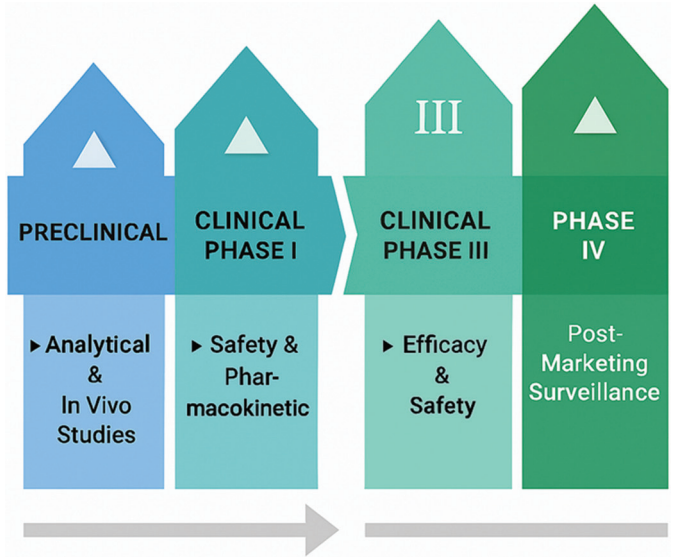

Figure 6: Sequential phases in biosimilar development.

therapeutic approach often involves biosimilars, particularly in areas such as oncology, rheumatology, and endocrinology [34,54]. Biosimilar offer similar clinical efficacy to reference biologics at lower prices, thereby increasing access to necessary therapies. Clinical decision-making is also aided by real-world evidence, physician education, and formulary inclusion policies that promote the use of biosimilars. Treatment sustainability extends beyond medication selection; it encompasses a coordinated, multidisciplinary process that incorporates ongoing patient monitoring, pharmacovigilance, and outcome measurement. With access to digital health platforms and electronic medical records, clinicians can monitor the efficacy and safety of biosimilar in real-time, allowing for responsive adjustments to care [55].

2.10. Target quality attributes (TAQs)

TQAs are the predefined physicochemical and biological attributes of a biosimilar that are crucial to ensure its safety, efficacy, and quality. They encompass characteristics such as protein structure, purity, charge variants, glycosylation profiles, and biological activity. Attainment and sustenance of TQAs during manufacturing are key to demonstrating bio similarity with the reference biologic [56]. Biosimilar must exhibit a "high degree of similarity" to the reference product; some controlled differences in quality characteristics are allowed, which provided that they are clinically irrelevant [57]. Statistical procedures are employed to analyze the operating characteristics of such similarities and determine acceptable windows of variability [58]. The pursuit of quality must fit in with sustainable production practices. Incorporating green chemistry principles into biologic manufacturing facilitates effective control of TQAs and reduces the environmental impact. It encompasses:

- Solvent reduction: Utilization of less toxic or biodegradable solvents for purification stages
- Energy efficiency: Improved temperature and pH control to minimize electricity and HVAC loads
- Material recycling: Adoption of media reuse policies and buffer recycling mechanisms [59].

Real-time monitoring using process analytical technology and predictive analytics enables perpetual process verification. This method assures consistency while maintaining eco-efficient manufacturing principles, hence balancing regulatory requirements with environmental responsibility. Figure 5 illustrates how sustainability is incorporated in quality control systems during biosimilar manufacturing.

2.11. Clinical Studies for Biosimilars

The conventional biologics/biosimilar development pipeline, spanning from preclinical studies through clinical phases to post-market surveillance, is foundational for ensuring the safety and efficacy of these products. Preclinical assessment (PK, PD, immunogenicity, and toxicity) *in vitro* and *in vivo* remains indispensable. However, mounting evidence suggests that reliance on animal models introduces significant

translational gaps, as interspecies differences in receptor expression, immune system architecture, metabolism, and glycosylation often lead to poor productivity of human outcomes (e.g., exaggerated immunogenic responses or misleading PK/PD metrics in non-human primates or rodents) [60]. Moreover, animal studies are limited by small group sizes, low sensitivity for detecting subtle functional differences between reference biologics and biosimilar, and ethical, cost, and time burdens [61]. Accordingly, regulatory frameworks are evolving, with non-clinical *in vitro* assays, humanized animal models, organ-chips, computational modeling, and advanced biomarkers being proposed to supplement or, in certain cases, replace traditional animal testing for preclinical evaluation where residual uncertainties after analytical characterization are minimal [62]. Emerging approaches, such as organ-on-chip systems, 3D bioprinting, and computational modeling, offer human-relevant alternatives; however, their regulatory acceptance and scalability remain under debate. In contemporary biosimilar development, Phases I, II, and III clinical trials remain the traditional pillars for evaluating PKs/safety, confirmatory efficacy, and post-market surveillance, respectively. Mounting evidence suggests that comparability studies utilizing high-resolution mass spectrometry, chromatographic profiling, and functional bioassays can often serve as a substitute for broad efficacy trials in many cases. A 2024 analysis found that Phase III trials for biosimilar typically cost a median of US$27.6 million and enrolled a median of ~538 patients, significantly more than analogous trials for originator biologics. Regulatory bodies (FDA, EMA, Health Canada) are now considering waivers of dedicated efficacy trials when robust analytical comparability, along with rigorous PK/PD data, shows no clinically meaningful differences.

While effective, the requirement for large Phase III trials in biosimilar has been criticized as scientifically redundant, given the precision of modern analytical and PK/PD assays. This not only prolongs timelines and increases costs but also restricts patient access to affordable therapies. While biosimilar of infliximab, trastuzumab, and adalimumab have expanded access to biologic therapies, regulatory discrepancies impede their global adoption. The EMA has approved a broader range of biosimilar for musculoskeletal diseases compared to the U.S. FDA [63]. Post-approval quality variations, such as glycan profile differences in infliximab biosimilar, underscore the challenges in achieving consistent therapeutic equivalence [64]. Future development pathways are shifting toward adaptive trial designs, the integration of real-world evidence, and digital health technologies to reduce reliance on lengthy and repetitive trials. Regulators are also considering streamlined approval models for biosimilar where robust analytical comparability may suffice, thereby lowering development costs and accelerating access. The challenge remains to balance rigorous safety evaluation with the urgent need for cost-effective biologic alternatives [Figure 6].

2.12. Regulatory Aspects

The regulatory environment for biosimilar and biologics is evolving rapidly, with a growing emphasis on harmonization, sustainability, and expedited access without compromising safety and efficacy. International regulatory agencies, including the U.S. FDA, the EMA, and India's CDSCO, have developed frameworks that delineate the criteria for biosimilar approval, comparability, and interchangeability [Table 1]. A comparative analysis of biosimilar regulatory frameworks between the FDA (United States), EMA (Europe), and CDSCO (India) is presented, based on key criteria such as approval routes, interchangeability, clinical trial requirements, and documentation standards. The table also incorporates sustainability linked incentives, indicating how agencies implement environmentally friendly practices, minimize duplication, and enhance digitalization in biosimilar development.

A closer comparative analysis of biosimilar regulatory frameworks reveals significant philosophical and operational differences among the FDA, EMA, and CDSCO, extending beyond surface-level procedural distinctions. The FDA's 351(k) pathway exemplifies a highly structured, evidence-driven approach, prioritizing the "totality-

Table 1: Comparative regulatory overview of biosimilar.

Criteria	FDA (USA)	EMA (Europe)	CDSCO (India)	Sustainability-linked incentives
Approval pathway	351(k) pathway under the Biologics Price Competition and Innovation Act	Well-established biosimilar regulatory pathway	Based on the WHO guidelines and the Indian biosimilar policy	Reduced duplication of animal/ clinical studies encouraged in all agencies
Interchangeability	Designation granted after switching studies	Not evaluated by EMA; left to national authorities	Not formally defined	FDA encourages use of RWE to minimize clinical trials
Comparability requirements	Emphasizes totality of evidence (analytical, PK/PD, clinical)	Emphasizes comparability via stepwise approach	Requires head-to-head comparisons; clinical trials usually required	Analytical and PK/PD focus reduces waste and resource use
Clinical trial requirements	Often limited to PK/PD and immunogenicity studies	Generally, fewer trials if analytical similarity is proven	Phase I and III trials often mandated	Abbreviated pathways support fewer trial participants and resources
Post-marketing surveillance	REMS and pharmacovigilance	Comprehensive pharmacovigilance required	Pharmacovigilance program mandatory	Digital reporting systems encouraged; reduce paperwork and transport
Documentation format	eCTD (electronic) submission	eCTD or NeeS	eCTD accepted; still transitioning from paper-based in some cases	Digital documentation lowers paper usage and carbon footprint
Harmonization efforts	Aligned with ICH, WHO	Actively engaged in global harmonization (ICH, WHO)	Follows WHO guidelines and regional harmonization efforts	Harmonized data avoids redundant studies across regions
Green incentives/ policies	Voluntary guidance on sustainable practices is emerging	EU green deal supports eco-conscious manufacturing	Not explicitly defined; scope for future integration	EMA leads with incentives under EU sustainability goals; others are in the early adoption phase

FDA: Food and Drug Administration, EMA: European Medicines Agency, CDSCO: Central Drugs Standard Control Organization, WHO: World Health Organization, PK: Pharmacokinetic, PD: Pharmacodynamics, REMS: Risk evaluation and mitigation strategies, EU: European Union, eCTD- (electronic) submission, ICH- International Council for Harmonisation

of-evidence" paradigm, which allows analytical, PK/PD, and limited clinical studies to substitute for extensive trials when similarity is clearly demonstrated. This facilitates accelerated market entry while maintaining stringent safety and efficacy standards. The unique interchangeability designation, reliant on controlled switching studies, reflects a proactive regulatory strategy that encourages real-world adoption and potentially reduces healthcare costs but also imposes rigorous evidence thresholds that smaller manufacturers may find challenging. EMA's framework, in contrast, emphasizes stepwise comparability with considerable regulatory discretion. By allowing reduced clinical trials when analytical similarity is robust, the EMA demonstrates a risk-based, science-driven pragmatism. Its delegation of interchangeability to national authorities introduces heterogeneity in real-world use across Europe, which may complicate multi-country commercialization strategies. CDSCO maintains a more conservative stance, generally requiring head to head clinical trials, including Phase I and III studies, reflecting a cautious, patient safety centric philosophy. While scientifically robust, this increases development time, costs, and resource consumption, potentially limiting access to biosimilar and slowing innovation relative to Western frameworks.

Sustainability and efficiency considerations further distinguish these agencies. The FDA and EMA actively promote digital documentation, harmonized data submission, and the integration of real-world evidence, thereby reducing the need for redundant trials and resource utilization. CDSCO is still transitioning from paper based systems, representing a latent opportunity for environmental and operational optimization. Post-marketing surveillance illustrates similar gradations: EMA's comprehensive pharmacovigilance contrasts with CDSCO's emerging systems, while FDA strategically leverages risk evaluation and mitigation strategies to minimize adverse outcomes without overburdening developers.

3. FUTURE SCOPE

As the worldwide biopharmaceutical industry evolves, the future development of biosimilars will be shaped by technological advancements, regulatory leniency, and sustainability requirements. Apart from accelerating developmental cycles, this approach is anticipated to minimize trial-and-error experimentation, thereby conserving valuable resources and enhancing the overall efficiency and reproducibility of research outcomes. Next-generation bioproduction systems, such as plant-based expression systems, continuous biomanufacturing, and microfluidic bioreactors, hold great promise for decreasing energy usage and operating costs while scaling up. Concurrently, synthetic biology and gene editing tools (e.g., CRISPR) may enable the design of customized cell lines with higher protein yield and consistency in quality.

4. CONCLUSION

Biosimilar represents a paradigm shift in biologic therapeutics, offering affordable alternatives without compromising safety or efficacy. Their manufacture, however, necessitates strong multi-step comparability studies, ranging from analytical to preclinical and clinical areas. With growing regulatory guidance, the incorporation of digital technologies, and sustainable manufacturing innovations such as single-use bioreactors and circular waste approaches, the biosimilar landscape is becoming increasingly efficient and environmentally friendly.

6. AUTHORS' CONTRIBUTIONS

All authors made substantial contributions to conception and design, acquisition of data, or analysis and interpretation of data; took part in drafting the article or revising it critically for important intellectual content; agreed to submit to the current journal; gave final approval of the version to be published; and agree to be accountable for all aspects of the work. All the authors are eligible to be author as per the International Committee of Medical Journal Editors (ICMJE) requirements/guidelines.

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9. ETHICAL APPROVALS

This study does not involve experiments on animals or human subjects.

10. DATA AVAILABILITY

The data supporting the findings of this study are available from the corresponding author on request.

11. PUBLISHER'S NOTE

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12. USE OF ARTIFICIAL INTELLIGENCE (AI)-ASSISTED TECHNOLOGY

The authors declare that they have not used artificial intelligence (AI) tools for writing and editing of the manuscript, and no images were manipulated using AI.

REFERENCES

1. Kang HN, Wadhwa M, Knezevic I, Ondari C, Simao M. WHO guidelines on biosimilar: Toward improved access to safe and effective products. Ann N Y Acad Sci. 2023;1521(1):96-103. https://doi.org/10.1111/nyas.14965
2. McDonnell S, Principe RF, Zamprognio MS, Whelan J. Challenges and emerging technologies in biomanufacturing of monoclonal antibodies (mAbs). In: Biotechnology - Biosensors, Biomaterials and Tissue Engineering Annual. Vol. 2022. London: IntechOpen; 2023. p. 1. https://doi.org/10.5772/intechopen.108565
3. Sahin E, Deshmukh S. Challenges and considerations in the development and manufacturing of high-concentration biologics drug products. J Pharm Innov. 2020;15(2):255-67. https://doi.org/10.1007/s12247-019-09414-3

4. Posner J, Barrington P, Brier T, Datta-Mannan A. Monoclonal antibodies: Past, present and future. Concepts Princip Pharmacol. 2019;260:81-141. https://doi.org/10.1007/164_2019_323
5. Nupur N, Joshi S, Gulliarme D, Rathore AS. Analytical similarity assessment of biosimilars: Global regulatory landscape, recent studies and major advancements in orthogonal platforms. Front Bioeng Biotechnol. 2022;10:832059. https://doi.org/10.3389/fbioe.2022.832059
6. Mallick P, Taneja G, Moorthy B, Ghose R. Regulation of drug-metabolizing enzymes in infectious and inflammatory disease: Implications for biologics-small molecule drug interactions. Expert Opin Drug Metab Toxicol. 2017;13(6):605-16. https://doi.org/10.1080/17425255.2017.1292251
7. Walsh G, Walsh E. Biopharmaceutical benchmarks 2022. Nat Biotechnol. 2022;40(12):1722-60. https://doi.org/10.1038/s41587-022-01582-x
8. Leader B, Baca QJ, Golan DE. Protein therapeutics: A summary and pharmacological classification. Nat Rev Drug Discov. 2008;7(1):21-39. https://doi.org/10.1038/nrd2399
9. Kim K, Choe D, Lee DH, Cho BK. Engineering biology to construct microbial chassis for the production of difficult-to-express proteins. Int. J. Mol. Sci 2020;21:990.
10. Rosano GL, Ceccarelli EA. Recombinant protein expression in *Escherichia coli*: Advances and challenges. Front Microbiol. 2014;5:172. https://doi.org/10.3389/fmicb.2014.00172
11. Ahmad M, Hirz M, Pichler H, Schwab H. Protein expression in *Pichia pastoris*: Recent achievements and perspectives for heterologous protein production. Appl Microbiol Biotechnol. 2014;98(12):5301-17. https://doi.org/10.1007/s00253-014-5732-5
12. Kim JY, Kim YG, Lee GM. CHO cells in biotechnology for production of recombinant proteins: Current state and further potential. Appl Microbiol Biotechnol. 2012;93(3):917-30. https://doi.org/10.1007/s00253-011-3758-5
13. Thomas DR, Penney CA, Majumder A, Walmsley AM. Evolution of plant-made pharmaceuticals. Int. J. Mol. Sci 2011;12:3220-36.
14. Walsh G. Biopharmaceuticals: Recent approvals and likely future trends. N Biotechnol. 2018;40:15-20.
15. Hamilton SR, Gerngross TU. Glycosylation engineering in yeast: The advent of fully humanized yeast. Curr Opin Biotechnol. 2007;18(5):387-92. https://doi.org/10.1016/j.copbio.2007.09.001
16. Walsh G. Biopharmaceutical benchmarks 2018. Nat Biotechnol. 2018;36(12):1136-45. https://doi.org/10.1038/nbt.4305
17. Kaplon H, Reichert JM. Antibodies to watch in 2023. mAbs. 2023;15(1):2153410. https://doi.org/10.1080/19420862.2022.2153410
18. Ansari RA, Saghir SA, Torisky R, Husain K. Recombinant protein production: From bench to biopharming. Front Drug Des Discov. 2021;10:1-31. https://doi.org/10.2174/9789811421563121100003
19. Lagassé HA, Alexaki A, Simhadri VL, Katagiri NH, Jankowski W, Sauna ZE, *et al.* Recent advances in (therapeutic protein) immunogenicity prediction and mitigation. Front Immunol. 2017;8:113. https://doi.org/10.12688/f1000research.9970.1
20. Walsh G. Biopharmaceutical benchmarks 2021. Nat Biotechnol. 2021;39(6):702-10.
21. Pamukcu C, Atik AE. Assessing the comparability of degradation profiles between biosimilar and originator anti-VEGF monoclonal antibodies under thermal stress. Pharmaceuticals (Basel). 2025;18(9):1267. https://doi.org/10.3390/ph18091267
22. Tripathi NK, Shrivastava A. Recent developments in bioprocessing of recombinant proteins: Expression hosts and process development. Front Bioeng Biotechnol. 2019;7:420. https://doi.org/10.3389/fbioe.2019.00420
23. Kannicht C, Ramström M, Kohla G, Tiemeyer M, Casademunt E, Walter O, *et al*. Characterisation of the post-translational modifications of a novel, human cell line-derived recombinant human factor VIII. Thromb Res. 2013;131(1):78-88. https://doi.org/10.1016/j.thromres.2012.09.011
24. Slavik I, Müller S, Mokosch R, Azongbilla JA, Uhl W. Impact of shear stress and pH changes on floc size and removal of dissolved organic matter (DOM). Water Res. 2012;46(19):6543-53. https://doi.org/10.1016/j.watres.2012.09.033
25. Jayakrishnan A, Wan Rosli WR, Tahir AR, Razak FS, Kee PE, Ng HS, *et al.* Evolving paradigms of recombinant protein production in the pharmaceutical industry: A rigorous review. Science. 2024;6(1):9. https://doi.org/10.3390/sci6010009
26. Chamberlain P, Hemmer B, Höfler J, Wessels H, von Richter O, Hornuss C, *et al*. Comparative immunogenicity assessment of biosimilar natalizumab to its reference medicine: A matching immunogenicity profile. Front Immunol. 2024;15:1414304. https://doi.org/10.3389/fimmu.2024.1414304
27. Tomaszewska-Kiecana M, Ullmann M, Petit-Frere C, Monnet J, Dagres C, Illes A. Pharmacokinetics of a proposed tocilizumab biosimilar (MSB11456) versus US-licensed tocilizumab: Results of a randomized, double-blind, single-intravenous dose study in healthy adults. Exp Rev Clin Immunol. 2023;19(4):439-46. https://doi.org/10.1080/1744666X.2023.2174104
28. Guo Y, Guo T, Di Y, Xu W, Hu Z, Xiao Y, *et al*. Pharmacokinetics, pharmacodynamics, safety and immunogenicity of recombinant, fully human anti-RANKL monoclonal antibody (MW031) versus denosumab in Chinese healthy subjects: A single center, randomized, double-blind, single-dose, parallel-controlled trial. Exp Opin Biol Ther. 2023;23(8):705-15.
29. Berlec A, Štrukelj B. Current state and recent advances in biopharmaceutical production in *Escherichia coli*, yeasts, and mammalian cells. J Ind Microbiol Biotechnol. 2023;40(3-4):257-74. https://doi.org/10.1080/14712598.2023.2178298
30. Joshi AK, Gupta SK. Single-use continuous manufacturing and process intensification for the production of affordable biological drugs. Process Control Intensif Digit Cont Biomanuf. 2022;14:179-208. https://doi.org/10.1002/9783527827343
31. Yang J, Yu S, Yang Z, Yan Y, Chen Y, Zeng H, *et al*. Efficacy and safety of anti-cancer biosimilar compared to reference biologics in oncology: A systematic review and meta-analysis of randomized controlled trials. Bio Drugs. 2019;33(4):357-71. https://doi.org/10.1007/s40259-019-00358-1
32. Vemula GK, Badale P, Mavrogenis P, Lalande-Luesink I, Borkowski M, Lewis DJ. A Risk-Based approach for safety case Follow-up of adverse event reports in pharmacovigilance. Adv Ther. 2024;41(1):82-91. https://doi.org/10.1007/s12325-023-02699-4
33. Wilsdon T, Pistollato M, Ross-Stewart K, Saada R, Barquin P, AlnaqbixKA, *et al*. Biosimilars: A global roadmap for policy sustainability. Charles River Associates. Washington, DC; 2022. p. 1.
34. Bas TG, Duarte V. Biosimilar in the era of artificial intelligence-international regulations and the use in oncological treatments. Pharmaceuticals (Basel). 2024;17(7):925. https://doi.org/10.3390/ph17070925
35. Ahmad AS, Olech E, McClellan JE, Kirchhoff CF. Development of biosimilar. Semin Arthritis Rheum 2016;45(5 Suppl):S11-8. https://doi.org/10.1016/j.semarthrit.2016.01.002
36. Gimpel AL, Katsikis G, Sha S, Maloney AJ, Hong MS, Nguyen TN, Wolfrum J, Springs SL, Sinskey AJ, Manalis SR, Barone PW. Analytical methods for process and product characterization of recombinant adeno-associated virus-based gene therapies. Molecular Therapy Methods & Clinical Development 2021;20:740-54.
37. Kirchhoff CF, Wang XZ, Conlon HD, Anderson S, Ryan AM, Bose A. Biosimilar: Key regulatory considerations and similarity assessment tools. Biotechnol Bioeng. 2017;114(12):2696-705. https://doi.org/10.1002/bit.26438
38. Schreitmüller T, Barton B, Zharkov A, Bakalos G. Comparative

immunogenicity assessment of biosimilar. Future Oncol. 2018;15(3):319-29. https://doi.org/10.2217/fon-2018-0553
39. Meunier S, De Bourayne M, Hamze M, Azam A, Correia E, Menier C, *et al*. Specificity of the T cell response to protein biopharmaceuticals. Front Immunol. 2020;11:1550. https://doi.org/10.3389/fimmu.2020.01550
40. Budzinski K, Constable D, D'Aquila D, Smith P, Madabhushi SR, Whiting A, *et al*. Streamlined life cycle assessment of single use technologies in biopharmaceutical manufacture. New Biotechnol. 2022;68:28-36. https://doi.org/10.1016/j.nbt.2022.01.002
41. Pietrzykowski M, Flanagan W, Pizzi V, Brown A, Sinclair A, Monge M. An environmental life cycle assessment comparison of single-use and conventional process technology for the production of monoclonal antibodies. J Clean Prod. 2013;41:150-62. https://doi.org/10.1016/j.jclepro.2012.09.048
42. Prior S, Metcalfe C, Hufton SE, Wadhwa M, Schneider CK, Burns C. Maintaining 'standards' for biosimilar monoclonal antibodies. Nat Biotechnol. 2021;39(3):276-80. https://doi.org/10.1038/s41587-021-00848-0
43. Andrade AM, da Motta Girardi J, da Silva ET, Barbosa JR, Pereira DC. Efficacy, safety, and immunogenicity of biosimilar compared with the biologic etanercept in patients with rheumatoid arthritis: A systematic review and meta-analysis. Syst Rev. 2024;13(1):291. https://doi.org/10.1186/s13643-024-02715-w
44. Bitoun S, Hässler S, Ternant D, Szely N, Gleizes A, Richez C, *et al*. Response to biologic drugs in patients with rheumatoid arthritis and antidrug antibodies. JAMA Netw Open. 2023;6(7):e2323098. https://doi.org/10.1001/jamanetworkopen.2023.23098
45. Bellinvia S, Edwards CJ. Explaining biosimilar and how reverse engineering plays a critical role in their development. Expert Opin Drug Discov. 2020;15(11):1283-9. https://doi.org/10.1080/17460441.2020.1796627
46. Gonçalves J, Araújo F, Cutolo M, Fonseca JE. Biosimilar monoclonal antibodies: Preclinical and clinical development aspects. Clin Exp Rheumatol. 2016;34(4):698-705.
47. Adami G, Saag KG, Chapurlat RD, Guañabens N, Haugeberg G, Lems WF, *et al.* Balancing benefits and risks in the era of biologics. Ther Adv Musculoskelet Dis. 2019;11:1759720X19883973. https://doi.org/10.1177/1759720X19883973
48. Gherghescu I, Delgado-Charro MB. The biosimilar landscape: An overview of regulatory approvals by the EMA and FDA. Pharmaceutics. 2020;13(1):48. https://doi.org/10.3390/pharmaceutics13010048
49. Mascarenhas-Melo F, Diaz M, Gonçalves MB, Vieira P, Bell V, Viana S, *et al*. An overview of biosimilar development, quality, regulatory issues, and management in healthcare. Pharmaceuticals (Basel). 2024;17(2):235. https://doi.org/10.3390/ph17020235
50. Halimi V, Daci A, Ancevska Netkovska K, Suturkova L, Babar ZU, Grozdanova A. Clinical and regulatory concerns of biosimilar: A review of literature. Int J Environ Res Public Health. 2020;17(16):5800. https://doi.org/10.3390/ijerph17165800
51. Alvarez DF, Wolbink G, Cronenberger C, Orazem J, Kay J. Interchangeability of biosimilars: What level of clinical evidence is needed to support the interchangeability designation in the United States? Bio Drugs. 2020;34(6):723. https://doi.org/10.1007/s40259-020-00446-7
52. Uozumi R, Hamada C. Adaptive seamless design for establishing pharmacokinetic and efficacy equivalence in developing biosimilar. Ther Innov Regul Sci. 2017;51(6):761-9. https://doi.org/10.1177/2168479017706526
53. Bujkiewicz S, Singh J, Wheaton L, Jenkins D, Martina R, Hyrich KL, *et al*. Bridging disconnected networks of first and second lines of biologic therapies in rheumatoid arthritis with registry data: Bayesian evidence synthesis with target trial emulation. J Clin Epidemiol. 2022;150:171-8. https://doi.org/10.1016/j.jclinepi.2022.06.011
54. Al Meslamani AZ. Short and long-term economic implications of biosimilar. Expert Opin Biol Ther. 2024;24(7):567-70. https://doi.org/10.1080/14712598.2024.2307353
55. Mielke J, Innerbichler F, Schiestl M, Ballarini NM, Jones B. The assessment of quality attributes for biosimilar: A statistical perspective on current practice and a proposal. AAPS J. 2019;21:7. https://doi.org/10.1208/s12248-018-0275-9
56. Dash R, Singh SK, Chirmule N, Rathore AS. Assessment of functional characterization and comparability of bio therapeutics: A review. AAPS J. 2022;24:15. https://doi.org/10.1208/s12248-021-00671-0
57. Stangler T, Schiestl M. Similarity assessment of quality attributes of biological medicines: The calculation of operating characteristics to compare different statistical approaches. AAPS Open. 2019;5(1):1-6. https://doi.org/10.1186/s41120-019-0033-9
58. Kwon O, Joung J, Park Y, Kim CW, Hong SH. Considerations of critical quality attributes in the analytical comparability assessment of biosimilar products. Biologicals. 2017;48:101-8. https://doi.org/10.1016/j.biologicals.2017.04.005
59. Vashishat A, Patel P, Das Gupta G, Das Kurmi B. Alternatives of animal models for biomedical research: A comprehensive review of modern approaches. Stem Cell Rev Rep. 2024;20(4):881-99. https://doi.org/10.1007/s12015-024-10701-x
60. Van Norman GA. Limitations of animal studies for predicting toxicity in clinical trials: Part 2: Potential alternatives to the use of animals in preclinical trials. Basic Transl Sci. 2020;5(4):387-97. https://doi.org/10.1016/j.jacbts.2020.03.010
61. Guideline IH. Preclinical safety evaluation of biotechnology-derived pharmaceuticals S6 (R1). In: Proceedings of the International Conference on Harmonisation of Technical Requirements for Registration of Pharmaceuticals for Human Use; 2011. https://database.ich.org/sites/default/files/S6_R1_Guideline_0.pdf
62. Niazi SK. Biosimilar: Harmonizing the approval guidelines. Biologics. 2022;2(3):171-95. https://doi.org/10.3390/biologics2030014
63. McGrath LN, Moodie D, Feldman SR. Biologics and biosimilar in musculoskeletal diseases: Addressing regulatory inconsistencies and clinical uncertainty. Explor Musculoskelet Dis. 2025;3:100798. https://doi.org/10.37349/emd.2025.100798
64. Shibata H, Harazono A, Kiyoshi M, Saito Y, Ishii-Watabe A. Characterization of biosimilar monoclonal antibodies and their reference products approved in Japan to reveal the quality characteristics in post approval phase. BioDrugs. 2025;39:645-67. https://doi.org/10.1007/s40259-025-00722-4

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