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DISSERTATION

“Investigating and Guiding Evidence Generation to Inform Regulatory Decision-Making for Advanced Therapy Medicinal Products (ATMPs)”

“Beurteilung und Begleitung der Generierung von Beweisen, um die regulatorische Entscheidungsfindung für Arzneimittel für neuartige Therapien (ATMPs) zu unterstützen”

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I. LIST OF ABBREVIATIONS

ATMPs	Advanced Therapy Medicinal Products
CAT	Committee for Advanced Therapies
CAR T cells	Chimeric Antigen Receptor T cells
CHMP	Committee for Medicinal Products for Human Use
CTD	Common Technical Document
CMA	Conditional Marketing Authorisation
CTMPs	Somatic Cell Therapy Medicinal Products
EPAR	European Public Assessment Report
EC	European Commission
EMA	European Medicines Agency
EU	European Union
GCP	Good Clinical Practice
GLP	Good Laboratory Practice
GMP	Good Manufacturing Practice
GTMPs	Gene Therapy Medicinal Products
HTA	Health Technology Assessment
LoOI	list of outstanding issues
LoQ	list of questions
MA	Marketing Authorisation
MAA	Marketing Authorisation Application
MCDA	Multi-criteria Decision Analysis
MoA	Mode of Action
PD	Pharmacodynamics
PK/BD	Pharmacokinetics/Biodistribution
SA	Scientific Advice
SD	Standard deviation
SME	Small and medium-sized enterprises
TEPs	Tissue-engineered products

II. PRELIMINARY REMARKS

This thesis summarises the work that has been published in:

- The Lancet Oncology (2020), 21, e104 - e116. “CAR T-cell product performance in haematological malignancies before and after marketing authorisation” **Elsallab, M.**, Levine, BL., Wayne, AS. and Abou-El-Enein, M.
  The publication contains the conducted preliminary work to evaluate the suitability of our approach in answering the research question.

- Molecular Therapy - Methods & Clinical Development (2020), 18, 269-276. “Mitigating Deficiencies in Evidence during Regulatory Assessments of Advanced Therapies: A Comparative Study with Other Biologicals” **Elsallab, M.**, Bravery, CA., Kurtz, A. and Abou-El-Enein, M.
  The publication contains the main body of work.

III. ABSTRACT (ZUSAMMENFASSUNG)

Abstract

Advanced Therapy medicinal products (ATMPs) are a novel, diverse group of pharmaceuticals that comprise cell therapies, gene therapies, and tissue-engineered products. ATMPs have provided new therapeutic approaches for several unmet medical needs. However, approved ATMPs in the EU have shown a disappointing market performance with five products being withdrawn from the market after acquiring marketing authorisations (MA). Such alarming numbers of withdrawals can indicate the presence of a gap between the evidence presented for the MA and the evidence deemed sufficient for market and patient access.

Here, we study the sufficiency of evidence in ATMPs submissions for MA. We employed two indicators of the sufficiency of evidence (i) the regulatory objections raised during the authorisation and (ii) the accepted divergence from traditional data requirements. To estimate how different the ATMPs evidence-packages compared to other closely related products, we conducted a retrospective quantitative matched-pair comparison between ATMPs and other biologicals. The comparison was carried out across four evidence domains (quality and manufacturing; experimental design and conduct of the study; efficacy and mode of action; safety and toxicity) that were created using a top-down value tree approach.

The analysis showed that approved ATMPs received significantly more objections in total ($p = 0.013$). Also, the evidence in ATMP submissions diverged more from the traditional data requirements ($p = 0.0001$). Quality issues represented the largest proportion of the objections in both groups with no significant difference between them. Furthermore, no divergence was identified in the quality sections. The experimental design and conduct of the studies, as well as efficacy requirements, had significantly more objections in approved ATMPs compared to biologicals ($p = 0.021$, 0.031, respectively). While no significant difference in the divergence was observed in the experimental design and conduct of the studies, divergences were more in the non-clinical data packages of ATMPs and affected the safety as well as efficacy domains. The comparison of time of solving the objections showed that ATMPs developers are subjected to more post-marketing commitments.

Our finding indicates that the clinical development of ATMPs suffers critical issues that affect the sufficiency of evidence at the time of the MA, particularly, the product efficacy. Furthermore, the non-clinical packages generate far less evidence compared to other biological. Finally, ATMPs' developers are subjected to more post-marketing commitments to account for such deficiencies in the evidence. This situation might increase the pressure on ATMP developers at the stage of market access. We believe that generating more evidence on the clinical efficacy before the submission might help mitigate such issues at the time of approval.

Zusammenfassung

Arzneimittel für neuartige Therapien (Advanced Therapy Medicinal Products, ATMPs) sind eine neuartige, vielseitige Gruppe von Arzneimitteln, die Zelltherapien, Gentherapien und Produkte aus dem „Tissue-Engineering“ umfasst. ATMPs haben neue therapeutische Ansätze für verschiedene Krankheiten mit hohem medizinischem Bedarf geliefert. Allerdings haben genehmigte ATMPs in Europa nach der Zulassung eine enttäuschende Marktperformance gezeigt, da fünf Produkte nach Erhalt der Marktzulassung (MZ) vom Markt genommen wurden. Solche eine alarmierende Zahl an Rückrufen können ein Hinweis auf eine Diskrepanz zwischen der für die MZ vorgelegte Evidenz und der für den Markt- und Patientenzugang als ausreichend erachteten Evidenz darstellen.

Hier untersuchen wir, ob die Evidenz in ATMPs, die für die MZ eingereicht wurden, ausreichend ist. Wir verwendeten zwei Indikatoren für die Beurteilung der Evidenz, zum einen die während der Zulassung erhobenen regulatorischen Widersprüche und zum anderen die Akzeptanz der Divergenz von den traditionellen Datenanforderungen. Um abzuschätzen, wie unterschiedlich die Evidenzdatensätze der ATMPs im Vergleich zu anderen nah verwandten Produkten sind, führten wir einen retrospektiven quantitativen „Matched-Pair“ Vergleich zwischen ATMPs und anderen biologischen Produkten durch. Der Vergleich wurde in vier Kategorien durchgeführt (Qualität und Herstellung; experimentelles Design und Durchführung der Studie; Wirksamkeit und Wirkungsweise; Sicherheit und Toxizität), die mit Hilfe eines „Top-Down-Value Tree“ Ansatzes erstellt wurden.

Die Analyse zeigte, dass zugelassene ATMPs insgesamt signifikant mehr Widersprüche erhielten ($p = 0,013$). Auch wich die Evidenz in den ATMP-Anträgen stärker von den traditionellen Evidenzanforderungen ab ($p = 0,0001$). Qualitätsprobleme machten in beiden Gruppen den größten Anteil der Einwände aus, ohne dass es zwischen ihnen einen signifikanten Unterschied gab. Auch in den Qualitätsparametern wurde keine Divergenz festgestellt. Das experimentelle Design und die Durchführung der Studien sowie die Wirksamkeitsanforderungen hatten bei zugelassenen ATMPs im Vergleich zu biologischen Produkten signifikant mehr Widersprüche ($p = 0,021$, bzw. $0,031$). Während im experimentellen Design und in der Durchführung der Studien kein signifikanter Unterschied in der Divergenz festgestellt wurde, waren die Abweichungen bei den nichtklinischen Datensätzen

der ATMPs größer und sowohl die Sicherheit als auch die Wirksamkeit betrafen. Der Vergleich der benötigten Zeit, die zur Lösung der Einwände aufgebracht werden musste, zeigte, dass die Entwickler von ATMPs mehr Verpflichtungen nach der Zulassung eingehen müssen.

Unser Ergebnis deutet darauf hin, dass die klinische Entwicklung von ATMPs unter kritischen Problemen leidet, die sich auf die ausreichende Evidenz zum Zeitpunkt der MZ auswirken, insbesondere auf die Produktwirksamkeit. Darüber hinaus erzeugen die generierten nichtklinischen Datensätze im Vergleich zu anderen biologischen Produkten weit weniger Evidenz. Schließlich sind die Entwickler von ATMPs nach der Zulassung mehr Verpflichtungen unterworfen, um solchen Mängeln in der Evidenz Rechnung zu tragen. Diese Situation könnte den Druck auf die Entwickler von ATMPs in der Phase des Marktzugangs erhöhen. Wir glauben, dass die Generierung von mehr Beweisen für die klinische Wirksamkeit vor der Beantragung der Zulassung dazu beitragen könnte, solche Probleme zum Zeitpunkt der Zulassung zu mildern.

1. SYNOPSIS

1.1. Introduction

1.1.1. The emergence of advanced therapy medicinal products (ATMPs) as a new class of medicine

The successful development of methods for the production of antibodies by immortalised cell lines in 1975 marked the beginning of a new era in medicine (1,2). Since then, the pharmaceutical industry, once dominated by small molecule development, invested heavily in the development of biological medicinal products. Currently, the development portfolio of the top 12 pharmaceutical companies comprises around 50% biologicals and specialty products (e.g. oncology and autoimmune pharmaceuticals) (3,4). In the past two decades, we are witnessing a new shift toward two new concepts in therapeutic development; precision medicine, and advanced therapies (2,5). Both concepts rely on the knowledge gained from high-throughput omics technologies, including genomics, transcriptomics, proteomics, and metabolomics, to design new diagnostic and therapeutic modalities (6). Such accumulating knowledge, coupled with the advances in biomedical engineering enabled the correction of defective genes, replacing damaged tissues, and isolation of specific cellular populations with therapeutic potential. By the beginning of the 21st century, the first wave of Advanced Therapy Medicinal Products (ATMPs) has reached the stage of marketing in Europe (7). However, due to their novelty, such products challenge our current traditional models of drug development (8). The challenges are embedded in all steps of development, spanning non-clinical and clinical development, manufacturing, marketing approval, to market access, and routine clinical practice integration. Despite their potential, ATMPs experienced low approval rates and high post-marketing withdrawals compared to other classes of medicinal products. These issues are believed to be influenced dramatically by the sufficiency of evidence generated during product development and submitted to the regulatory authorities to obtain marketing approval. As such, in this project, we focus on investigating the sufficiency of the evidence in the ATMPs regulatory submissions while benchmarking this against other established class of products such as biologicals. In the following sections, an overview on ATMPs, their definitions, and regulatory framework in the EU is provided. Then we present current issues with the approvals, prior studies, knowledge gap, and the aim of the project. Finally, we summarise the methods, results, and discussions of the published work.

1.1.2. What is an Advanced Therapy Medicinal Product?

To achieve adequate regulatory oversight for advanced therapies, regulations concerning medicine for human use had to evolve. This change started by introducing legal definitions for gene therapy medicinal products (GTMPs) and somatic cell therapy medicinal products (CTMPs) to Directive 2001/83/EC. Afterwards, Tissue-engineered products (TEPs), and Combined ATMPs were defined in regulation 1394/2007. Finally, all these products were included under the Advanced Therapy Medicinal Products (ATMPs) in Directive 2001/83/EC.

a) Gene Therapy medicinal products (GTMPs)

According to Directive 2001/83/EC, “Gene therapy medicinal product means a biological medicinal product which has the following characteristics: (a) it contains an active substance which contains or consists of a recombinant nucleic acid used in or administered to human beings to regulate, repair, replacing, adding or deleting a genetic sequence; (b) its therapeutic, prophylactic or diagnostic effect relates directly to the recombinant nucleic acid sequence it contains, or to the product of gene expression of this sequence. Gene therapy medicinal products shall not include vaccines against infectious diseases.”

b) Somatic cell therapy medicinal product (CTMPs)

The directive also defines CTMPs as following; “Somatic cell therapy medicinal product means a biological medicinal product which has the following characteristics: (a) contains or consists of cells or tissues that have been subject to substantial manipulation so that biological characteristics, physiological functions or structural properties relevant for the intended clinical use have been altered, or of cells or tissues that are not intended to be used for the same essential function(s) in the recipient and the donor; (b) is presented as having properties for, or is used in or administered to human beings to treat, prevent or diagnosing disease through the pharmacological, immunological or metabolic action of its cells or tissues.” Annex I to Regulation (EC) No 1394/2007 contains a list of techniques that shall not be considered as substantial manipulation:

- cutting,
- grinding,
- shaping,
- centrifugation,

- soaking in antibiotic or antimicrobial solutions,
- sterilisation,
- irradiation,
- cell separation, concentration or purification,
- filtering,
- lyophilisation,
- freezing,
- cryopreservation,
- vitrification.

Based on this definition, when the cells are cultured even to be used for the same function they physiologically perform, they are considered substantially manipulated and regulated as medicinal products. Such considerations are important because, based on the definition, many cellular therapies offered in unproven stem cell clinics are considered illegal as they are substantially manipulated cells and need to be thoroughly evaluated and approved by the European Medicine Agency (EMA) in the EU for human use (9).

c) Tissue Engineered Products (TEPs)

“Tissue-engineered product means a product that: – contains or consists of engineered cells or tissues, and – is presented as having properties for, or is used in or administered to human beings to regenerate, repair or replacing a human tissue. A tissue-engineered product may contain cells or tissues of human or animal origin or both. The cells or tissues may be viable or non-viable. It may also contain additional substances, such as cellular products, bio-molecules, biomaterials, chemical substances, scaffolds or matrices. Products containing or consisting exclusively of non-viable human or animal cells and/or tissues, which do not contain any viable cells or tissues and which do not act principally by pharmacological, immunological or metabolic action, shall be excluded from this definition.” “Cells or tissues shall be considered ‘engineered’ if they fulfil at least one of the following conditions: – the cells or tissues have been subject to substantial manipulation, so that biological characteristics, physiological functions, or structural properties relevant for the intended regeneration, repair or replacement are achieved. – the cells or tissues are not intended to be used for the same essential function or functions in the recipient as in the donor.”

Based on the definition of tissue-engineered products, overlap with CTMP can be observed. However, the main difference is the function the medicinal product needs to perform. In the case of TEP, the product is administered to regenerate, repair or replacing a human tissue. In contrast, in CTMP the product is administered to treat, prevent or diagnose disease through the pharmacological, immunological or metabolic action of its cells or tissues.

d) Combined Advanced Therapy Medicinal Products

The last category of ATMPs is the combined ATMPs. Regulation 1394/2007 defines combined ATMPs as the products which "fulfils the following conditions: – it must incorporate, as an integral part of the product, one or more medical devices or one or more active implantable medical devices, and – its cellular or tissue part must contain viable cells or tissues, or – its cellular or tissue part containing non-viable cells or tissues must be liable to act upon the human body with action that can be considered as primary to that of the devices referred to." As in the previous cases, the regulation also considers that "where a product contains viable cells or tissues, the pharmacological, immunological or metabolic action of those cells or tissues shall be considered as the principal mode of action of the product."

1.1.3. ATMPs approval process in the EU

ATMPs are regulated in the EU as biological medicinal products (known as biologicals). According to Directive 2001/83/EC; “A biological medicinal product is a product, the active substance of which is a biological substance. A biological substance is a substance that is produced by or extracted from a biological source, and that needs for its characterisation and the determination of its quality a combination of physicochemical-biological testing, together with the production process and its control. The following shall be considered as biological medicinal products: immunological medicinal products and medicinal products derived from human blood and human plasma as defined, respectively in paragraphs (4) and (10) of Article 1; medicinal products falling within the scope of Part A of the Annex to Regulation (EC) No 2309/93; advanced therapy medicinal products as defined in Part IV of this Annex.” Accordingly, ATMPs are subjected to the same regulatory requirements for marketing approvals and post-marketing monitoring as other biologicals.

In the EU, a centralised marketing authorisation (MA) procedure via the EMA is compulsory for ATMPs. For submissions of a marketing authorisation application (MAA), a unified format across Europe, USA, and Japan, known as the Common Technical Document (CTD), is used (10). The CTD contains scientific evidence on product quality, efficacy, and safety. The data requirements that need to be included in the CTD are listed in Annex I of the Directive 2001/83/EC. The data is the cumulative scientific evidence collected from the non-clinical and clinical studies as well as manufacturing and quality. To clarify how ATMPs are expected to fulfil the requirements and to specify additional requirements, a specific part for ATMPs was added in Annex I of Directive 2001/83/EC. The regulations also introduced the concept of regulatory flexibility where the applicant can be exempt from some of the data requirements if a risk-based approach was used or specific guidance that exempts ATMP developer from such data requirements is available.

Upon application submission, the EMA’s Committee for Advanced Therapies (CAT) performs scientific assessments of the MAA of ATMPs. The CAT reviews the application and provide the applicant with a list of questions (LoQ) that contain the objections and concerns on the provided evidence. The applicant then responds to these questions within a period between three to six months. After that, the application is reviewed again, and a list of outstanding issues (LoOI) is formulated and sent to the

applicant. The applicant has then one to three months to respond to the outstanding issue. After that, the CAT formulates an opinion. Based on the CAT draft opinion, the EMA's Committee for Medicinal Products for Human Use (CHMP) adopts a final opinion on whether a MA should be granted or not. Finally, the European Commission (EC) (the authorising body for centrally authorised products) makes a legally binding decision on the MA based on the EMA recommendations (11,12).

1.1.4. Regulatory tools to support ATMPs development

Regulators have been offering different tools to increase engagement with developers, guide development, and accelerate the evaluation of innovative therapies, including ATMPs. In 1996, the EMA introduced Scientific Advice (SA) to foster communication between the developers of new therapeutics and regulators (13). Such a tool offers applicants regulatory advice on the required scientific evidence and the appropriate studies sufficient to favour a positive benefit-risk balance. Further support is given to developers of treatments for rare diseases (fewer than 5 in 10,000 people across the EU) under orphan incentives. Products designated as orphan medicinal products benefit from a specific type of SA known as protocol assistance, which is free-of-charge. Additionally, developers are offered ten-year market exclusivity and a reduction in fees at submissions (14). Another approach to support innovative therapies that fulfil unmet medical needs is the introduction of expedited authorisation schemes (14). For instance, conditional and exceptional circumstances authorisations allow for the marketing of pharmaceutical products with limited safety and efficacy evidence. This flexibility is offered if the product addresses a serious or a rare condition with unavailable standard-of-care, and clear limitations hamper further evidence generation on product safety and efficacy (14).

Within the same context, the EMA also supports small and medium-sized enterprises (SMEs) by reducing fees for SA and MAA (15). The PRIority Medicines (PRIME) is the EMA's latest scheme that offers multiple early access tools under one initiative. The PRIME scheme started in March 2016 to accelerate the regulatory evaluation of innovative therapies that target unmet medical needs. Applicants granted a PRIME status could benefit from regulatory support through an EMA appointed rapporteur, who provides the applicant guidance on the development plan and regulatory strategy for filling the MAA. Developers also benefit from early scientific advice with reduced fees as well as the eligibility for accelerated assessment, which reduces the evaluation timeframe from 210 days to 150 days (14,16).

1.1.5. Current issues with ATMPs marketing approval

a) ATMPs has lower marketing approval rates

As of August 2020, there are 29 ATMPs' submissions for MA to the EMA, out of which six submissions are under evaluation. Gene therapy submissions increased over the years, with the submissions in the last four years being only for gene therapies (Figure 1). On the other hand, the submissions for cell therapy and tissue-engineered products were markedly less. Out of the 23 submissions with a final opinion, 15 (65%) has been approved for marketing in Europe (**Figure *2***). This approval rate is lower than the general approval rates of all medicinal products submissions to the EMA (76%) (7).

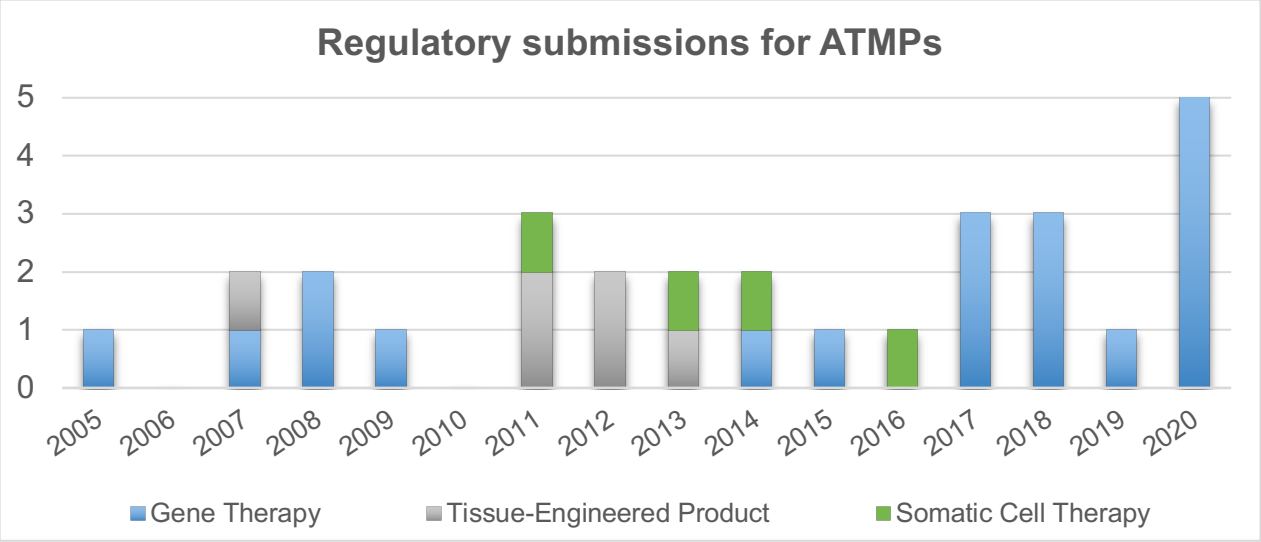

Figure 1 *Type of ATMPs regulatory submissions over the years*

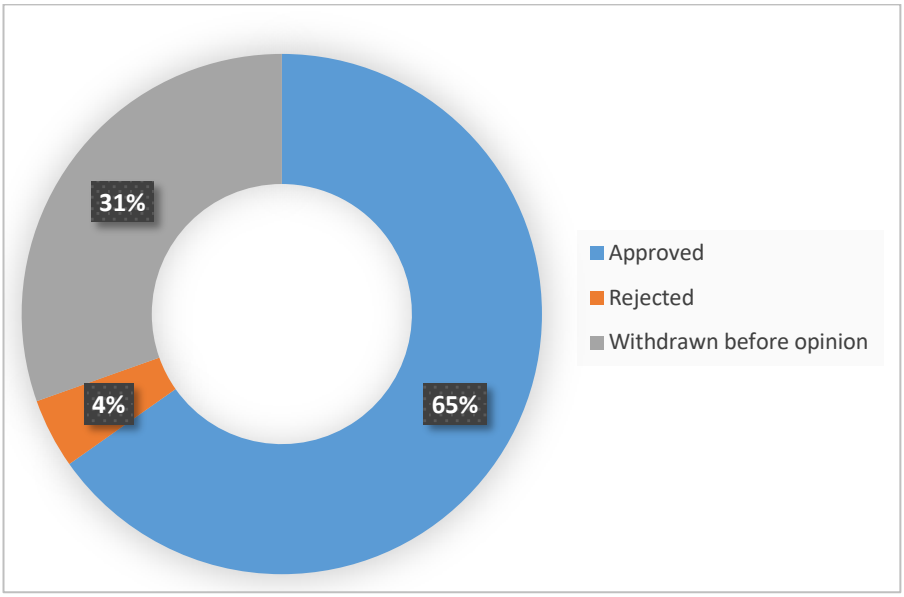

Figure 2 *Outcome of ATMPs regulatory submissions*

b) ATMPs has a high rate of post-marketing withdrawals

Five out of the 15 approved ATMPs have been withdrawn from the market (33%). The withdrawn products were approved for an average of 3.60 years (SD, 2.30; range, 1.40–6.82). Accordingly, only three marketed ATMPs are approved for more than three years, with Holoclar being the longest at 4.37 years). Comparing those numbers to the medicinal products withdrawal rates post-marketing (**Figure *3***), ATMPs show more withdrawals. The latest withdrawal was in October 2019 for Zalmoxis due to un-favourable results of the post-marketing phase III clinical trial. Other ATMPs were withdrawn due to reimbursement, manufacturing, and limited market demand (17–19).

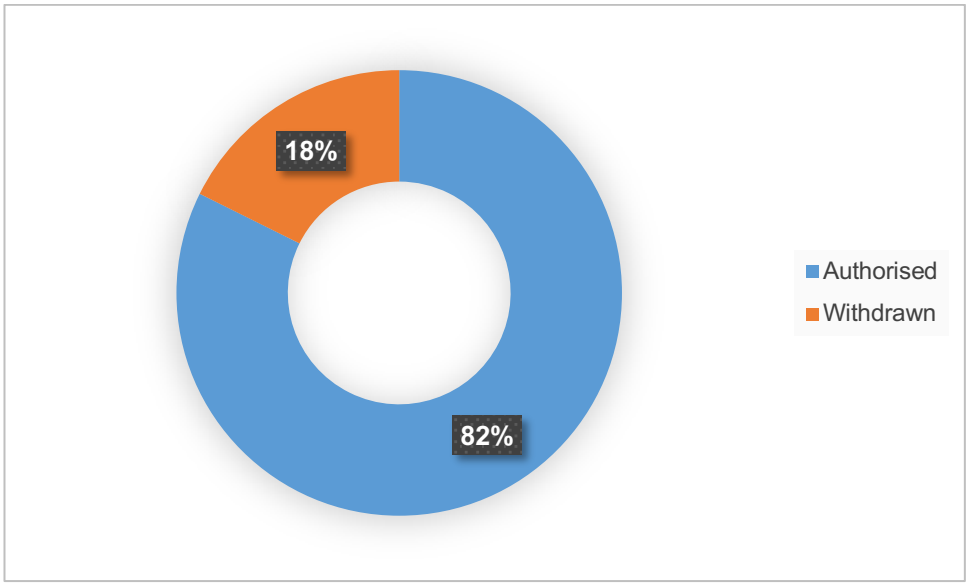

Figure 3 *General Post-marketing withdrawal rates.*
The analysis is based on the data obtained from the EMA website
https://www.ema.europa.eu/en/medicines/download-medicine-data (cut-off date: 12.04.2019)

1.2. Problem Statement & Hypothesis

The reasons for the below-average approval rates, as well as the high post-marketing withdrawals, need to be investigated. One central aspect that contributes to any medicinal product success is the amount, quality, and sufficiency of the evidence generated in the pre-marketing stage. Furthermore, the alignment of the evidence with post-marketing requirements (e.g., HTA evidence requirements) can play a crucial rule in product success (19–21). Hence, we hypothesised that the observed differences in the approval rates and post-marketing performances stem from differences in the evidence generated during product development. Understanding and characterising the evidence in ATMPs regulatory submissions can provide insights, unravel issues in the development and explain the struggling market performance. Therefore, we set the goal of investigating the sufficiency of the evidence in the ATMP submissions during the regulatory approval process. We compare the regulatory evaluation of the evidence between ATMPs and other biological submissions (e.g., monoclonal antibodies) to benchmark ATMP evidence packages against such more established class of products.

1.3. Previous Studies and Knowledge Gap

The only available source for studying the regulatory submissions of medicinal products in Europe is public regulatory documents. The European public assessment reports (EPARs) are public documents released on the EMA website after finishing the MAA revision. The EPARs contain information on the submitted quality, non-clinical and clinical data as well as the major objection and concerns raised during the evaluation of the medicinal product. The EPARs has been utilised, by others, to investigate the regulatory evaluation and to quantify and classify the regulatory objections for various kind of products and from different types of developers (22–26). For ATMPs, Wilde et al. investigated 14 submissions and identified the major objections in them. However, the issues were minimalistic in terms of the details provided and analysis performed (27). Carvalho M. et Al. studied seven gene therapy submission and provided a comprehensive description of their objections (28). Members of the EMA released the summary of the objections raised against the first 20 MAA of ATMPs (29). Bravery et al. performed secondary analyses on this data in another study (7,29). However, two main gaps can be identified in the conducted studies. First, all the studies did not include a well-established comparator to benchmark the ATMP submission against it. Having a comparator will identify specific

weaknesses in the submissions that are unique to ATMPs. Second, no study investigated the impact of regulatory flexibility on the submitted evidence and how the developer diverge from the traditional data requirements.

1.4. Aim of Project

In this project, we aimed at investigating the sufficiency of the evidence in the regulatory submissions of ATMPs in Europe. First, to evaluate the suitability of our approach in answering the research question, we conducted a pilot study by analysing the regulatory submissions of two recently approved CAR T cell products (**Publication 1**). The two products were approved for a similar indication, which created an unprecedented opportunity to assess the differences between their regulatory evaluations. First, the submitted evidence was reviewed from the EPARs of both products, and the regulatory objections against the submitted evidence were extracted and analysed. The largest part of the regulatory objections was observed in the clinical part of the evidence, particularly in the quality of the conducted studies, the historical comparators' suitability, and the products' efficacy outcomes (**Publication 1**). Our analysis revealed that the regulators accepted suboptimal non-clinical studies and showed a high degree of flexibility with the products. This finding prompted us to move forward with studying the evidence sufficiency in the entire ATMP cohort and compare it against biologicals (**Publication 2**).

For the comparison, we utilised two indicators of the evidence sufficiency; the regulatory objections and the divergence from the regulatory requirement. A retrospective quantitative matched-pair comparison of these indicators between ATMPs and other biologicals was then performed. The comparison was carried out across four evidence domains (quality and manufacturing; experimental design and conduct of the study; efficacy and mode of action; safety and toxicity). The evidence domains were created using a top-down value tree approach. We were able to identify unique weaknesses in the ATMPs submissions that differs from other biologicals. Finally, we investigated the causes of the observed variabilities, particularly in the domains with significant observable differences.

1.5. Material and Methods

1.5.1. Defining the indicators of evidence sufficiency

a) Objections (Publication 1 & 2)

During the evaluation of an MA application, the applicant receives a list of identified issues in the applications under two categories: first is “major objections,” defined as critical issues that preclude a recommendation for MA (30); second is “other concerns,” defined as issues that do not preclude a recommendation of the MA, as it can be solved through modifying the summary of product characteristics, or implementation of risk minimisation measures (30). However, in case of failure to solve the other concerns, the product cannot be authorised. Since EPARs do not differentiate between major objections and other concerns, all issues extracted from the EPAR are referred to in our work as objections.

b) Divergence (Publication 2)

Any studies stated as a requirement for the MAA in Annex I of Directive 2001/83/EC and that have not been performed by the applicant should be justified. Justifications include the availability of specific guidelines that deem these studies unnecessary for this kind of therapy, through a rational justification from the applicant or by the application of a risk-based approach. We quantified the degree of divergence by collecting the number of studies that were omitted in the submission and accepted during the evaluation of the application.

1.5.2. Retrieving and clustering Data Requirements into Evidence Domains (Publication 2)

The data requirements for MA application were retrieved from Annex I of Directive 2001/83/EC of the European Parliament and the Council (31). While the data are traditionally categorised according to their source (manufacturing, non-clinical, and clinical data), such categorisation does not reflect the role of the data in the decision-making process. Therefore, we clustered the data requirements based on the objectives of scientific evaluation and decision-making. This was achieved by employing a top-down value tree approach that is utilised in multi-criteria decision analysis (MCDA), commonly used in HTA studies (Figure 1) (Table S3 in **publication 2**). (32–34)

1.5.3. Search Strategy and data retrieval (Publication 1 & 2)

The EMA website (https://www.ema.europa.eu/en/medicines/download-medicine-data) was accessed on July 01st, 2019. We constructed a database that comprises two separate spreadsheets: one for all of the products with an EPAR (authorised and refused), and the other for withdrawn products with a withdrawal assessment report. The corresponding administrative information about each identified product was then collected from the EMA website (https://www.ema.europa.eu/en/medicines). Information on product developer and their status (size of the company) was collected by searching the company name in the SME register database (https://fmapps.emea.europa.eu/SME/). In case the company was not registered, the EMA definition of SME was used to categorise the company based on the financial and organisational status at the year of submission.

1.5.4. Selecting confounders for matching (Publication 2)

Several confounding factors can influence the amount of evidence in the regulatory submissions of pharmaceutical products. To minimise the impact of such factors on the comparative analysis, matching-method was employed. The identified factors that were selected for matching are as follows:

1- The outcome of the submission (authorised, refused, or withdrawn)
2- The disease indication as they affect the choice of the animal models and the design of the clinical trials (e.g., in case of oncology treatments)(35)
3- The rarity of indication which can complicate the clinical trial design, and patient recruitment (36)
4- Type of approval (full, Conditional, or under exceptional circumstances)
5- Time of approval as evidence requirements for MA evolve

1.5.5. Matching methods (Publication 2)

Initially, an exact match on submission outcome, orphan designations, and the type of approval were achieved. Exact matching on the disease area was done by searching the potential biologicals matches. A nearest-neighbour matching without replacement was then utilised to match the decision date of the submission (37).

1.5.6. Data Extraction and Statistical Analysis (Publication 1 & 2)

Objections and divergences were collected, sorted, and coded. The supervisor of the study then verified the data. Upon disagreement, discussions were conducted to reach a decision. IBM SPSS Statistics for Windows, (Version 25.0. Armonk, NY: IBM Corp.) was utilised for statistical analysis. Data were described in Means, ranges, and standard deviations. A non-parametric Wilcoxon signed-rank test was predefined for the analysis. Such choice was due to the small sample size and the matched design. No adjustment for multiple comparisons was made as the study is exploratory. Two-tailed p values less than 0.05 were considered statistically significant. Figures were produced by SPSS version 25 and R studio (version 1.2.1335) (38) using the tidyverse package (version 1.3.0) (39).

1.6. Results

1.6.1. Clustering the data requirements and creating the evidence domains

Using a top-down value tree approach, the data requirements listed in Annex I of Directive 2001/83/EC were clustered into four main domains: manufacturing and quality testing, experimental design and conduct of studies, efficacy and mode of action (MoA), and safety and toxicity (Figure 4). The first two domains represent “confidence criteria”, and the other two domains are considered “outcome criteria”. The confidence criteria reflect the regulator assessment of the manufacturing process and its ability to produce a consistent product without introducing additional risks to the product (e.g., impurities, contaminations, formulation). The other aspect of the confidence criteria concerns the integrity of the performed studies and the reliability of such studies in estimating product safety and efficacy. Having issues in the confidence criteria will affect the reliability of the outcome criteria.

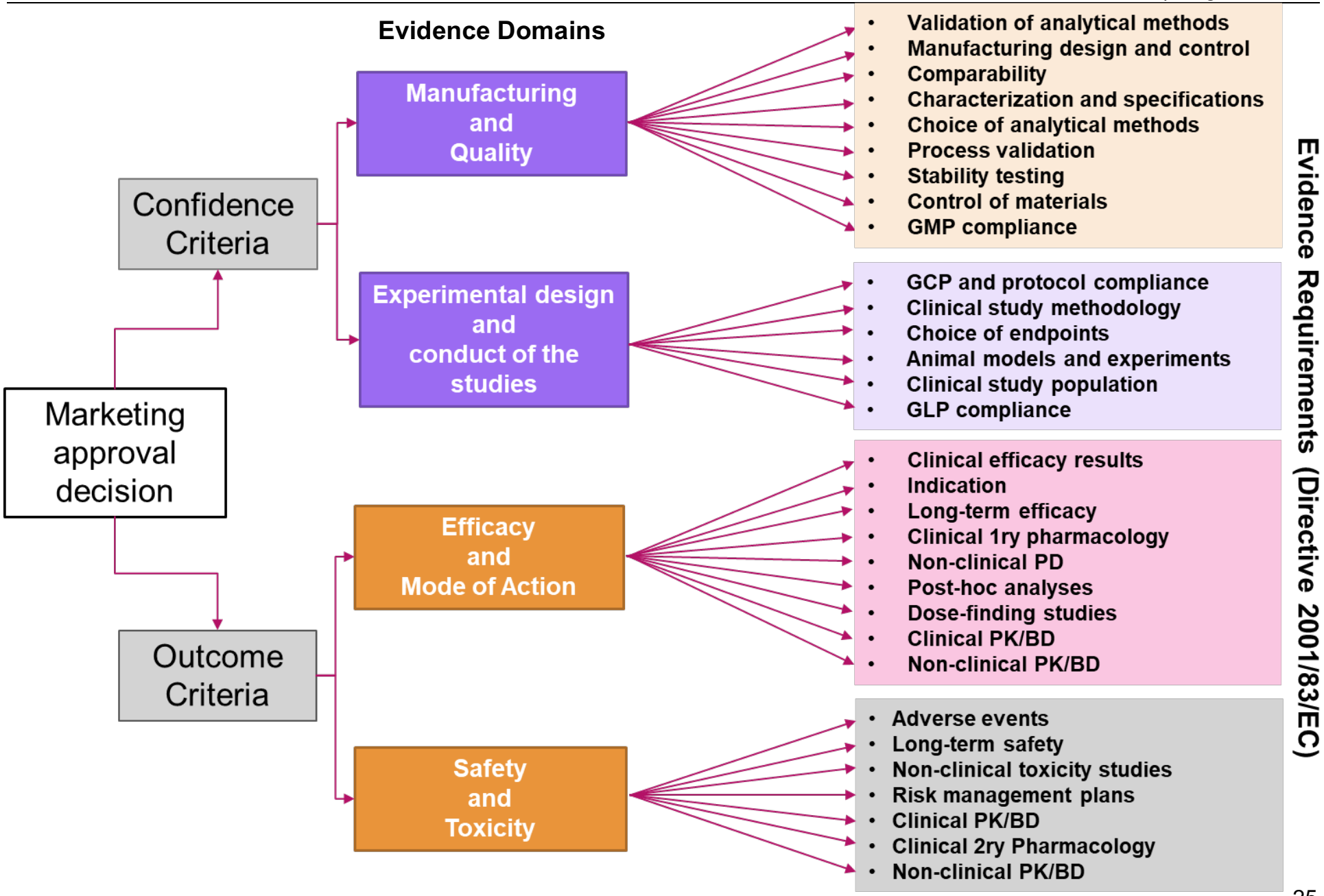

Figure 4 *Value tree for regulatory decision-making for marketing approval.*

1.6.2. Retrieval and matching of ATMP Submissions

The screening of the EMA databases yielded 22 ATMP submissions, out of which 12 were for gene therapy products (55%, including genetically modified cells), 6 were for tissue-engineered products (27%), and Four were for somatic cell therapy products (18%). The average number of ATMP submissions per year was 1.6 (standard deviation [SD], 0.9; range, 0–3). Seventeen ATMPs were successfully matched to other biologicals with the same characteristics as highlighted (Table 1 in **publication 2**). For comparison, two cohorts were created; the authorised cohort and the failed cohort (withdrawn and rejected). The average difference in time of the approval between the matched authorised products was 15.6 months (SD, 21.8 months; range, 0–67) while in the failed cohorts, the average difference between the withdrawal or rejection date of matched pairs was 41.4 months (SD, 30.9 months; range, 11–86).

1.6.3. Comparing sufficiency of the evidence of ATMPs to other biologicals

The ATMPs and the matched biologicals in the authorised and the failed cohorts were then compared for objections and divergence from the data requirement. When comparing the objections between the authorised ATMPs (n = 12) to the other authorised biologicals (n = 12), ATMPs received significantly more objections in total against the submitted evidence (p = 0.013) (Table 2 in **publication 2**). When comparing the number of objections across the evidence domains, the number of objections in the experimental design and conduct of studies was significantly higher in ATMPs (p =0.021) as well as in the efficacy and MoA domain (p =0.031) (table 2 in **publication 2**). Importantly, no significant difference in the number of objections against the quality and manufacturing evidence was observed. Also, the objections against safety and toxicity evidence showed no significant difference. In the failed cohort, there were no observed differences in objections in total as well as among the comparison domains (table 2 in **publication 2**).

The comparison of the divergence showed that ATMPs submissions tended to diverge more from the traditional data requirements by omitting or combining studies (e.g. combining toxicokinetics and local tolerance with general toxicity studies). This was evident in the total number of omitted studies from ATMPs submissions. The total number of divergence was higher in authorised ATMP cohort compared to other biologicals (p = 0.0001) (table 3 in **publication 2**). Also, across the evidence domains, divergence was more in the outcome domains (safety and toxicity as well as efficacy and MoA) of the approved ATMP submissions. In contrast, no differences in

divergence were observed in the confidence domains of the approved ATMP submissions. In the failed cohorts, no differences in divergence were observed in the total numbers or across the domains (table 3 in **publication 2**).

1.6.4. Distribution of the objections and divergence across the data requirements

We then explored the most reported objections and divergence in the ATMPs and compared them to those reported in the other biological submissions (Figure 1 in **publication 2**). The most reported and recurrent objections in ATMPs submission were related to good clinical practice and protocol compliance (76% of ATMPs submissions vs 41% of other biological submissions), which belonged to the experimental design and conduct of the studies domain. Four other data requirements had an equal frequency of objections among the products (71% of ATMP submissions each). One is the clinical efficacy results which belong to efficacy and mode of action domain. The other three data requirements were the validation of the analytical methods, the manufacturing process design and control, and the comparability of the product, all of which belonging to the manufacturing and quality domain.

Divergences in the ATMP submission were localised in two data requirements: the non-clinical toxicity studies and the clinical pharmacokinetics and biodistribution studies. Both data requirements benefited more from regulatory flexibility in the ATMPs group than other biologicals (Figure 3 in **publication 2**).

1.6.5. Differences in the timing of solving the raised objections

The raised objections can be solved either before or after the MA. Solving the objections can be achieved by submitting new data, additional analysis, additional risk minimisation measures, or modifications of the summary of product characteristics. When such solutions cannot be achieved before the approval, and the objection can be solved in the post-marketing stage, the applicant is subjected to post-marketing commitments and recommendations that are mandatory to fulfil to maintain the MA. Comparing the timing at which the issues were solved, objections in authorised ATMPs group are solved more in the post-marketing stage compared to other biologicals. This was particularly seen in the objections against clinical efficacy results, long term safety and efficacy. Also, some manufacturing and quality raised issues can be completed in the post-marketing stage, such as continuing the validation runs, improving the control of the manufacturing process, and adjusting and tightening the product specifications.

1.6.6. Investigating possible reasons for the observed differences

The observed differences in the objections in the evidence domains motivated us to investigate other causes that might indirectly affect the sufficiency of the evidence in the submissions. We first explored the type of applicant which can either be a large biopharmaceutical company or a small or medium-sized enterprise (SME). ATMPs submissions came mainly from SMEs (76% vs 12% in the other biologicals), with only 4 ATMPs submissions from large pharma. We then analysed the interaction of the developers with the regulators through scientific advice. Strikingly, the utilisation of scientific advice was nearly equal in both groups. ATMPs developers sought scientific advice three times on average while other biological developers sought scientific advice at 3.1 times on average. Finally, due to the higher objection frequency in the clinical efficacy and the conduct of the clinical studies in the ATMP group, we investigated the difference in the design and number of patients recruited in the main studies. Randomised controlled trials, considered the gold standard for demonstrating efficacy, were less in ATMP submissions (58% in ATMPs vs 83% in other biologicals). Additionally, the number of patients recruited in the main clinical study was significantly less in the approved ATMPs vs in the other biologicals (median: 114 vs 241; $Z = -2.510$, $p = 0.009$).

1.7. Discussion

The main objective of this study was to investigate the sufficiency of the evidence in ATMP submissions and provide lessons learned to mitigate such pitfalls in future developments. We employed two main indicators for the sufficiency of evidence from the regulatory point of view; the objections and the divergence from data requirement. Both indicators enabled us to estimate the sufficiency of the evidence at the time of regulatory approval. The objections enabled us to understand the aspects of the submissions where the regulators will accept no or minimum compromises in the evidence. On the other hand, the divergence enabled us to identify the area where the regulatory flexibility will allow the regulators to accept more compromises in the evidence. We employed a control group in our analysis, which comprised a matched cohort of other biologicals such as monoclonal antibodies. The control group created several advantages that were missing in the previous studies that investigated ATMP submissions (7,27,28). First, the comparison provided an estimate of how different the regulatory evaluation of ATMPs from the other closely related biological products. Second, the matched pair comparison and the data visualisation across the evidence domains exposed the areas of evidence from which the differences are originating. Third, the comparison showed to which degree ATMPs submissions diverge from the traditional data requirement as opposed to the other biologicals. Finally, matching both cohorts eliminated, to a great extent, the impact of the confounders that can affect the amount of evidence available for each product.

It is acknowledged that the study has some limitations. First, ATMPs and other biologicals are a heterogeneous group of products that might share a biological precursor material but differ in aspects such as manufacturing and the expected long-term efficacy. However, such limitation does not invalidate the comparison, given that ATMPs as well as biologicals are subjected to the same regulations and need to comply with the same data requirements. Hence, the amount and the quality of evidence in ATMP submissions should not differ significantly from other biological products. Second, is the small sample size; however, the matched comparison design and the use of non-parametric analysis assisted in overcoming the limitation of the sample size and the overestimation of the effect size. Finally, the use of public documents for extracting the information can be a limiting factor since commercially confidential information is usually omitted (40). However, since the same type of

document was used across all products, it is expected that lack of information in a certain section will not skew the data comparison between both cohorts.

The comparison of the evidence domains showed that manufacturing and quality testing is a challenging aspect to all biological medicinal products, whether it is an ATMP or not. However, the distribution of the objection in this domain showed that validation of the analytical methods and controlling of the manufacturing process are particularly challenging for ATMPs.

An unexpected outcome of the comparison is the prevalence of objections among aspects that are not adherently related to the product type, mainly the GCP compliance and the protocol adherence for the conducted clinical studies. Furthermore, the approved ATMPs received more objections regarding clinical efficacy and long-term safety and efficacy. Such issues, coupled with the observation that ATMPs clinical trials had more uncontrolled trials and smaller sample sizes, point toward the clinical development as the most challenging aspect for ATMPs submission.

An initial argument is that ATMPs mainly target rare diseases with fewer patients, so it is expected that the trial designs and recruited numbers are affected. However, the matched group of other biologicals contained the same number of products developed for rare indications and showed significantly more patients recruited in the clinical trials and more robust results. Hence, the plausible explanation of the difference is related to the experience and size of the developer. As we showed, ATMPs are mainly developed by SMEs, which might lack the experience and the finance to conduct large scale clinical trials and assemble a strong body of evidence before the submission. As a result, the developers try to move a considerable amount of the raised objections as post-marketing commitments. This strategy might be based on the notion that; once generating revenue from the marketing of the product, the developer can bear the fulfilment of further requirements.

The implications of having many issues in the evidence at the market entry stage are the inability to achieve favourable HTA assessments and reimbursement decisions. Therefore, obtaining an MA while having a gab in evidence can lead to product stagnation due to failure to secure reimbursement, consequently, the lack of market access. This observation is confirmed by other studies showing that the suboptimal clinical efficacy data was the main result of failing in reaching an agreement on

reimbursement (20,41). Such circumstances have created paradoxical settings where the applicant needs to conduct confirmatory trials in the post-marketing phase to fulfil the regulatory commitments while, at the same time, struggling to fulfil the data requirements for HTA (42). Therefore, ATMPs developers become overwhelmed with addressing issues that should be solved before reaching the market, which adds significant financial and workload burden. This analysis might partially explain why ATMPs has more post-marketing withdrawals compare to the general withdrawal rates.

1.8. Concluding Remarks

Our results have shown that regulations offer a reasonable degree of flexibility to ATMPs developers to bring these products to the market. This flexibility comes with caveats, however. Authorized ATMPs had more issues in the submitted evidence compared to other biologicals, particularly in the clinical data packages. In addition, given the identified high divergence from traditional data requirement in the non-clinical package, conducting a well-informed benefit-risk assessment by regulators might be challenging. To overcome the shortcomings, the regulators impose extensive post-marketing measures on applicants. However, these issues complicated the post-marketing phase, where the applicant struggles with achieving a positive reimbursement decision due to the suboptimal evidence. Such post-marketing settings create a demanding situation for the developers and increase the risk of failure. As such, developing a strong regulatory submission package can help in mitigate potential evidence deficiencies that may jeopardise product market success.

1.9. Future Perspectives

The gained insights enable us to propose recommendations for developing the next-generation cell and gene-based therapeutics. First, the quality and the robustness of clinical trial design for ATMPs need to be improved. The number of recruited patients and the statistical robustness of the trials need to increase. The interaction with the regulators through scientific advice from the local authorities and the EMA can be better utilised. Such interactions can help establish better trial designs and familiarise both sides with the products. On a larger scale, financial aid to SMEs can help them conduct more powered and controlled clinical trials, whenever possible. Lastly, increasing the regulatory knowledge and experience among the academic institutions and the SMEs can greatly impact their product development.

Another way to streamline product approval and market access is to bridge the evidence requirements between the HTAs and regulatory assessments. There is currently a trend in the HTA field toward using multi-criteria decision analysis (MCDA) for reimbursement decisions. In our project, we already introduced this concept in clustering the data requirements using a value tree similar to the ones utilised for decision analysis. The value tree can be utilised for an MCDA framework for regulatory evaluations. Another utility of the MCDA framework is using it for evaluating translational scientific projects in academic and industrial settings. The framework can be adapted to various stages of development to track the performance of the projects and estimate the probability of success. This can be achieved by assigning a weight for each data requirement in the value tree (Figure 4). By scoring the performance of the projects in fulfilling these data requirements, an estimate for the probability of success can be calculated. Such an approach can be used as a Go/no-Go decision-making tool for academic institutes and as a project prioritising tool for companies.

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2. STATUTORY DECLARATION / DECLARATION OF OWN CONTRIBUTION

2.1. Statutory Declaration

"I, Magdi, Elsallab, by personally signing this document in lieu of an oath, hereby affirm that I prepared the submitted dissertation on the topic "Investigating and Guiding Evidence Generation to Inform Regulatory Decision-Making for Advanced Therapy medicinal products (ATMPs)" – "Beurteilung und Begleitung der Generierung von Beweisen, um die regulatorische Entscheidungsfindung für Arzneimittel für neuartige Therapien (ATMPs) zu unterstützen", independently and without the support of third parties, and that I used no other sources and aids than those stated.

All parts which are based on the publications or presentations of other authors, either in letter or in spirit, are specified as such in accordance with the citing guidelines. The sections on methodology (in particular regarding practical work, laboratory regulations, statistical processing) and results (in particular regarding figures, charts and tables) are exclusively my responsibility.

Furthermore, I declare that I have correctly marked all of the data, the analyses, and the conclusions generated from data obtained in collaboration with other persons, and that I have correctly marked my own contribution and the contributions of other persons (cf. declaration of contribution). I have correctly marked all texts or parts of texts that were generated in collaboration with other persons.

My contributions to any publications to this dissertation correspond to those stated in the below joint declaration made together with the supervisor. All publications created within the scope of the dissertation comply with the guidelines of the ICMJE (International Committee of Medical Journal Editors; www.icmje.org) on authorship. In addition, I declare that I shall comply with the regulations of Charité – Universitätsmedizin Berlin on ensuring good scientific practice.

I declare that I have not yet submitted this dissertation in identical or similar form to another Faculty.

The significance of this statutory declaration and the consequences of a false statutory declaration under criminal law (Sections 156, 161 of the German Criminal Code) are known to me."

Date **Signature**

2.2. Declaration of Own Contribution

Magdi Elsallab contributed the following to the below-listed publications:

- **Publication 1:**

Authors: Magdi Elsallab; Bruce L Levine; Alan S Wayne; Mohamed Abou-El-Enein.

Title: CAR T-cell product performance in haematological malignancies before and after marketing authorisation.

Journal: The Lancet Oncology. Volume: 21, Issue: 2, Pages: e104 - e116.

Year: 2020

Detailed description of the contribution:

- conception and design
- literature search
- Data collection
- Generating tables (1, 2, 3) and supplementary tables (1, 2, 3, 4, 5)
- Generating figures (1, 2, 3)
- Data analysis
- Data interpretation
- Writing the manuscript

- **Publication 2:**

Authors: Magdi Elsallab; Christopher Bravery; Andreas Kurtz; Mohamed Abou-El-Enein.

Title: Mitigating Deficiencies in Evidence during Regulatory Assessments of Advanced Therapies: A Comparative Study with Other Biologicals.

Journal: Molecular Therapy. – Methods & Clinical Development. Volume: 18, Pages: 269–279.

Year: 2020

Detailed description of the contribution:

- Literature review
- Study design
- Defining indicators for the comparison
- Database search and identification of ATMPs submissions
- Text mining and text extraction from the European public assessment reports (EPARs)
- Quantification and coding of the raw data
- Statistical data analysis in SPSS
- Data coding and visualisation using R studio
- Generating tables (1,2,3) in the submitted publication as well as tables (1, 2, and 3) in the supplementary materials of publication 2

- Generating figures (1,2,3) in the publication as well as figures (1,2,3,4,5) in the supplemental materials of the publication 2
- Interpretation of the results
- Draft writing of the publication

_____
Signature, date and stamp of first supervising university professor/lecturer

_____
Signature of the doctoral candidate

3. PUBLICATIONS

3.1. Publication 1

3.1.1. Journal Summary List

Journal Data Filtered By: **Selected JCR Year: 2018** Selected Editions: SCIE,SSCI
Selected Categories: **"ONCOLOGY"** Selected Category Scheme: WoS
Gesamtanzahl: 229 Journale

Rank	Full Journal Title	Total Cites	Journal Impact Factor	Eigenfactor Score
1	CA-A CANCER JOURNAL FOR CLINICIANS	32,410	223.679	0.077370
2	NATURE REVIEWS CANCER	50,529	51.848	0.074080
3	LANCET ONCOLOGY	48,822	35.386	0.146770
4	Nature Reviews Clinical Oncology	9,626	34.106	0.031890
5	JOURNAL OF CLINICAL ONCOLOGY	154,029	28.245	0.281750
6	Cancer Discovery	13,715	26.370	0.064810
7	CANCER CELL	36,056	23.916	0.091050
8	JAMA Oncology	9,488	22.416	0.048340
9	ANNALS OF ONCOLOGY	40,751	14.196	0.103620
10	Journal of Thoracic Oncology	16,601	12.460	0.038810
11	Molecular Cancer	11,626	10.679	0.021350
12	JNCI-Journal of the National Cancer Institute	36,790	10.211	0.051650
13	NEURO-ONCOLOGY	11,858	10.091	0.029150
14	LEUKEMIA	24,555	9.944	0.054750
15	SEMINARS IN CANCER BIOLOGY	6,992	9.658	0.010730
16	CLINICAL CANCER RESEARCH	78,171	8.911	0.134870
17	Trends in Cancer	1,420	8.884	0.006040
18	Journal of Hematology & Oncology	5,366	8.731	0.013620
19	Journal for ImmunoTherapy of Cancer	2,716	8.676	0.011350
20	Cancer Immunology Research	5,420	8.619	0.025380
21	CANCER RESEARCH	130,932	8.378	0.123870

3.1.2. Publication: "CAR T-cell product performance in haematological malignancies before and after marketing authorisation."

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Personal View

CAR T-cell product performance in haematological malignancies before and after marketing authorisation

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Chimeric antigen receptor (CAR) T cells represent a potent new approach to treat haematological malignancies. Two CAR T-cell therapies, tisagenlecleucel and axicabtagene ciloleucel, have been approved in Europe and the USA, as well as several other countries, for the treatment of leukaemia and lymphoma. These approvals marked a major milestone in the field of cell and gene therapies. However, the clinical development and regulatory evaluation of these innovative therapies faced several challenges that are considered important lessons learned for future similar products. Here, we examine the products' non-clinical and clinical data packages to outline the challenges encountered during the regulatory evaluation process in Europe, and to provide an update on their performance after authorisation.

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See Online for appendix

Introduction

On Aug 27, 2018, the European Commission granted marketing authorisation to axicabtagene ciloleucel (Yescarta, Kite Pharma [Gilead]; Santa Monica, USA) and tisagenlecleucel (Kymriah, Novartis; Basel, Switzerland). The products are autologous, genetically modified, chimeric antigen receptor (CAR) T cells that were approved for treating various haematological malignancies. Axicabtagene ciloleucel is approved for the treatment of adults with relapsed or refractory diffuse large B-cell lymphoma (DLBCL) and mediastinal large B-cell lymphoma (appendix p 1). Tisagenlecleucel is approved for the treatment of adult relapsed DLBCL, as well as paediatric and young adult (25 years old or younger) acute lymphoblastic leukaemia. In addition to the EU, both products are approved in the USA, Canada, and Switzerland; tisagenlecleucel is also approved in Japan and Australia.

The novelty in CAR T cells lies in part in the genetically engineered chimeric receptor,[1,2] which is a fusion protein with an extracellular antibody-derived domain, known as a single-chain variable fragment (ScFv), and an intracellular signalling component usually comprised of primary and costimulatory signalling domains. The ScFv is responsible for specific antigen recognition on the surface of tumour cells, whereas the intracytoplasmic domains are responsible for T-cell activation, eliciting targeted killing of tumour cells.[1,2] The gene encoding the receptor is delivered to T cells via a viral vector or by membrane permeabilisation techniques such as electroporation. Both products use second-generation CAR constructs with CD19 as the target surface antigen, which is expressed on healthy B cells and in B-cell malignancies (appendix p 2). The primary T-cell signalling domain in both products is CD3ζ. The costimulatory signalling domain in axicabtagene ciloleucel is CD28 and the costimulatory signal in tisagenlecleucel is produced by 4–1BB (CD137, TNSFR9).[3,4] CD28 and 4–1BB are the most widely used costimulatory domains in clinical studies investigating CAR T-cell therapy.[5] CD28 promotes effector T-cell differentiation with an exhausted phenotype (potent, short-lived cells), leading to an initial intense activation and cytokine production that diminishes rapidly,[4] whereas 4–1BB induces differentiation predominantly to memory cell subtypes that promote cellular persistence and less cytokine production.[4,6] By harnessing the specificity of antibodies and the cytotoxicity of T cells, CAR T cells have shown high potency in treating haematological malignancies, with new generations of CAR T cells being tested for the treatment of many subtypes of haematological malignancies and solid tumours.[7,8]

In the EU, CAR T cells are subject to the advanced therapy medicinal product (ATMP) legislation and guidelines. The scientific evaluation of marketing authorisation applications for ATMPs is assessed by the European Medicines Agency (EMA) via a mandatory centralised procedure.[9] Given the complex nature of developing a living drug, meeting the traditional data requirements for marketing authorisation is challenging. As a result, regulatory guidance and incentives have been continuously evolving to address the unique biomanufacturing characteristics of ATMPs, the lack of suitable animal models, and the restrictive nature of the targeted medical indications. For instance, CAR T-cell products aim to treat life threatening or debilitating conditions and thus qualify for multiple regulatory initiatives to accelerate their development,[10] such as the priority medicines scheme (PRIME) and the orphan drug designation programme (appendix p 1). However, some doubts were cast on the completeness and strength of clinical evidence submitted to the EMA to support the marketing authorisation of these products.[11–14] Furthermore, the initial negative evaluation of the products by reimbursement bodies supported the argument that authorisation decisions on these drugs were premature.[11–13] Nevertheless, the EMA tries to strike a balance between timely market availability, patient safety, and postmarketing knowledge gains, by subjecting such products to more stringent postauthorisation measures.

Since their approval in 2018, tisagenlecleucel and axicabtagene ciloleucel are subject to additional monitoring, and their developers are obligated to supplement the safety and efficacy evidence by conducting post-authorisation studies and close follow-up of treated patients for an extended period (between 5 years and 15 years).[15–17] Understanding the added clinical value of

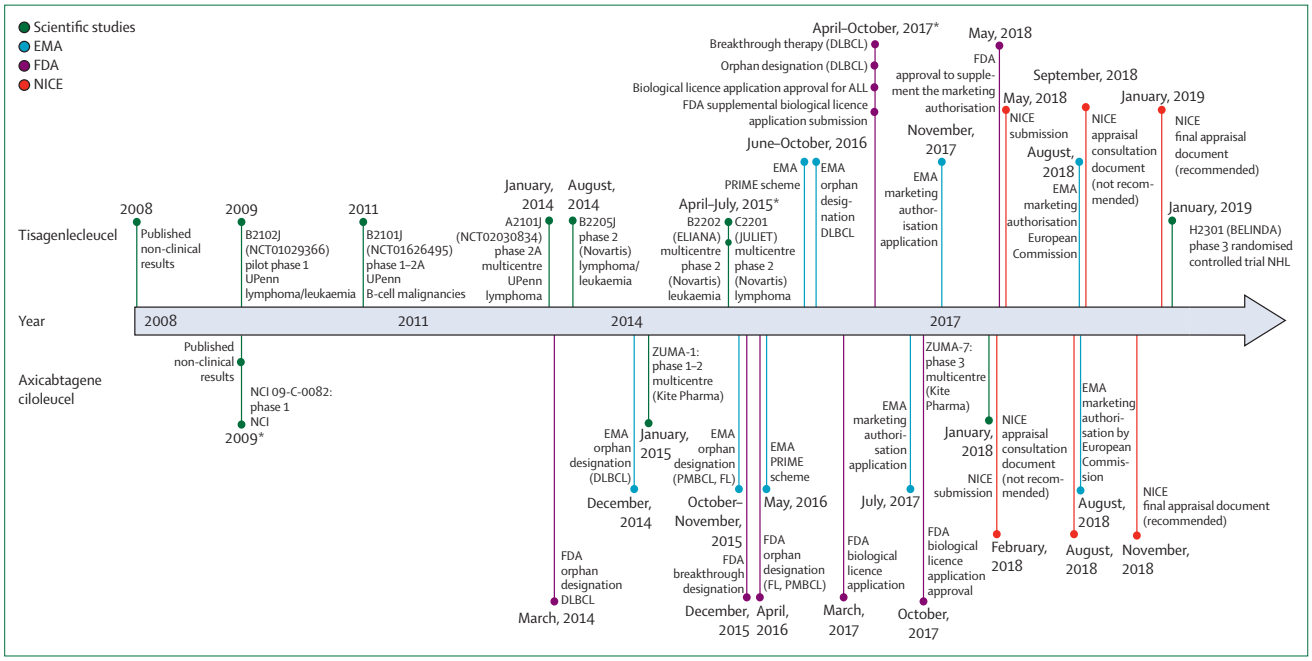


Figure 1: **Development timeline of axicabtagene ciloleucel and tisagenlecleucel**
The spaces between the lines are not to scale. EMA=European Medicines Agency. FDA=US Food and Drug Administration. NICE=National Institute for Health and Care Excellence. UPenn=University of Pennsylvania. NCI=National Cancer Institute. DLBCL=diffuse large B-cell lymphoma. PRIME scheme=Priority Medicines scheme. PMBCL=primary mediastinal B-cell lymphoma. FL=follicular lymphoma. ALL=acute lymphoblastic leukaemia. NHL=non-Hodgkin lymphoma. *Points on the same line represent the events arranged chronologically from top to bottom.

these products, and analysing gaps in evidence, could provide essential information and lessons for future ATMP development. Moreover, having two CAR T-cell products approved at the same time for similar indications created an unprecedented opportunity to scrutinise the ability of these different development pathways to inform clinical and regulatory decisions for orphan oncology therapies (figure 1). In this analysis, we examined the preauthorisation data packages submitted to the EMA to obtain marketing authorisation and then identified regulatory objections and concerns raised during the evaluation of both products. Finally, we present the postauthorisation evidence-generation strategies to fulfil the regulatory requirements and summarise the real-world data available on the use of these products.

Non-clinical proof-of-concept assessment

Our analysis reveals that the majority of the regulatory concerns raised during the evaluation of axicabtagene ciloleucel and tisagenlecleucel pertained to the clinical data and product quality packages, whereas more regulatory flexibility was shown with the non-clinical data (table 1). Nevertheless, animal models provided valuable information about the pharmacokinetics, pharmacodynamics, and some toxicological aspects of the products. Axicabtagene ciloleucel was tested by use of a CD19-expressing 38c13 mouse lymphoma cell line in an immunocompetent syngeneic lymphoma mouse model,[18] whereas tisagenlecleucel treatment studies used an immunodeficient NOD/Shi-scid IL-2Rγ^{null} human leukaemia xenograft mouse model.[6]

The disadvantage of using immunocompetent mice in the axicabtagene ciloleucel studies is that these mice only support the growth of mouse lymphoma, which hampered the efficacy and safety testing of the human-derived CAR T cells. As a result, murine-derived CAR T cells were developed and tested as a surrogate model for the proposed CAR T-cell therapy. The main limitation of these cells is that their manufacturing and cellular dynamics differ from the final human CAR T-cell product. The EMA highlighted this point and accepted the animal studies as a proof-of-concept, deeming the murine model as the most appropriate for testing.[15]

Conversely, the immunodeficient mice used in tisagenlecleucel non-clinical studies could be injected with human acute lymphoblastic leukaemia cells, allowing for the testing of the human CAR T-cell product. However, the absence of an intact immune system in this model less accurately simulates the disease in humans than does a model using immunocompetent mice, and the safety testing of on-target–off-tumour activity and cytokine-release syndrome could not be done.[19] Moreover, several CAR constructs were tested in the leukaemia model (second-generation CD28, second-generation 4–1BB, and third-generation CD28 and

	Tisagenlecleucel	Axicabtagene ciloleucel
Quality aspects		
Major objections	Documentation of GMP compliance	Inconsistent viral transduction
Other concerns	NA	No initial data on comparability and equivalence of the different processes (CLP 1.0. and CLP 2.2.); lower transduction rate in the last manufacturing process
Recommendations	Characterisation and testing of the viral vector, leukapheresis starting material, and the finished product	Enhancing manufacturing process and control of the product
Non-clinical aspects		
Major objections	NA	NA
Other concerns	Not using both CD28 and 4-1BB as the intracellular domain in the CAR construct	NA
Recommendations	NA	NA
Clinical pharmacology		
Major objections	NA	NA
Other concerns	No dose exposure relationship; less proliferation of the cells in patients with DLBCL than patients with ALL; high variability of cellular kinetics in the study groups	No relation between product characteristics and efficacy outcomes; no correlation between biomarkers and positive treatment outcomes
Recommendations	Investigate cellular kinetic parameters	NA
Clinical efficacy		
Major objections	Absence of CD19 tumour expression as a requirement for infusion in the summary of product characteristics	NA
Other concerns	ALL: delayed assessment of the tumour stage after patient enrolment affects baseline characteristics; not reflecting the study population for the submitted indication; DLBCL: testing the null hypothesis of overall response at 20% against the EMA scientific advice recommendation (overall response of 40%); excluding the effect of bridging therapy in the clinical assessment by use of modified intention-to-treat analysis; long time span (54 days) from enrolment to the infusion of tisagenlecleucel due to longer than expected manufacturing time (4–5 weeks); patients dropping out of the study with poor prognostic factors due to disease progression; introducing bias to the efficacy analysis by use of the infused modified intention-to-treat population; not including stable disease and progressive disease populations in the overall survival analysis; different baseline characteristics between non-infused patients and infused patients	DLBCL: Not doing the baseline-PET scan in the prespecified time before conditioning chemotherapy; not reflecting the study population for the submitted indication; absence of comparison with SCHOLAR-1 for a worst-case scenario by excluding patients with an Eastern Cooperative Oncology Group score of 2–4 or unknown
Recommendations	NA	NA
Clinical safety		
Major objections	NA	NA
Other concerns	Severe and life-threatening adverse effects; missing information in several patient groups	High incidence of adverse drug reactions; missing information in several patient groups
Recommendations	NA	NA

GMP=good manufacturing practice. NA=not applicable. CAR=chimeric antigen receptor. DLBCL=diffuse large B-cell lymphoma. ALL=acute lymphoblastic leukaemia. EMA=European Medicines Agency.

Table 1: **Major objections and concerns raised by the EMA during the evaluation of tisagenlecleucel and axicabtagene ciloleucel**

4–1BB).[6] Although CAR T cells with the third-generation CD28 and 4-1BB construct persisted for longer in the tumour-bearing mice, 4–1BB was the construct of choice for clinical testing, a decision that was accepted during the regulatory evaluation process.[16]

Notably, no lymphoma animal model was developed and tested as a proof of concept for tisagenlecleucel. The EMA flagged this observation; nevertheless, the agency found the absence of this animal model acceptable considering the available clinical experience and approved the product for this indication.[16] Overall, the regulatory flexibility in accepting suboptimal non-clinical data packages for both products was evident.

Clinical investigation of CAR T-cell pharmacology

Data on axicabtagene ciloleucel pharmacology were generated by the phase 1–2 ZUMA-1 trial[20] and the supportive National Cancer Institute 09-C-00082 study[21] (figure 1, table 2), whereas tisagenlecleucel relied on the pivotal phase 2 ELIANA trial[22] and supportive studies (Pedi CART19[23] [NCT01626495] and ENSIGN[22] [NCT02228096]) for the acute lymphoblastic leukaemia indication, and the JULIET study[24] for the DLBCL indication (figure 1, table 2). In these trials, proliferation, distribution, and persistence of anti-CD19 CAR T cells were measured in peripheral blood and bone marrow by qPCR and flow cytometry.[25,26]

	ELIANA (NCT02435849)	JULIET (NCT02445248)	ZUMA-1 (NCT02348216)	SCHOLAR-1
Treatment	Tisagenlecleucel	Tisagenlecleucel	Axicabtagene ciloleucel	Salvage chemotherapy
Centres in countries	25 in 11	27 in 10	24 in 1	NA
Study population	Paediatric and young adult patients with relapsed or refractory B-cell ALL	Relapsed or refractory DLBCL after two lines or more of chemotherapy and not eligible for stem cell transplantation	Relapsed or refractory DLBCL, PMLBCL, or FL after two lines or more of chemotherapy or an autologous stem cell transplantation	Refractory aggressive B-cell non-Hodgkin lymphoma (DLBCL, PMBCL, or TFL)
Median age, years (range)	11 (3–23)	59 (22–76)	58 (23–76)	55 (19–81)
Study design	Phase 2, single-arm, open-label, multicentre	Phase 2, single-arm, open-label, multicentre compared with historical data	Phase 2, single-arm, open-label, multicentre compared with historical data	Retrospective meta-analysis
Conditioning chemotherapy	Fludarabine (30 mg/m^2, intravenous daily for four doses) and cyclophosphamide (500 mg/m^2, intravenous daily for two doses); cytarabine (500 mg/m^2 daily for 2 days) and etoposide (150 mg/m^2 daily for 3 days)	Fludarabine (25 mg/m^2) and cyclophosphamide (250 mg/m^2); intravenous daily for three doses	Fludarabine (30 mg/m^2) and cyclophosphamide (500 mg/m^2); intravenous daily for three doses; treatment starts 5 days before infusion of the CAR T cells	NA
Dose	0·2–5·0 × 10^6 cells per kg (for patients ≤ 50 kg) and 0·1–2·5 × 10^8 cells (for patients >50 kg)	1·0–5·0 × 10^8 cells single infusion	2 × 10^6 (± 20%) cells per kg (minimum 1 × 10^6 cells per kg)	Salvage chemotherapy with an anti-CD20 monoclonal antibody such as rituximab
Enrolled/infused	92/75	165/111	111/101	636/523
Primary endpoints				
Overall response	Best overall disease response as a CR or CRi	Best overall disease response as a CR or PR	Best overall disease response as a CR or PR	..
Response	..	..	..	Best response as a CR or PR
Secondary endpoints	Overall response (CR and CRi) from US manufacturing facilities; percentage of patients with a best overall response of CR or CRi, with negative MRD, from all manufacturing facilities; percentage of patients with a best overall response of CR or CRi, with negative MRD, from US manufacturing facilities	Duration of response, overall survival, time to relapse, event-free survival, progression-free survival	Duration of response, progression-free survival, overall survival	CR and overall survival
Safety endpoints	Incidence of adverse events	Incidence of adverse events	Incidence of adverse events	NA

CAR=chimeric antigen receptor. EMA=European Medicines Agency. NA=not applicable. ALL=acute lymphoblastic leukaemia. DLBCL=diffuse large B-cell lymphoma. PMLBCL=primary mediastinal large B-cell lymphoma. FL=follicular lymphoma. TFL= transformed follicular lymphoma. CR=complete response. CRi=complete response with incomplete haematological recovery. PR=partial response. MRD=minimal residual disease.

Table 2: **Pivotal clinical trials for CAR T-cell products and historical controls submitted in the marketing authorisation application to the EMA**

The non-compartmental analysis of tisagenlecleucel in ELIANA and supportive studies showed an initial rapid expansion of CAR T cells in acute lymphoblastic leukaemia responders, reaching the maximal expansion in peripheral blood (C_{max}) after nearly 10 days (T_{max}).[22,23] Acute lymphoblastic leukaemia responders showed 68% more cellular expansion (C_{max}) and 43% higher exposure (area under the curve for 0–28 days; $AUC_{0–28}$) of tisagenlecleucel than did non-responders. The cells persisted in responders for longer than in non-responders, with the median time until the last measured concentration being 170·0 days in responders versus 28·9 days in non-responders. The pharmacokinetic properties of tisagenlecleucel in the peripheral blood have shown a direct correlation with endpoints in the trial for acute lymphoblastic leukaemia, including event-free survival for more than 3 months and overall response at day 28. Conversely, in the JULIET study,[24] a correlation between the cellular kinetics of tisagenlecleucel in the peripheral blood and treatment efficacy could not be shown in patients with lymphoma as no differences in the geometric means of the C_{max} or $AUC_{0–28}$ were observed between responders and non-responders.

Patients with lymphoma who responded to axicabtagene ciloleucel in the ZUMA-1 trial showed a 205% higher median C_{max} (43·6 cells per µL *vs* 21·2 cells per µL) and two times higher median $AUC_{0–28}$ (7·1 days per cells per µL *vs* 222·0 days per cells per µL) than did non-responders. The number of cells then declined to near background amounts within 3 months, with a median of 0·4 cells per µL (range of 0–15.8 cells per µL). Unlike with tisagenlecleucel, the C_{max} and $AUC_{0–28}$ of axicabtagene ciloleucel directly correlated with the clinical response in patients with lymphoma (responders tended to have more cells and longer exposure). In axicabtagene ciloleucel, the robust cellular proliferation and cytokine release promoted by the CD28 signalling domain might have influenced the high response observed in lymphoma. However, previous studies reported that CD28 CAR T cells might lack

durability and persistence, raising questions about the actual value of treatment, long-term efficacy, and the possible need for subsequent treatment.[4,27,28] The UK National Institute for Health and Care Excellence (NICE) also raised this concern during their health technology assessment of axicabtagene ciloleucel.[29]

Other factors, such as disease burden and location, T-cell phenotype, T-cell subpopulations, conditioning chemotherapy, and the tumour microenvironment have also been reported to affect the cellular kinetics of CAR T cells.[30,31] For instance, differences in the cellular kinetics of CAR T-cells between leukaemia and lymphoma might be attributed in part to the fact that leukaemia cells are often present in peripheral blood, whereas lymphoma cells mostly reside in lymphoid tissues. As noted, 4-1BB costimulation promotes cellular differentiation of memory cell phenotypes leading to longer persistence but weaker initial response compared to CD28 costimulation. Such characteristics of 4-1BB, coupled with the difference in microenvironment, can partially explain the observed variation in tisagenlecleucel's cellular kinetics between lymphoma and leukaemia. These factors prompted the EMA to recommend further characterisation of the cellular kinetics of tisagenlecleucel for both indications as part of the postauthorisation measures. In their efforts to address this point, the developers of tisagenlecleucel established a mixed-effects model describing the effect of tocilizumab and corticosteroids—treatments that are used to manage cytokine-release syndrome—on cellular kinetics.[32] The model can be adapted to characterise the expansion and persistence of CAR T cells across different disease indications, within various cell types, and between different costimulatory domains.[32]

Post-treatment outcomes and analysis of results

We further analysed the European public assessment reports for the submitted clinical data packages of both products.[15,16] Tisagenlecleucel showed a clear efficacy profile in patients with acute lymphoblastic leukaemia. The results of the ELIANA study (data cutoff: April 25, 2017) showed that of 92 patients, 61 (66%) achieved an overall response and 45 (49%) a complete response using the intention-to-treat population, and of 75 patients, 61 (81%) achieved an overall response and 45 (60%) a complete response using the infused modified intention-to-treat population, with a median overall survival of 19·4 months after a median follow-up of 10·5 months.[16] However, when exploring the results of lymphoma clinical trials for tisagenlecleucel, there were more noticeable differences between the intention-to-treat population versus the infused modified intention-to-treat population in the efficacy analysis of JULIET (data cutoff: Dec 8, 2017; figure 2; appendix pp 3–4). These differences also extended to the median overall survival, which was 8·2 months for the intention-to-treat analyses and 11·7 months for the modified intention-to-treat analysis.[16] These differences were not seen for axicabtagene ciloleucel in the ZUMA-1 trial, where overall and complete response proportions were similar between the intention-to-treat and modified intention-to-treat analyses (figure 2), as was median overall survival (17·4 months in the intention-to-treat analysis and not reached in the modified intention-to-treat analysis [data cutoff: Aug 11, 2017]; figure 2; appendix p 4).[15] The results of pivotal trials in lymphoma and leukaemia with both CAR T-cell products met the primary endpoint of best overall response in more than 20% of patients—an endpoint that was decided based on data obtained from historical studies.[23,24,33]

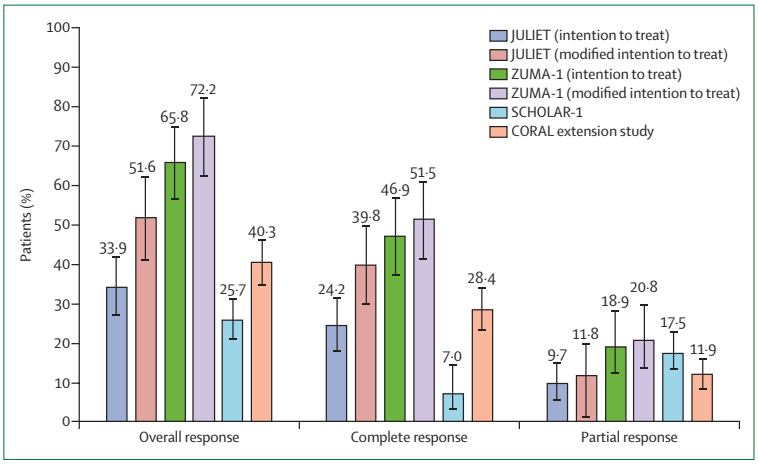
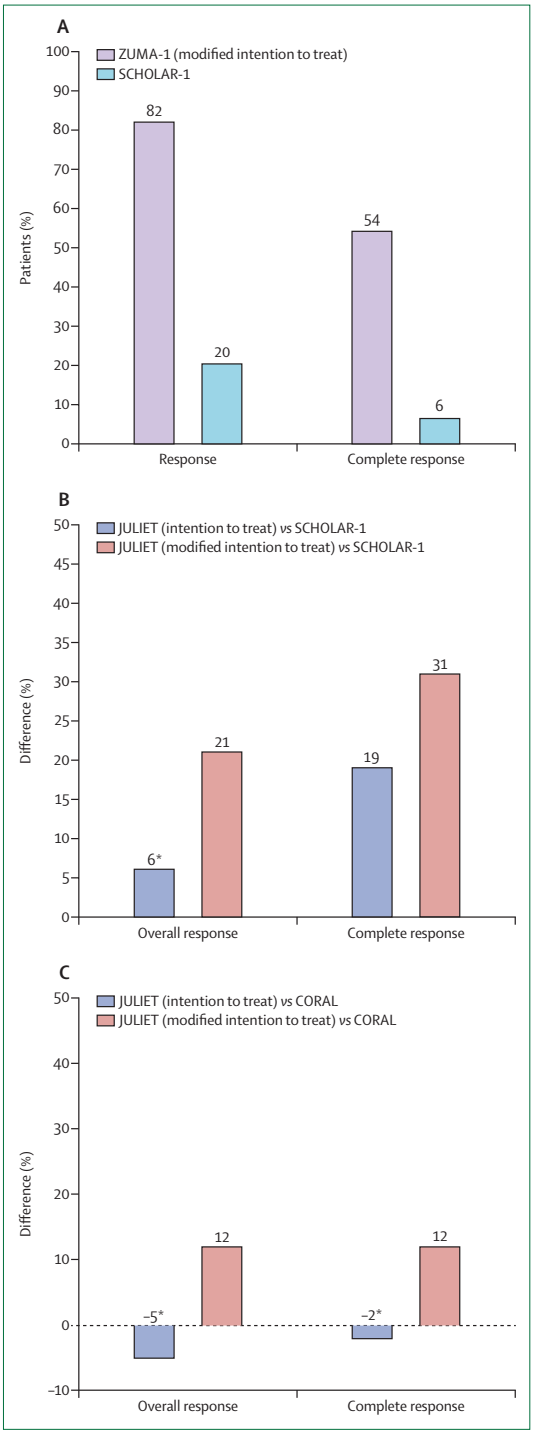


Figure 2: **Unadjusted aggregated efficacy results for JULIET (tisagenlecleucel), ZUMA-1 (axicabtagene ciloleucel), SCHOLAR-1, and CORAL extension studies with different analysis populations for the treatment of DLBCL**

Error bars represent the CI. The number of patients in each study analysis group (n=165, 93, 111, 101, 523, or 278) in order of the key from top to bottom. Data cutoff for ZUMA-1: Aug 11, 2017, with a median follow-up of 15·1 months. Data cutoff for JULIET: Dec 8, 2017, with a median follow-up of 13·9 months. DLBCL=diffuse large B-cell lymphoma.

For the tisagenlecleucel JULIET study, the EMA explored the reasons for the variability seen in the different analyses of clinical outcomes. They found that this variability in results could be attributed to the high dropout (30%), which changed the number of patients included in each analysis. This dropout resulted from a strict inclusion criterion where enrolled patients should not have had any substantial worsening of their disease status before the administration of the cellular product.[16] However, the median time from enrolment to infusion was 54 days due to manufacturing delays, which led to patient deterioration and exclusion from the study.[16] As such, the EMA concluded that selection bias was introduced in the modified intention-to-treat population. Additionally, 20% of patients who dropped out had a response to the bridging chemotherapy that was administered while waiting for product manufacturing (patients in the axicabtagene ciloleucel ZUMA-1 trial did not receive bridging chemotherapy). These observations have prompted the Inter-Committee Scientific Advisory Group on Oncology to advise the EMA that the evaluation of the intervention should be based on the whole treatment regimen, and not only on the infused cellular

product. Taking all these points into consideration, the EMA concluded that the reliability of using the outcomes of the infused modified intention-to-treat population as efficacy estimators was not sufficient to reflect an accurate assessment of clinical benefit (table 1). As such, the EMA used the enrolled intention-to-treat population data to evaluate the differences in outcomes against the historical controls, and to conduct the benefit–risk assessment for both products.[16]



The role of historical controls in evaluating clinical outcomes

The assessment of treatment benefit for both tisagenlecleucel and axicabtagene ciloleucel was supplemented by comparisons with historical control groups. In the case of single-arm studies with no control arms, regulatory and health technology assessment agencies show more flexibility in allowing comparisons with historical data. Analytical tools, such as matching-adjusted indirect comparisons and network meta-analyses, have been introduced for regulatory submissions and health technology assessments.[34,35] However, the choice of a suitable comparator remains challenging, and caution is needed during the interpretation and evaluation of the results.[35] Novartis tried to establish a comparison for the leukaemia indication for tisagenlecleucel by pooling data from their leukaemia studies (ELIANA [NCT02435849], ENSIGN [NCT02228096] and Pedi CART19 [NCT01626495]) and matching the data to other studies of marketed therapies, such as blinatumomab; a combination of clofarabine, cyclophosphamide, and etoposide; and clofarabine monotherapy.[36–40] Despite the potential bias due to small sample size, confounding patient populations, and matching on few variables, tisagenlecleucel showed consistent superiority across all the comparators, endpoints, and sensitivity analyses.[16]

In the lymphoma indication, tisagenlecleucel and axicabtagene ciloleucel were compared with SCHOLAR-1,[41] which is a retrospective, patient-level, pooled analysis of the outcome of currently available standard of care in patients with refractory, aggressive non-Hodgkin lymphoma. The comparison of response in ZUMA-1 with SCHOLAR-1 is shown in figure 2 for the unmatched and unadjusted data. The reliability of SCHOLAR-1 as a comparator with ZUMA-1 was thoroughly assessed during health technology assessments by NICE,[29] and was eventually accepted. This acceptance was attributed to the availability of individual patient data to Kite Pharma

***Figure 3*: Matched comparisons of results from axicabtagene ciloleucel and tisagenlecleucel pivotal clinical trials with historical comparators for the treatment of DLBCL**

Figures are reproduced from data presented in the European public assessment reports for both products, and a published article.[15,16,42] (A) Comparison of responses between ZUMA-1 and SCHOLAR-1 (data cutoff ZUMA-1: Aug 11, 2017, median follow-up 15·1 months). (B) The differences in overall response and complete response between JULIET and SCHOLAR-1 by analysis population (data cutoff: Dec 8, 2017, median follow-up 13·9 months). (C) The differences in overall response and complete response between JULIET and CORAL extension studies (data cutoff: Dec 8, 2017, median follow-up 13·9 months).
*No significant difference in responses (p>0·05).

(Gilead), which was the sponsor of SCHOLAR-1, enabling the company to match patients in both trials. In the matched analysis, axicabtagene ciloleucel showed superiority over the standardised historical data, even after adjusting the populations to a stricter baseline in a worst-case scenario analysis (figure 3A).

Since only the published aggregated data of SCHOLAR-1 were available to developers of tisagenlecleucel, other historical comparators were explored. In addition to SCHOLAR-1, the pooled CORAL extension data were used for comparisons (figure 2, appendix p 4).[16] The pooled CORAL extensions emerged from the main CORAL study and were considered by the EMA and NICE as a more suitable comparator than SCHOLAR-1 for evaluating tisagenlecleucel due to similarities in the populations enrolled.[43] The main CORAL study[44] compared salvage chemotherapy regimens followed by stem cell transplantation, whereas the pooled extension studies followed up patients who did not proceed to stem cell transplantation, or had a second relapse after transplantation.[45,46] Novartis used matching-adjusted indirect comparisons to match the individual patients from JULIET to both historical controls. When running the matched analysis with the modified intention-to-treat population, tisagenlecleucel showed a significant difference in overall response and complete response compared with that in both the pooled CORAL extensions and SCHOLAR-1 (figure 3B, C). However, when analysing the intention-to-treat population, the product did not show a significant difference in overall response when compared with the pooled CORAL extensions and SCHOLAR-1 studies (figure 3B, C). Nevertheless, tisagenlecleucel showed a significantly longer median overall survival (10·6 months for intention to treat and 16·3 months for modified intention to treat) compared with the pooled CORAL results where the median overall survival was 5·8 months.

Due to the aforementioned inconsistencies in efficacy analysis for the different populations, 12 members of two EMA committees involved in the evaluation process disagreed with granting authorisation for tisagenlecleucel in the lymphoma indication. Eventually, the product was authorised in lymphoma by taking into account the higher response durability in tisagenlecleucel compared with the controls. Nevertheless, Novartis was mandated to do extensive postauthorisation efficacy studies in the form of data collection on treated patients in dedicated registries and an interventional phase 3, randomised, controlled trial of tisagenlecleucel versus platinum-based immunochemotherapy (BELINDA; NCT03570892; table 3). BELINDA began enrolment in May, 2019, with a target enrolment of 318 patients across the USA, Australia, Germany, Japan, and Spain.

Associated risks and measures to ensure patient safety

Both tisagenlecleucel and axicabtagene ciloleucel used integrating viral vectors (appendix p 2), which might raise the concern of insertional oncogenesis due to semi-random integration patterns. Lentivectors used in tisagenlecleucel are considered safer than γ-retroviral vectors used in axicabtagene ciloleucel, as their integration patterns do not favour transcriptional start sites.[54] However, mature T cells are resistant to malignant transformation after transduction with an integrating viral vector,[55] which was Kite Pharma's (Gilead) justification for using a γ-retroviral vector.[15] Notably, axicabtagene ciloleucel received advice from the EMA in the form of early discussions on the risks of insertional mutagenesis under the PRIME scheme. Another concern of the use of viral vectors is the generation of a replication-competent virus.[56] The risk of replication-competent virus formation was considered low by the EMA as both vectors are replication incompetent and stringently tested for the absence of replication-competent virus.[15,16] Studies have shown that the risk of formation of replication-competent virus either by a lentiviral or retroviral vector is very low.[57] As a result, the US Food and Drug Administration is revising the regulations on testing for replication-competent virus, which might result in a reduction of follow-up testing in the case of vectors where there is substantial experience with safety.[58] Nevertheless, to ensure patient safety and accumulate more data about the products, the EMA requires postauthorisation safety studies where data from patients treated with these products must be collected for a period of up to 15 years to assess the long-term safety of both vector types as part of the risk minimisation plan (table 3).

During clinical testing, all patients infused with either of the two products had adverse events (appendix p 5). Serious adverse events were mainly attributed to cytokine-release syndrome and neurological complications. Other frequent serious adverse events were infections, tumour lysis syndrome, and febrile neutropenia. Axicabtagene ciloleucel showed a higher incidence of cytokine-release syndrome and neurological events than did tisagenlecleucel (appendix p 5), and these events were associated with higher concentrations of cytokines and a higher maximum number of axicabtagene ciloleucel cells in the blood (C_{max}).[33] The clinical management plan for adverse events in both studies was seen as sufficient by the EMA. For instance, CAR T-cell therapies were to be provided only in qualified centres that also had available tocilizumab as a treatment for cytokine-release syndrome. Additionally, the clinical trial sponsors had to offer an educational programme for each participating centre that was targeted towards centre personnel and patients. As part of the postauthorisation measures, each applicant had to collect postauthorisation safety data in dedicated registries. For tisagenlecleucel, the data were collected through the European Society for Blood and Marrow Transplantation and the Center for International Blood and Marrow Transplant Research registries.[59] As part of the ongoing effort, the EMA released the proposed data

	Indication	Primary objective	Obligatory by EMA	Study type	Phase	Control	Randomised	Start date	Number of Patients	Current status
Tisagenlecleucel										
Stein et al (2019)[32]	ALL or DLBCL	Cellular kinetic parameters and the effect of CRS medications	No	Experimental	NA	NA	NA	NA	NA	Published mixed-effects model analysing the effects of CRS medications on cellular kinetics
CCTL019B2401[47]	ALL	Evaluate the efficacy in patients with ALL younger than 3 years	Yes	Observational; registry-based	Phase 4	NA	NA	Q4, 2018	NA	Data from EBMT and CIBMTR registries will be used for the observational study; February, 2019: statistical plan for the study submitted to the committee for advanced therapies
ELIANA[48] (NCT02435849, CCTL019B2202)	ALL	Long-term efficacy and safety of tisagenlecleucel in the ELIANA study	No	Follow-up	Phase 2 multicentre	No	No	April, 2015	97 enrolled, 79 infused at last data cutoff	Official 24-month report of ELIANA (expected Q4, 2019); last published results: April, 2018, data cutoff; 24-month median follow-up; median duration of response not reached; median overall survival not reached; 66% overall survival (modified intention to treat; 24 months)
CCTL019B2401[47]	DLBCL	Evaluate efficacy outcome measures, including the manufacturing time	Yes	Observational; registry-based	Phase 4	No	No	Q4, 2018	NA	February, 2019: statistical plan for the study submitted to the committee for advanced therapies
JULIET[49] (NCT02445248, CCTL019C2201)	DLBCL	Long-term efficacy and safety of tisagenlecleucel in the JULIET study	Yes	Follow-up	Phase 2 multicentre	No	No	July, 2015	167 enrolled, 115 infused	Official 24-month report of JULIET (expected in September, 2019); last published results: May, 2018, data cutoff; 19-month median follow-up; median duration of response not reached; median overall survival of 11·1 months for infused patients; 43% overall survival at 18 months
BELINDA (CCTL019H2301, NCT03570892)	DLBCL	Efficacy of tisagenlecleucel *vs* standard of care in adult patients with refractory or relapsed NHL	Yes	Interventional	Phase 3 multicentre	Yes (active comparator)	Yes	May, 2019	318 (estimated)	Recruiting; primary endpoint: event-free survival
CCTL019B2401[47]	ALL or DLBCL	Long-term safety of tisagenlecleucel in patients with ALL and DLBCL based on disease registry	Yes	Observational; registry-based	Phase 4	NA	NA	Q4, 2018	NA	February, 2019: statistical plan for the study submitted to the committee for advanced therapies
(NCT02445222, CCTL019A2205B)[16]	ALL or DLBCL	Long-term follow-up of patients exposed to lentiviral-based CD19 directed CAR T-cell therapy	Yes	Observational; registry-based	Phase 4	NA	NA	Nov, 2015	620 (estimated)	Follow-up of all the patients who have been infused with tisagenlecleucel for 15 years; annual safety reports and 5-yearly interim reports will be submitted to the EMA; final report of study results in December, 2038
Axicabtagene ciloleucel										
ZUMA-1[50] (NCT02348216)	DLBCL, PMLBCL, or FL	Long-term efficacy and safety of axicabtagene ciloleucel in the ZUMA-1 study	No	Follow-up	Phase 2 multicentre	No	No	January, 2015	111 enrolled, 101 infused	EMA 24-month result update based on intention-to-treat (n=111); 68% overall response; 50% CR; median duration of response not reached; median overall survival of 17·4 months; 48% 24-month overall survival

(Table 3 continues on next page)

	Indication	Primary objective	Obligatory by EMA	Study type	Phase	Control	Randomised	Start date	Number of Patients	Current status
(Continued from previous page)										
Non-interventional registry study[15]	DLBCL	Long-term safety of axicabtagene ciloleucel in the postmarketing setting	Yes	Observational; registry-based	Phase 4	NA	NA	NA	NA	Planned
ZUMA-2 (NCT02601313)	MCL	Efficacy of axicabtagene ciloleucel in patients with refractory or relapsed MCL	No	Interventional	Phase 2 multicentre	No	No	November, 2015	105	Active; expected primary completion date in July, 2019; primary endpoint: overall response
ZUMA-3[51] (NCT02614066)	ALL	Safety and efficacy of axicabtagene ciloleucel in adult participants with refractory or relapsed ALL	No	Interventional	Phase 1–2 multicentre	No	No	March, 2016	100 (estimated)	Recruiting; expected primary completion date in January, 2020; end of phase 1 results: September, 2018, data cutoff; 45 infused patients; 41 evaluable patients; 16-month median follow-up; 68% overall response (CR + CRi); 73% minimal residual disease negative; no DLT
ZUMA-4[52] (NCT02625480)	ALL	Safety and efficacy of axicabtagene ciloleucel in paediatric and adult participants with refractory or relapsed ALL	No	Interventional	Phase 1–2 multicentre	No	No	February, 2016	100	Recruiting; expected primary completion date in July, 2021; end of phase 1 results October, 2018, data cutoff; 24 infused patients; 13-month median follow-up; overall response of 100% (2×10^6), 64% (1×10^6; 68 mL), and 71% (1×10^6; 40 mL) in three dose groups
ZUMA-5 (NCT03105336)	NHL	Safety and efficacy of axicabtagene ciloleucel in patients with indolent refractory or relapsed indolent NHL	No	Interventional	Phase 2 multicentre	No	No	June, 2017	160 (estimated)	Recruiting; expected primary completion date in March, 2020; primary endpoint: overall response
ZUMA-6[53] (NCT02926833)	DLBCL	Safety and efficacy of axicabtagene ciloleucel in combination with atezolizumab in adults with refractory or relapsed DLBCL	No	Interventional	Phase 1–2 multicentre	No	No	September, 2016	37 (estimated)	Active; end of phase 1 results: January, 2018, cutoff; 12 infused patients; 4·4 median follow-up; dose-limiting toxicity in 1 patient; all patients had at least one adverse effect (92%, grade ≥3); overall response in 9 (90%) of 10 evaluable patients
ZUMA-7 (NCT03391466)	DLBCL	Efficacy of axicabtagene ciloleucel against the standard of care in relapsed or refractory DLBCL	No	Interventional	Phase 3 multicentre	Yes	Yes	December, 2017	350 (estimated)	Recruiting; 71 study locations (Europe, North America, Australia, Israel); primary endpoint: event-free survival; secondary endpoints: overall response, overall survival, progression-free survival, duration of response

EMA=European Medicines Agency. ALL=acute lymphoblastic leukaemia. DLBCL=diffuse large B-cell lymphoma. CRS=cytokine release syndrome. NA=not applicable. Q4=fourth quarter of the year (October, November, and December). EBMT=European Society for Blood and Marrow Transplantation. CIBMTR=Center for International Blood and Marrow Transplant Research. NHL=non-Hodgkin lymphoma. CAR=chimeric antigen receptor. PMLBCL=primary mediastinal large B-cell lymphoma. FL=follicular lymphoma. CR=complete response. MCL=mantle cell lymphoma. CRi=complete response with incomplete haematological recovery. DLT=dose limiting toxicity.

Table 3: **Postauthorization studies for tisagenlecleucel and axicabtagene ciloleucel based on the submitted risk management plan**

elements that should be fulfilled by the registries to capture all the necessary information on the safety and efficacy of CAR T-cell products.[60]

Complex logistics and regulatory considerations

Although clear clinical benefits were obtained from clinical trials investigating tisagenlecleucel and axicabta-

gene ciloleucel, issues pertaining to manufacturing and supply chain management should be highlighted. For instance, the locations of the studies might have influenced the outcomes of both treatments and their evaluation by the EMA. ZUMA-1 was done in the USA, except for one patient, who was treated in Israel (table 2). Due to the absence of European patients in ZUMA-1, the developer was advised, under the PRIME scheme, to include European patients in the planned phase 3 trial (ZUMA-7, NCT03391466).[15] Conversely, JULIET was done at 27 sites in ten countries across four continents. Even though clinical, collection, and infusion sites were global, tisagenlecleucel for JULIET was mainly manufactured in the USA, with some manufacturing in Germany. This restricted capacity of the manufacturing might have posed a challenge to the product supply chain and manufacturing coordination, and prolonged the time from enrolment to infusion in the JULIET study. As a result, details on tisagenlecleucel manufacturing turnaround time was required by the EMA as part of the postauthorisation efficacy studies.[16]

Postmarketing performance of CAR T-cell products

The up-to-date clinical follow-up shows that both tisagenlecleucel and axicabtagene ciloleucel elicit a durable response in the approved leukaemia and lymphoma indications (table 3). In patients with leukaemia, the median duration of response and overall survival were not reached at a median follow-up of 24 months in the ELIANA study.[48] In patients with lymphoma, the last update from the tisagenlecleucel JULIET trial showed a median overall survival of 11·1 months, and the median duration of response was not reached (table 3).[49] The 24-month results of the axicabtagene ciloleucel ZUMA-1 study showed a median overall survival of 17·4 months, and the median duration of response was not reached.[15]

Two postmarketing real-world studies were published evaluating patients with non-Hodgkin lymphoma that were treated with standard-of-care axicabtagene ciloleucel in the USA.[61,62] Nastoupil and colleagues[61] reported results of 295 patients treated as of August, 2018, at 17 academic USA centres. 240 of 274 patients had cytokine-release syndrome, of which 18 individuals were grade 3 or worse, and 85 patients had grade 3 or worse neurological complications.[63] Overall response was seen in 81% of patients after a median follow-up of 3·9 months.[61,64] Jacobson and colleagues[62] reported a lower overall response in 67 (71%) of 95 patients infused with axicabtagene ciloleucel, after a median follow-up of 5·6 months.[62,65,66] 95% of the patients had cytokine-release syndrome, of which 17 (16%) patients were grade 3 or worse, whereas neurological complications were reported in 29 (38%) of the treated patients.[65] These real-world experiences extend earlier clinical evidence generated from investigational trials. Further real-world safety and efficacy data on the use of axicabtagene ciloleucel in the USA is expected through the expanded access trial, ZUMA-9 (NCT03153462).

In September, 2019, the European Society for Blood and Marrow Transplantation reported that 155 patients treated with either commercial (80%) or investigational (20%) CAR T cells in 40 centres across nine countries in Europe were registered in their registry.[67] Individual clinical reports on patients receiving CAR T cells in different European countries have also been released. In Germany, of 23 patients who underwent leukapheresis, 20 patients with acute lymphoblastic leukaemia were given tisagenlecleucel, while the remaining 3 patients could not be treated as the manufactured products did not meet the prespecified release criteria.[68] Of these patients, nine (45%) were in remission at the last follow-up visit. The study reported that at a median follow-up of 11 months, the overall survival was 69% and event-free survival was 65%. 17 (74%) of the 23 enrolled patients received tisagenlecleucel either through the expanded access programme (n=6) or as a commercial product (n=11).[68] Grade 4 cytokine-release syndrome was reported in three patients. In Spain, a report released in January, 2019, showed that seven hospitals had treated 84 patients with CAR T cells, out of which only six patients received the product in commercial settings, with the remaining treated in clinical trials.[69] In France, 60 patients with DLBCL were treated with either tisagenlecleucel (n=30) or axicabtagene ciloleucel (n=30) across five centres between April, 2018, and February, 2019, under the temporary authorisation for use programme.[70] Although the actual numbers of treated patients are yet to be disclosed, the uptake of this treatment in Europe has been steady but smaller compared with the USA.

To investigate the activities of specialised treatment centres in adopting CAR T-cell therapies in Europe, a survey study was done between November, 2018, and January, 2019. 566 European Society for Blood and Marrow Transplantation centres were surveyed, of which 134 centres across 22 countries responded.[69] The study showed that 34 centres have already administered CAR T cells to patients, primarily within clinical trials (93% of patients). Furthermore, 57 additional centres located in Europe were planning to administer a CAR T-cell product within the 6 months following the study.[69] In the UK, patients of the National Health Service with acute lymphoblastic leukaemia (children, adolescents, and young adults [up to 25 years old]) can receive CAR T-cell therapy in nine centres and adult patients with DLBCL can receive CAR T-cell products in seven centres, with more centres planning to enrol patients in the future.[71] Although the data indicate a limited number of centres currently available in Europe for commercial CAR T-cell treatments, they also reflect a strong willingness toward the adoption of the therapy.

Ongoing investigations of authorised products in other oncology indications

Tisagenlecleucel and axicabtagene ciloleucel are being investigated for other indications and treatment strategies. A phase 3 trial (OBERON, NCT03628053) is expected to start in late 2019 to further test the efficacy and safety of tisagenlecleucel for the treatment of acute lymphoblastic leukaemia compared with bispecific (blinatumomab) and monoclonal (inotuzumab ozogamicin) antibody-based therapies. Tisagenlecleucel is also being investigated as a treatment for high-risk paediatric acute lymphoblastic leukaemia (positive minimal residual disease at the end of consolidation) in a phase 2 trial (CASSIOPEIA, NCT03876769). Concurrently, axicabtagene ciloleucel is expanding into chronic lymphocytic leukaemia (ZUMA-8, NCT03624036) and acute lymphoblastic leukaemia indications (ZUMA-3, NCT02614066; ZUMA-4, NCT02625480).[51,52] Preliminary results from phase 1 trials were promising,[51,52] and axicabtagene ciloleucel has moved on to phase 2 testing for the treatment of these two conditions (table 3).

In lymphoma, tisagenlecleucel is being tested in combination with pembrolizumab, a PD-1 inhibitor, and with ibrutinib, a BTK inhibitor, in patients with DLBCL (NCT03630159, NCT03876028). The product is also being tested for the treatment of paediatric non-Hodgkin lymphoma (NCT03610724) and relapsed or refractory follicular lymphoma (NCT03568461). In large B-cell lymphoma, axicabtagene ciloleucel is being tested in combination with various anticancer drugs: a PD-1 inhibitor, atezolizumab, with promising results (ZUMA-6, NCT02926833), a 4–1BB agonist (utomilumab; ZUMA-11, NCT03704298), and rituximab or lenalidomide (ZUMA-14, NCT04002401). The developer is also testing axicabtagene ciloleucel as a first-line treatment in high-risk large B-cell lymphoma (ZUMA-12, NCT03761056), and as a treatment for mantle cell lymphoma and indolent non-Hodgkin lymphoma. Data generated from these axicabtagene ciloleucel studies will support the pharmacovigilance plan of this product in Europe.

Search strategy and selection criteria

We obtained the European public assessment reports from the database of the European Medicines Agency website (accessed April 11, 2019). We extracted the manufacturing and product quality, and non-clinical and clinical data into a spreadsheet. When needed, the relevant scientific literature mentioned in the European public assessment reports was also reviewed. Revision of the clinical data packages in the European public assessment reports relied on datasets with the longest possible follow-up time: 12-month update of the ZUMA-1 clinical study of axicabtagene ciloleucel in lymphoma (data cutoff: Aug 11, 2017), the JULIET clinical study of tisagenlecleucel in lymphoma (data cutoff: Dec 8, 2017), and the ELIANA study of tisagenlecleucel in leukaemia (data cutoff: April 25, 2017). To control for investigator bias, we relied on the results reported by the central independent review committee, rather than the results stated by the investigators. Regarding historical comparators, the SCHOLAR-1 study outcomes used as a comparator for both products were extracted from the axicabtagene ciloleucel European public assessment reports. Outcomes of the pooled CORAL extension studies used as a comparator for tisagenlecleucel were not detailed in the European public assessment reports. To reproduce the pooled analysis of the studies, we extracted the data published in scientific literature that were referenced in the European public assessment reports. The population of the pooled studies comprised patients that had relapsed after a second stem cell transplantation (n=75) and patients who did not proceed to stem cell transplantation (n=203). The responses achieved by patients in the clinical studies and historical comparators were then reproduced with the extracted patient numbers. Postauthorisation studies submitted as additional pharmacovigilance activities were extracted from the risk management plan section in the European public assessment reports for each product. To collect the latest published results of these studies, we searched ClinicalTrials.gov, PubMed, Google, the agendas, minutes, and reports of the Committee for Advanced Therapies using the developers and the ClinicalTrials.gov identifiers of the studies (data cutoff: July, 2019).

Conclusion

The two approved CAR T-cell products, tisagenlecleucel and axicabtagene ciloleucel, provided a unique opportunity to explore the effect of choices made by developers during product development on the regulatory evaluation processes. Due to the still undetermined long-term benefits and high price tag, the products face tremendous pressure to have proven long-lasting clinical benefits, particularly when compared with other established treatment options in the market that are more cost-effective, such as haemopoietic stem cell transplantation. The clinical efficacy of the products was identified as the most challenging aspect during development because of the nature of the disease under study, the single-arm study designs, the complex treatment regimens, and the absence of suitable comparators. Both developers were able to implement effective measures to partially mitigate serious adverse events during clinical testing. Further measures were mandated by the regulators in the postmarketing setting to ensure patient safety. The products are being tested for various indications, and more data will further inform their benefit–risk profile. Our analysis suggests that regulatory authorities tend to accept more uncertainty in the evidence generated for CAR T-cell therapies at the time of marketing authorisation submissions compared with small molecules and conventional biologics. Of note, the outlined hurdles and challenges faced by these two products should not discourage more developers from pursuing CAR T-cell therapy development, nor are they intended to call for stricter regulatory assessments. This analysis of the development experiences and regulatory approval processes provide a roadmap to improve the generation of evidence and dossiers for future CAR T-cell therapies, and their integration into routine clinical practice.

Contibutors

ME and MA contributed to the conception and design of the Personal View. ME contributed to data collection and figures, and ME and MA analysed the data. All authors contributed to the literature search, data interpretation, and the writing of the manuscript. MA approved the final manuscript.

Declaration of interests

BLL reports grants and personal fees from Novartis, during the conduct of the work; personal fees from Novartis, Avectas, Brammer Bio,

Incysus, CRC Oncology/Cure Genetics, Novartis, Vycellix, Immuneel, and Ori Biotechand, and equity in Tmunity Therapeutics of which he is a cofounder, outside the submitted work. He has patent methods for the treatment of cancer (US 8906682, US 8916381, US 9101584), patent compositions for the treatment of cancer (US 8911993, US 9102761, US 9102760), a patent method for treating chronic lymphocytic leukaemia (US 9161971), patent compositions and methods for the treatment of cancer (US 9464140, US 9518123, US 9481728, US 9540445), a patent use of CAR-modified T cells to treat cancer (US 9328156, US 9499629), and patent method for assessing the suitability of transduced T cells for administration (US 9572836), with all royalties paid to the University of Pennsylvania. ASW reports advisory board membership and consultation fees from Servier, and grants and consultation fees from Kite Pharma, during the conduct of the work; consultation fees from AbbVie, and grants and consultation fees from Spectrum Pharmaceuticals, outside the submitted work. ME and MA declare no competing interests.

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Supplementary appendix

This appendix formed part of the original submission and has been peer reviewed. We post it as supplied by the authors.

Supplement to: Elsallab M, Levine BL, Wayne AS, Abou-El-Enein M. CAR T-cell product performance in haematological malignancies before and after marketing authorisation. *Lancet Oncol* 2020; **21:** e104–116.

Table 1: Overview of CAR T cell products approved in the EU

	Tisagenlecleucel	Axicabtagene ciloleucel (Axi-cel)
Classification by the European Medicine Agency (EMA)	Advanced Therapy Medicinal Products (ATMP) Gene Therapy Medicinal Product (GTMP)	Advanced Therapy Medicinal Products (ATMP) Gene Therapy medicinal Product (GTMP)
Marketing Authorization Holder (MAH)	Novartis Europharm Limited	Kite Pharma EU B.V.
Academic partners	The University of Pennsylvania, Children's Hospital of Philadelphia (CHOP), United States	National Cancer Institute (NCI), United States
Submission date	November 2017	July 2017
Marketing authorization date	August 27, 2018	August 27, 2018
Duration from submission to Authorization	294 days (9 months, 21 days)	390 days (1 year, 25 days)
Manufacturing sites	Germany, United States	United States
Europe batch release	Novartis Pharma GmbH, Germany	Lonza Netherlands B.V. Kite Pharma EU B.V., Netherlands
Indication	Acute lymphoblastic leukaemia (ALL) Treatment of pediatric and young adult patients up to 25 years of age with B-cell acute lymphoblastic leukaemia (ALL) that is refractory, in relapse post-transplant or second or later relapse.	Large B cell lymphomas Treatment of adult patients with relapsed or refractory diffuse large B-cell lymphoma (DLBCL) and primary mediastinal large B-cell lymphoma after two or more lines of systemic therapy.
	Diffuse large B cell lymphoma (DLBCL) Treatment of adult patients with relapsed or refractory diffuse large B-cell lymphoma (DLBCL) after two or more lines of systemic therapy.	
Price	475,000 US dollars (ALL) 373,000 US dollars (DLBCL)	373,000 US dollars
PRIME scheme	Yes	Yes
Accelerated Assessment procedure	Yes, reverted to a standard timetable on May 2018	Yes, reverted to a standard timetable December 2017

Table 2: Summary of CAR T cell design in each product

	Tisagenlecleucel	Axicabtagene ciloleucel (axi-cel)
viral vector		
Manufacturer	Oxford BioMedica	Contract manufacturing organisation (CMO)
Vector type	Lentiviral vector	Gamma retroviral vector
Vector origin	HIV-1	Murine stem cell virus (MSCV) with gibbon ape leukaemia virus (GaLV) envelope
Self-inactivation	Yes	No
Integration into the host genome	Yes	Yes
Manufacturing	Upstream process: - Thawing the working cell bank (WCB) - Expansion of the production cell bank - plasmid transfection - induction and harvest Downstream purification process: - filtration, - Chromatography - Nuclease treatment	Stably transduced PG13 (ATCC CRL-10686™) cell line. - Expanded cells from a working cell bank (WCB) - Culture supernatant is harvested, filtered, and filled into cryostorage bags.
Transduction capabilities	Non-dividing and dividing cells	Dividing cells only
CAR construct		
CAR construct	Anti CD19, 4-1BB, CD3 zeta	Anti CD19, CD28, CD3 zeta
CAR generation	Second generation	Second generation
Promoter	EF-1alpha	5'LTR (include the promoter)
ScFv	Anti CD19	Anti CD19
ScFv source	FMC63 mouse hybridoma	FMC63 mouse hybridoma
Hinge and Transmembrane region	CD8 derived	CD28 derived
Cytoplasmic region	4-1BB (CD137), CD3 zeta	CD28, CD3 zeta

Table 3: Patient characteristics from different populations of the pivotal CAR T cells' trials and historical controls

	JULIET (ITT) (165) (Enrolled)	JULIET (FAS)* (111) (infused)	ZUMA-1 (ITT) (111)	ZUMA-1 (mITT) (101)	SCHOLAR 1
Median Age (range)	59 (22-76)	56 (22-76)	58 (23-76)	58 (23-76)	55 (19-81)
Male %	62%	61%	69%	67%	64%
Disease Type					
DLBCL	77%	79%	NA	76%	87%
PMBCL	NA	NA	NA	8%	2%
TFL	21%	19%	NA	16%	4%
Other	2%	2%	NA	-	1%
ECOG PS, %					
0	47%	55%	41%	42%	-
1	53%	45%	59%	58%	(0+1) 73%
2-4	-	-	-	-	14%
Missing					13%
Disease stage					
I-II	22%	24%	15%	15%	27%
III-IV	78%	76%	85%	85%	72%
Missing	-	-	-	-	<1%
IPI risk classification, %					
<2 risk factors	21%	28%	NA	27%	25%
≥2 risk factors	79%	72%	NA	73 %	57%
Missing	-	-	-	-	18%
Refractory category					
Primary refractory	NA	NA	3%	2%	28%
Refractory to ≥ second-line therapy	NA	NA	77%	77%	50%
Relapsed ≤ 12 months post ASCT	NA	NA	20%	21%	22%
Total no. of lines of chemotherapy and ASCT received, %			Median no. 3	Median no. 3	
1	4%	5%	NA	2%	28%
2	44%	44%	NA	29%	49%
3	31%	31%	NA	30%	<1%
≥4	21%	20%	NA	39%	-

*Full analysis set (FAS): include all the infused patients, Efficacy analysis set (EAS): include infused patients who were at least followed for three months.

ASCT: autologous stem cell transplantation. DLBCL: diffuse large B-cell lymphoma. ECOG PS: Eastern Cooperative Oncology Group performance status. IPI: International Prognostic Index. PMBCL: primary mediastinal large B-cell lymphoma. TFL: transformed follicular lymphoma.

Table 4: Efficacy results of the pivotal CAR T cells' trials using different analysis populations and historical controls

	JULIET (ITT)	JULIET (mITT) (EAS)*	ZUMA-1 (ITT)	ZUMA-1 (mITT)	SCHOLAR-1	CORAL extension studies
sample	165	93	111	101	523	278
RR n	56	48	73	73	135	112
CR n	40	37	52	52	45	79
PRn	16	11	21	21	90	33
RR % (95% CI)	33.9 (26.8-41.7)	51.6 (41-62,1)	65.8 (56.1-74.5)	72.2 (62.5-80.7)	25.7 (20.9-31.3)	40,3 (34.5-46.3)
CR % (95% CI)	24.2 (17.9-31.5)	39.8 (29,8-50)	46.9 (37.3-56.6)	51.5 (41.3 61.6)	7.0 (3.2-14.5)	28.4 (23.2-34.1)
PR % (95% CI)	9.7 (5.6-15.3)	11.8 (0.06-20.1)	18.9 (12.1-27.5)	20,8 (13.4-30)	17.5 (13.3-22.7)	11.9 (8.3-16.3)

Full analysis set (FAS): include all the infused patients, Efficacy analysis set (EAS): include infused patients who were at least followed for three months.
CR: complete response. ITT: intention to treat. mITT: modified intention to treat. PR: partial response. RR: response rate.

Table 5: Safety profiles and adverse reactions from clinical trials of tisagenlecleucel and axi-cel

	Tisagenlecleucel		Axicabtagene ciloleucel (axi-cel)
Indication (patient numbers)	ALL (N=104)	DLBCL (N=99)	DLBCL, PMLBCL, or TFL (N=108)
Adverse Events	100%	100%	100%
Serious Adverse events	78%	65%	55%
Death	- Four deaths within 30 days of infusion - 25 deaths after 30 days of infusion - 22/29 due to disease progression	- Three deaths within 30 days post-infusion - 47 after 30 days of infusion - 45/50 due to disease progression	- Two within 30 days post-infusion - 42 after 30 days of infusion - 37/44 due to disease progression
Cytokine release syndrome	81%	58%	93%
Neurological events	37.5%	21%	66%
Tumour lysis syndrome	4%	1%	1%
Infections	68%	53%	38%
Febrile neutropenia	35%	13%	35%

ALL: Acute lymphoblastic leukaemia. DLBCL: diffuse large B-cell lymphoma. PMBCL: primary mediastinal large B-cell lymphoma. TFL: transformed follicular lymphoma.

3.2. Publication 2

3.2.1. Journal Summary List

Journal Data Filtered By: **Selected JCR Year: 2018** Selected Editions: SCIE,SSCI
Selected Categories: **"MEDICINE, RESEARCH and EXPERIMENTAL"**
Selected Category Scheme: WoS
Gesamtanzahl: 136 Journale

Rank	Full Journal Title	Total Cites	Journal Impact Factor	Eigenfactor Score
1	NATURE MEDICINE	79,243	30.641	0.162840
2	Science Translational Medicine	30,485	17.161	0.121980
3	JOURNAL OF CLINICAL INVESTIGATION	108,879	12.282	0.139970
4	TRENDS IN MOLECULAR MEDICINE	9,946	11.028	0.018900
5	JOURNAL OF EXPERIMENTAL MEDICINE	63,983	10.892	0.071790
6	EMBO Molecular Medicine	7,507	10.624	0.025980
7	Annual Review of Medicine	6,068	10.091	0.009030
8	MOLECULAR THERAPY	16,991	8.402	0.030050
9	MOLECULAR ASPECTS OF MEDICINE	5,568	8.313	0.009020
10	Theranostics	8,769	8.063	0.020270
11	EBioMedicine	5,401	6.680	0.022310
12	ALTEX-Alternatives to Animal Experimentation	1,361	6.183	0.001920
13	Wiley Interdisciplinary Reviews-Nanomedicine and Nanobiotechnology	2,345	6.140	0.004130
14	JCI Insight	4,351	6.014	0.020440
15	Molecular Therapy-Nucleic Acids	3,189	5.919	0.010410
16	Molecular Therapy-Oncolytics	486	5.710	0.001990
17	Nanomedicine-Nanotechnology Biology and Medicine	10,131	5.570	0.014480
18	Cold Spring Harbor Perspectives in Medicine	6,223	5.564	0.016730
19	CLINICAL SCIENCE	10,951	5.237	0.014190
20	JOURNAL OF BIOMEDICAL SCIENCE	4,083	5.203	0.006300

Rank	Full Journal Title	Total Cites	Journal Impact Factor	Eigenfactor Score
21	npj Vaccines	282	5.020	0.001120
22	AMYLOID-JOURNAL OF PROTEIN FOLDING DISORDERS	1,335	4.919	0.003270
23	Translational Research	3,669	4.915	0.008530
24	Molecular Therapy-Methods & Clinical Development	1,078	4.875	0.004020
25	Vaccines	1,077	4.760	0.003910
26	JOURNAL OF MOLECULAR MEDICINE-JMM	7,195	4.746	0.010880
27	EXPERIMENTAL AND MOLECULAR MEDICINE	4,046	4.743	0.007380
28	Stem Cell Reviews and Reports	2,436	4.697	0.004690
29	CANCER GENE THERAPY	2,842	4.681	0.003200
30	EPMA Journal	815	4.661	0.001320
31	JOURNAL OF CELLULAR AND MOLECULAR MEDICINE	12,391	4.658	0.015760
32	Stem Cell Research & Therapy	6,132	4.627	0.015810
33	Cancer Biology & Medicine	1,043	4.467	0.003040
34	EXPERT REVIEWS IN MOLECULAR MEDICINE	1,758	4.407	0.001450
35	mAbs	4,415	4.405	0.011150
36	MOLECULAR PHARMACEUTICS	16,792	4.396	0.028020
37	CYTOTHERAPY	5,969	4.297	0.009690
38	JOURNAL OF INHERITED METABOLIC DISEASE	5,868	4.287	0.008410
39	PPAR Research	1,434	4.186	0.001600
40	ARCHIVES OF PATHOLOGY & LABORATORY MEDICINE	10,039	4.151	0.012620
41	Journal of Translational Medicine	10,831	4.098	0.022910
42	CTS-Clinical and Translational Science	1,351	3.989	0.003190

2

Selected JCR Year: 2018; Selected Categories: "MEDICINE, RESEARCH and EXPERIMENTAL"

3.2.2. Publication: "Mitigating Deficiencies in Evidence during Regulatory Assessments of Advanced Therapies: A Comparative Study with Other Biologicals"

- **Title:**

  Mitigating Deficiencies in Evidence during Regulatory Assessments of Advanced Therapies: A Comparative Study with Other Biologicals

- **Authors:**

  Magdi Elsallab; Christopher A. Bravery; Andreas Kurtz; Mohamed Abou-El-Enein

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Molecular Therapy
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Original Article

Mitigating Deficiencies in Evidence during Regulatory Assessments of Advanced Therapies: A Comparative Study with Other Biologicals

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Advanced therapy medicinal products (ATMPs) comprising cell therapy, gene therapy, and tissue-engineered products, offer a multitude of novel therapeutic approaches to a wide range of severe and debilitating diseases. To date, several advanced therapies have received marketing authorization for a variety of indications. However, some products showed disappointing market performance, leading to their withdrawal. The available evidence for quality, safety, and efficacy at product launch can play a crucial role in their market success. To evaluate the sufficiency of evidence in submissions of advanced therapies for marketing authorization and to benchmark them against more established biological products, we conducted a matched comparison of the regulatory submissions between ATMPs and other biologicals. We applied a quantitative assessment of the regulatory objections and divergence from the expected data requirements as indicators of sufficiency of evidence and regulatory flexibility, respectively. Our results demonstrated that product manufacturing was challenging regardless of the product type. Advanced therapies displayed critical deficiencies in the submitted clinical data. The submitted non-clinical data packages benefited the most from regulatory flexibility. Additionally, ATMP developers need to comply with more commitments in the post-approval phase, which might add pressure on market performance. Mitigating such observed deficiencies in future product development, may leverage their potential for market success.

INTRODUCTION

The pharmaceutical industry is shifting focus toward disease areas with high unmet medical needs such as oncology and rare diseases.[1] Advancements in biotechnology have enabled such a shift by introducing novel therapeutic approaches, particularly cell therapies, gene therapies, and tissue-engineered products, known in the European Union (EU) as advanced therapy medicinal products (ATMPs).[2] To date, 14 ATMPs have received marketing authorization (MA) in the EU; however, 5 have subsequently been withdrawn from the market. Most recently, Zalmoxis was withdrawn in October 2019 after unfavorable results reported from the post-approval phase III clinical trial,[3] a requirement for conditional MA, which was obtained in 2016. Reimbursement and commercial issues, limited market demand and manufacturing problems contributed to the other withdrawals.[4,5] It is expected that pharmaceutical development programs generate safety and efficacy evidence that is not only sufficient to support MA decisions but also decisions made by health technology assessment (HTA) agencies and other relevant stakeholders.[6–9] However, such alarming numbers of withdrawals can indicate that there is a gap between the evidence presented for MA and the evidence deemed sufficient for market and patient access.

ATMPs are also biological medicinal products,[10] a family of products extracted from or manufactured from biological sources. These products include monoclonal antibodies, enzymes, and hormones, the majority of which are produced by recombinant DNA technologies (hereafter referred to as other biologicals). After 30 years of experience with recombinant proteins, their development path has become well established.[11] In contrast, ATMPs are a more diverse group of products, often with little in common with each other, and many of them are a poor fit for existing development and business models. This situation challenges developers to identify an appropriate development strategy and determine how much evidence is needed to increase the probability of success in acquiring MA and achieving commercial viability.[12]

The expected evidence that should be collected on a therapeutic candidate during its development for inclusion in a MA application (MAA) is laid down in Annex I of Directive 2001/83/EC (hereafter referred to as data requirements). Sections for specific types of therapeutics, such as ATMPs, are provided in the Annex to acknowledge the complexity of these products and guide developers on how to comply with additional requirements, whenever applicable. Moreover, to emphasize the need for flexibility when developing and testing ATMPs, which are very diverse in nature, Annex I encourages the use of a risk-based approach.[10,13] Such risk analysis can be conducted by the applicant to determine the extent of quality, non-clinical, and clinical evidence to be

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Table 1. Basic Characteristics of the Matched Cohorts

		ATMPs/ Total (N = 22)	ATMPs/ Matched (n = 17)	Other Biologicals/ Matched (n = 17)
MAA outcome (%)	authorized	14 (64)	12 (71)	12 (71)
	failed (refused and/or withdrawn)	8 (36)	5 (29)	5 (29)
MA type (%)	full authorization	10 (45)	10 (59)	10 (59)
	conditional marketing authorization	3 (14)	1 (6)	1 (6)
	marketing authorization under exceptional circumstances	1 (5)	1 (6)	1 (6)
	withdrawn (pre-approval)[a]	7 (32)	4 (24)	4 (24)
	refused	1 (5)	1 (6)	1 (6)
Orphan designation (%)		13 (60)	11 (65)	11 (65)
Disease area (%)	non-hematological malignant neoplasms	7 (32)	5 (29)	5 (29)
	musculoskeletal diseases	4 (18)	4 (24)	4 (24)
	hematological malignant neoplasms	3 (14)	3 (18)	3 (18)
	endocrine, nutritional, and metabolic diseases	2 (9)	2 (12)	2 (12)
	digestive system diseases	1 (5)	1 (6)	1 (6)
	eye diseases	3 (14)	1 (6)	1 (6)
	diseases of blood, blood-forming organs, and certain immune disorders	2 (9)	1 (6)	1 (6)

MAA, marketing authorization application; MA, marketing authorization.
[a]Withdrawn refers to the withdrawal of the marketing authorization application before issuing a final opinion from the Committee for Medicinal Products for Human Use (CHMP).

included in the MAA, and to provide scientific justification when deviating from the requirements of this Annex (hereafter referred to as divergence).[10] However, the degree of divergence of ATMPs from the expectations in Annex I and its effect on the sufficiency of the evidence and ability to reach a conclusion on the overall risks and benefits of the product have not been thoroughly investigated.

Previous studies have attempted to investigate the evidence in ATMP submissions through the quantification of objections raised by regulatory authorities during the assessment procedure of MAAs.[14–17] de Wilde et al.[14] and Carvalho et al.[15] relied on the European public assessment report (EPAR), a document published by the European Medicines Agency (EMA) for all submissions that reach the first stage of assessment, whether approved, refused, or withdrawn. Barkholt et al.[16] at the EMA quantified the objections for the first 20 MAAs for ATMPs. The study by de Wilde et al.[14] showed considerable discrepancies in the results compared to the other two studies[15,16] that performed a more thorough analysis, with Barkholt et al. deemed to be the most reliable data source, as they relied on internal EMA data.[16] Nevertheless, to benchmark the sufficiency of submitted evidence for ATMPs, a comparison with more established biological products is needed, as suggested by Bravery et al.[17] This approach can help ATMP developers mitigate deficiencies in evidence by identifying the weaknesses in existing submissions and understanding the impact on post-approval commitments and performance. To our knowledge, no existing research has attempted to assess the sufficiency of evidence presented for ATMPs in MAA submissions against other biologicals, by not only the quantification of objections, but also by identifying areas of regulatory flexibility, where applicants diverged from data requirements in Annex I.

In this study, we conducted a retrospective, head-to-head, nearest neighbor matched comparison of submitted evidence between ATMPs and other biologicals using data extracted from the EPARs. We accounted for several confounding factors that may impact the extent and the source of the evidence expected in the MAA by matching them in both groups. The data requirements provided in Directive 2001/83/EC, Annex I, were clustered into four evidence domains: the manufacturing and quality testing domain, the experimental design and conduct of studies domain, the efficacy and mode of action (MoA) domain, and the safety and toxicity domain. We then employed the quantitative assessment of the objections and divergence in each domain as indicators of evidence sufficiency and compared them between both groups. The differences in the timing of addressing the detected objections between the authorized cohorts were then explored. Finally, we investigated the possible reasons for the observed differences in evidence sufficiency.

RESULTS

Retrieval and Characteristics of ATMP Submissions

Screening of 1,604 submissions (data cutoff, July 1, 2019) in the EMA databases (authorized or refused submissions, 1,382; withdrawn submissions, 222) identified 22 ATMP submissions (Tables 1 and S1). Out of the 22 submissions, 12 were for gene therapy products (55%, including genetically modified cells), 6 were for tissue-engineered products (27%), and 4 were for somatic cell therapy products (18%). Products that contained autologous cells were 11/22 (50%), while 3/22 products contained allogeneic cells (14%). The first submission was for Cerepro in 2005, while the last identified submission was in 2018 for Zynteglo. The average number of ATMP submissions per year was 1.6 (standard deviation [SD], 0.9; range, 0–3). MA was granted to 14/22 submissions (Table 1), 10 of which were full MA (72%), 3 were conditional MA (CMA) (21%), while 1 (Glybera) was authorized under exceptional circumstances (7%). 21/22 (95%) EPARs were available since one product (Raligize) was withdrawn before the end of the first stage of evaluation (day 120), meaning that no EPAR was released. Out of the 14 approved ATMPs, 5 have been subsequently withdrawn. The screening of the EMA databases and selection of the ATMP submissions is depicted in (Figure S1).

Retrieval and Characteristics of the Matched Biological Products

The same EMA databases were screened to identify suitable matches to ATMPs from other biologicals. In total, 17/21 (81%) ATMPs were

Table 2. Matched Comparison of Objections between ATMP and Biologicals Submissions

Evidence Domain	Differences in Objections between Successful ATMPs and Biologicals Submissions (n = 24)		Differences in Objections between Failed ATMPs and Biologicals Submissions (n = 10)	
	Z	p (Two-Tailed)	Z	p (Two-Tailed)
Manufacturing and quality	−1.380	0.186	−0.674	0.625
Experimental design and conduct of the studies	−2.221	0.021*	−0.674	0.625
Efficacy and MoA	−2.108	0.031*	−0.137	1
Safety and toxicity	−0.431	0.727	−0.552	0.750
Total number of objections	−2.396	0.013*	−0.674	0.625

*p < 0.05. p values were determined by a Wilcoxon signed-rank test.

Table 3. Matched Comparison of Divergence between ATMPs and Biologicals Submissions

Evidence Domains	Differences in the Divergence between Authorized ATMPs and Biologicals Submissions (n = 24)		Differences in the Divergence between Failed ATMPs and Biologicals Submissions (n = 10)	
	Z	p (Two-Tailed)	Z	p (Two-Tailed)
Experimental design and conduct of the studies	−2.081	0.063	0	1.000
Efficacy and MoA	−3.070	0.0001*	−1.633	0.188
Safety and toxicity	−2.669	0.006*	−1.214	0.313
Total number of divergence	−3.063	0.0001*	−1.483	0.188

*p < 0.05. p values were determined by a Wilcoxon signed-rank test.

matched to other biologicals submissions (Tables 1 and S2) and compared statistically for objections and divergence. In the authorized ATMP cohort, 12/14 (86%) ATMPs were matched to other authorized biologicals. Two products (Zynteglo and Holoclar) could not be matched, as they received a CMA, and biological products with a CMA in the same disease areas (blood diseases and eye diseases, respectively) could not be identified. In the failed authorization cohort, 5/7 (71%) ATMPs were matched. Contusugene Ladenovec Gendux (CLG) and OraNera could not be matched due to the unavailability of other withdrawn biological products for eye diseases and non-hematological malignancies (not orphan), respectively. Of the 17 matched biologicals, 16 were recombinant products (94%), while the remaining product (Oncophage) was an autologous tumor-derived protein-peptide complex (6%). The 16 recombinant products, comprised, nine monoclonal antibodies (56%), three enzymes (19%), three hormones, cytokines, or growth factors (19%) and one coagulation factor (6%). Out of the 12 approved matched biologicals, only 1 has been subsequently withdrawn. The matching characteristics of the ATMPs and the other biologicals are summarized in Table 1.

To examine whether each ATMP and matched biological underwent the regulatory evaluation at a close time frame, the duration between the dates of the regulatory decisions (authorization, withdrawal, or rejection) for each matched pair was calculated. In the authorized cohorts, the average duration between the date of authorization of matched pairs was 15.6 months (SD, 21.8 months; range, 0–67). In the failed cohorts, the average duration between the withdrawal or rejection date of matched pairs was 41.4 months (SD, 30.9 months; range, 11–86).

Comparing ATMP Regulatory Submissions to Matched Biologicals

The available information in the EPARs on the objections raised on the submitted evidence was then extracted and sorted according to the corresponding evidence domains as defined (Table S3). When comparing the authorized matched paired products (n = 24), the total number of the identified objections in the EPARs of the ATMPs was significantly higher (p = 0.013) (Table 2; Figure S2). When comparing the objections in each evidence domain, objections in the experimental design and conduct of studies domain were significantly higher in authorized ATMPs (p = 0.021). Furthermore, a greater number of objections were raised on the evidence of efficacy and MoA in authorized ATMPs (p = 0.031) (Table 2; Figure S2). In contrast, no significant differences were observed in the product manufacturing and quality domain (p = 0.186) or issues related to product safety (p = 0.727) (Table 2; Figure S2). For the failed submissions (withdrawn or rejected, n = 10), no statistically significant differences were found in either the total number of objections or within any of the four domains (Table 2; Figure S3).

The impact of the regulatory flexibility on the evidence was evaluated by estimating the degree of divergence from the data requirements and then comparing them between groups. This was achieved by quantifying the studies that were not submitted in the application, as stated in the EPARs. When comparing the authorized cohorts, in total, significantly more divergence was detected in the EPARs of the ATMPs as compared to the other biologicals (p = 0.0001) (Table 3; Figure S4). Divergence in authorized ATMPs was significantly higher than in other biologicals, in the safety and toxicity evidence domain (p = 0.006), as well as in the clinical efficacy and MoA domain (p = 0.0001) (Table 3; Figure S4). Despite the application of more novel technologies and methods for ATMP manufacture and testing as compared to other biologicals, no divergence from the data requirements was detected in this domain. Additionally, no significant difference in divergence was found in the experimental design and conduct of studies evidence (p = 0.063), despite being greater in authorized matched ATMPs than in matched biologicals (Z = −2.081) (Table 3; Figure S4). No statistically significant differences were observed between the failed authorization cohorts (n = 10) (Table 3; Figure S5).

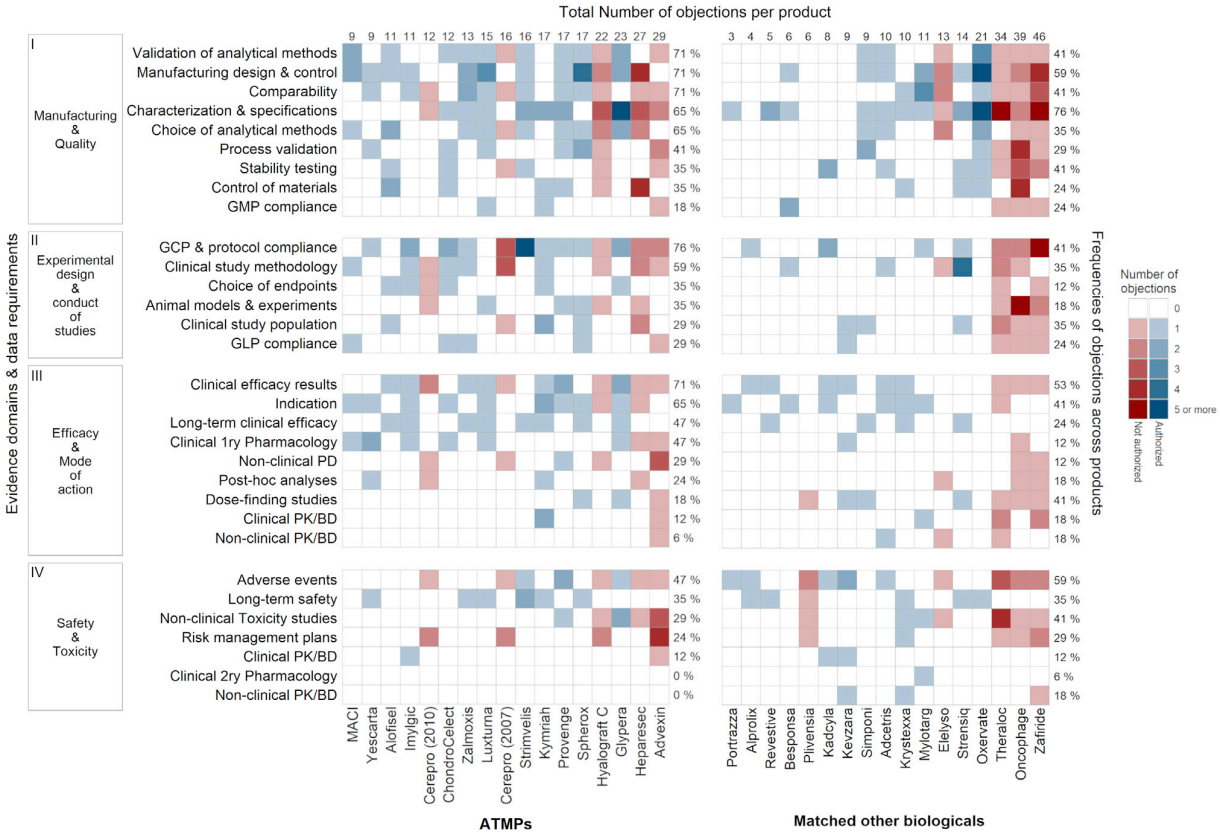

Figure 1. Heatmaps for the Distribution and Number of Objections among the Advanced Therapy Medicinal Products (ATMPs) and Matched Other Biologicals

The data requirements were clustered into four evidence domains (left y axis). The objections were then identified from the European public assessment reports (EPARs) and sorted to the relevant data requirement. The data requirements are arranged (top-downward) in each domain according to the frequency of objections in ATMP submissions. The total number of objections identified in each EPAR is shown on the top x axis. The frequency of objections and concerns across the products in each data requirement is shown on the right y axis of each heatmap.

Distribution of Objections across ATMPs Compared to Matched Biologicals

The distribution of the objections among the products and evidence domains revealed a clear heterogeneity in the distribution within the ATMP cohort (Figure 1). Most of the objections in both groups were concentrated in the manufacturing and quality domain, followed by the experimental design, and then the efficacy and safety domains. The spread of the objections across the products was greater in the ATMPs for most of the data requirements (Figure 1).

The most commonly identified objections in ATMP submissions were on compliance with good clinical practice (GCP) and clinical trial protocols (Figure 1, domain II, row 1). Such objections were due to substantial changes in the trial protocols, inadequate documentation of studies, and GCP non-compliance. These issues were not detected as frequently in the EPARs of the other biologicals (Figure 1, domain II, row 1). Another common objection for ATMPs was related to the efficacy results of the main clinical studies (Figure 1, domain III, row 1). Out of the 12 ATMPs with such detected objections, 7 were successful submissions.

Objections in the manufacturing and quality domain were mostly related to validation of the analytical methods, design and control of the manufacturing process, and comparability (Figure 1, domain I, rows 1–3). Most objections in the design and control of the manufacturing of ATMPs were due to deficiencies in microbiological control (8/12, 67% of the products). Other notable manufacturing objections were related to the choice and justification of the analytical methods (Figure 1, domain I, row 5). The most frequent reason for these objections was the choice of the potency assays (8/11, 73%). Objections around characterization and specifications of ATMPs were also common; however, they were slightly more common in other biologicals (Figure 1, category I, row 4). Safety-related objections were not common and were closely similar in both cohorts.

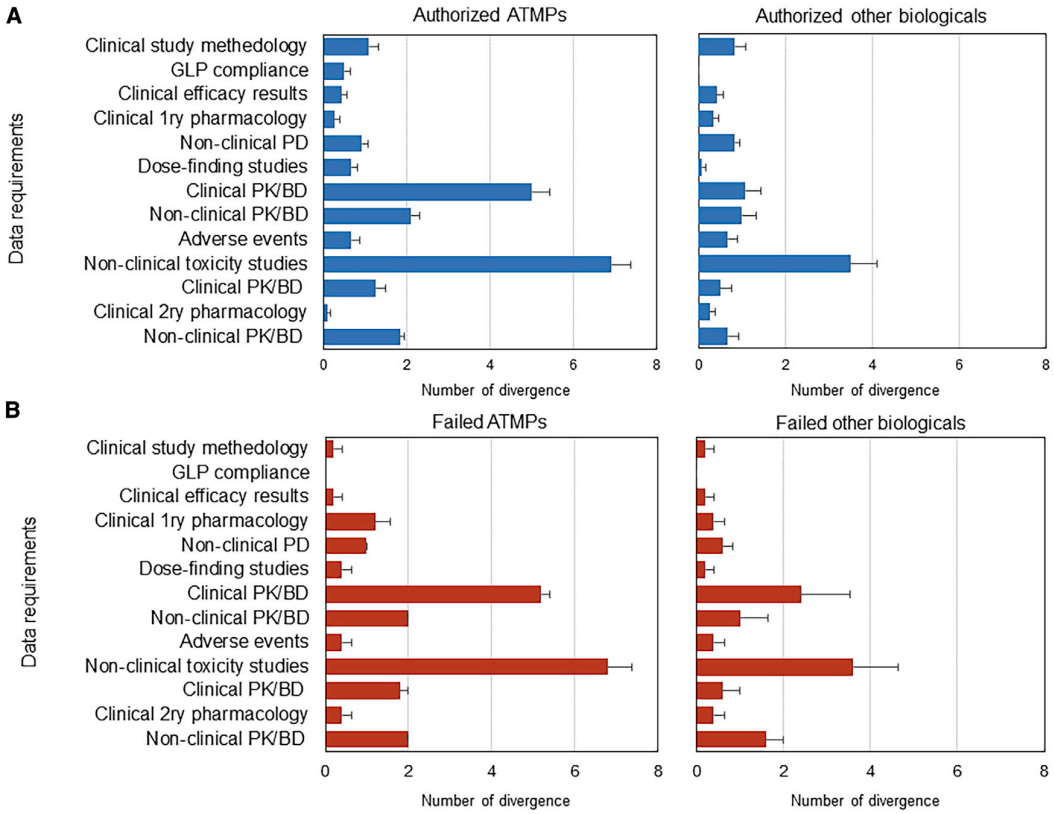

Figure 2. Average Numbers of Divergences in Each Data Requirement per Submission across Authorized and Failed ATMPs and Matched Other Biologicals
Divergence from the regulatory data requirements for marketing authorization applications laid down in Annex I of Directive 2001/83/EC was assessed through the quantification of omitted studies in the EPARs. Regardless of the approval status, differences in divergence are evident in the non-clinical toxicity studies and clinical pharmacokinetics and biodistribution (PK/BD) studies between ATMPs and other matched biologicals. Error bars represent the standard error of the mean (SEM). (A) Authorized ATMPs and matched other biologicals (Blue). (B) Failed ATMPs and matched other biologicals (Red).

Main Points of Divergence in ATMP Submissions Compared to Other Biologicals

Sources of divergence were primarily identified in non-clinical studies and, to a lesser degree, in clinical studies (Figure 2). The inability to undertake *in vivo* toxicity studies such as toxicokinetics, reproduction toxicity, local tolerance, and, in some cases, carcinogenicity studies in the ATMP safety and toxicity domain led to a greater number of divergences (Figure 2). Moreover, a full understanding of MoA was not achievable by conducting animal studies, particularly in cell-based product submissions. Difficulties in the application of good laboratory practice (GLP) principles in non-clinical studies of ATMPs has led to the acceptance of non-compliant studies in the submissions, a divergence not seen with other biologicals (Figure 2).

The absence of pharmacokinetics/biodistribution studies in human subjects (Figure 2) resulted in a significantly higher number of divergences for ATMPs (especially those approved). Absorption, distribution, metabolism, and excretion studies are not expected to be conducted in the case of ATMPs, but other studies such as target organ distribution, migration, and persistence were not conducted in human subjects for some of the products. In those cases, the study was not technically possible, and the available non-clinical evidence was considered sufficient. Furthermore, for only 6/17 (35%) of ATMPs, dose-escalation studies were conducted, while for 15/17 (88%) of other biologicals, traditional dose-escalation studies were carried out.

Differences in Solving the Raised Objections between the Matched Cohorts

Raised regulatory objections can be solved during the MAA procedure with the submission of new data, additional analysis, additional risk minimization measures, or modifications of the summary of product characteristics. Where such solutions are not possible during the procedure and the issue does not preclude approval, applicants can be asked to commit to solving the outstanding issues after approval through submission of more data on the quality, safety, or efficacy of the product. When comparing the approaches to address outstanding objections in successful applications, post-approval commitments were more frequent for ATMP submissions than for

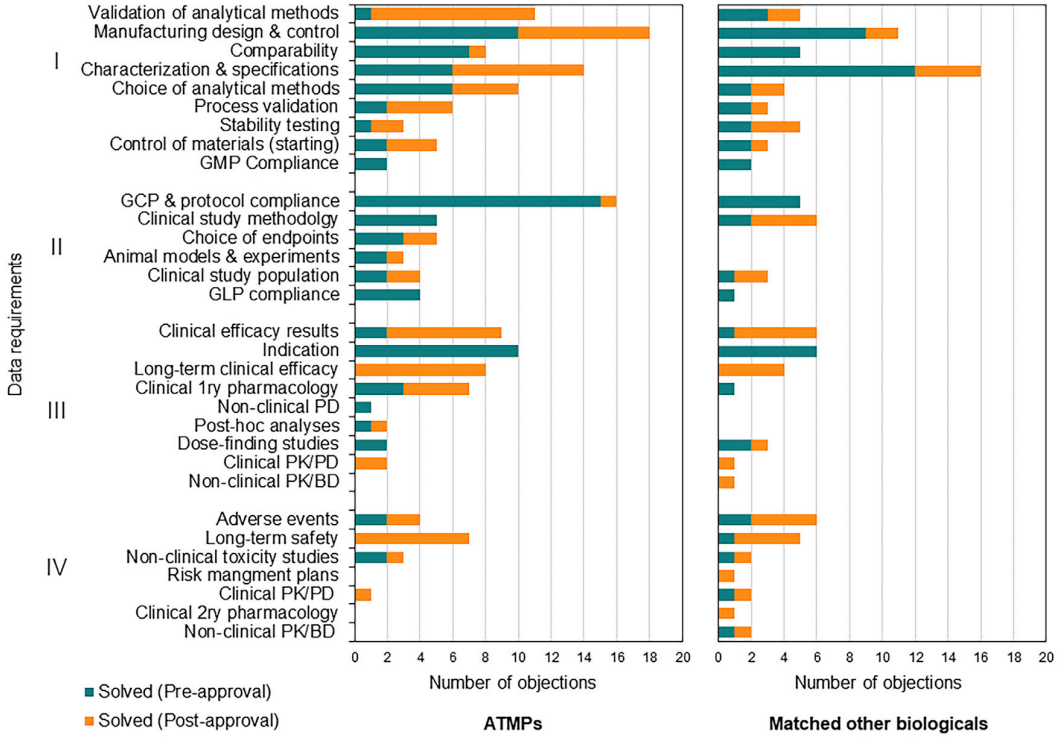

Figure 3. Differences in When Regulatory Objections Were Addressed between ATMPs and Matched Other Biologicals

Each solved objection was categorized as solved either in the pre-approval or the post-approval stage based on the information in the EPARs. Note the difference between both cohorts in quality data requirements (top of the chart). Note also the categories of long-term safety and efficacy as well as the clinical efficacy results that were addressed more in the case of ATMPs through post-approval approaches. (I) manufacturing and quality testing domain (II) experimental design and conduct of studies domain (III) efficacy and mode of action domain (IV) safety and toxicity domain.

other biologicals (Figure 3). Further analysis showed that more manufacturing and quality objections for ATMPs were mentioned in the EPAR to be addressed in the post-approval phase as compared to other biologicals (Figure 3). These objections were mostly related to validations of the analytical methods, improving process control, developing new analytical methods, performing further characterization, and tightening of the proposed specifications.

Furthermore, developers of ATMPs committed to more post-approval approaches to address issues related to the pivotal trial results, long-term efficacy and long-term safety, as compared to biologicals (Figure 3). These approaches mainly included the obligation to perform post-authorization safety studies (PASSs) and post-authorization efficacy studies (PAESs) (Figure 3). Additionally, ATMP developers were obliged to collect specific safety and efficacy information through the use of patient registries.

Other Factors Influencing the Sufficiency of Evidence

Possible differences in the development strategy in both cohorts were explored. The nature of the organization that developed the product was considered and divided into two categories: established large biopharma and micro, small, and medium-sized enterprises (SMEs). The use of scientific advice is reported in the EPAR, so those data were also collected. Most of the ATMP submissions came from SMEs, with only 4/17 (24%) of ATMP submissions from large companies, as compared to 15/17 (88%) for other biologicals. Despite ATMPs being more complex products that may require regulatory advice at several stages of development, EMA scientific advice was sought at nearly equal frequency. On average, developers of authorized ATMPs sought EMA scientific advice 3.0 times (SD, 1.3; range, 1–5), while the developers of the other approved biologicals sought scientific advice 3.1 times (SD, 2.0; range, 0–7).

The main clinical studies utilized for the benefit-risk assessment also showed significant differences between the matched authorized cohorts. Single-arm trials were more frequent among authorized ATMPs, with controlled trials being conducted in only 7/12 (58%) of the authorized ATMPs, as compared to 10/12 (83%) of the other biologicals. Furthermore, there was a significant difference in the number of patients in the main clinical trials of the authorized ATMPs, as compared to the other biologicals ($Z = -2.510$, $p = 0.009$). On average, authorized ATMP main clinical trials had 158

patients per clinical trial (SD, 160; range, 12–512), while the other biologicals had an average of 434 patients per clinical trial (SD, 431; range, 13–1,197,). Finally, all authorized other biological trials were multicenter trials, while two ATMPs (Glybera and Strimvelis) were single-center trials. Despite not included in the analysis, one authorized ATMP (Holoclar) used a historic clinical case series as the main study for the MA instead of designing and conducting a clinical trial.

DISCUSSION

ATMPs are a new and more complex group of therapeutic products with a wide range of development challenges. To acknowledge the complexity and novelty of ATMPs, the EU medicines directive (Directive 2001/83/EC) provides some specific requirements for their development in Annex I. Previous studies have explored the reasons for the success and failure of ATMPs by evaluating the objections, duration of review, and outcomes against other factors such as orphan status, company size, and use of scientific advice.[18–22] None to date has tried to evaluate the more subtle question of whether the data provided were consistent with Annex I and whether a risk-based approach was used and, more importantly, accepted. The backdrop to this question was the number of ATMPs withdrawn after approval, reaching a staggering 36% (5/14). Those five products had been approved for an average of 3.60 years (SD, 2.30; range, 1.40–6.82), leaving only three ATMPs that were approved for more than 3 years: Holoclar (longest at 4.37 years), Imlygic, and Strimvelis. Given the small numbers of ATMP submissions, a comparator group was needed to benchmark the performance of ATMPs against more established biological products (other biologicals). We were able to match 17 ATMPs to other biological products based on known confounding factors, thus minimizing potential bias in the comparison (Table 2). Our objectives were as follows: (1) to investigate the sufficiency of evidence through the quantification of objections raised by regulatory authorities, (2) to measure regulatory flexibility where applicants diverged from data requirements in Annex I, and (3) to assess whether any identified weaknesses have post-approval implications.

First, we acknowledge the limitations of this analysis. The only public sources of information available are the EPARs; these are edited versions of the EMA internal assessment reports, with confidential details removed, primarily in the manufacturing and quality section.[23] Moreover, some of the solved issues may have been removed from the final reports, leading to a potential underestimation of the objections raised during the evaluation. Furthermore, the EPAR format has been updated to address the needs of HTAs between 2012 and 2015.[24] Nevertheless, these limitations were addressed by applying a strict text mining and analysis framework and matching ATMPs and biologicals on the date of the regulatory decision, respectively.

We scored the objections raised during the regulatory assessments of MAA submissions for both ATMPs and other biologicals (Figure 1) and sorted them to the predefined evidence domains. Even though the manufacturing and quality evidence domain had the highest proportion of objections in both groups, as reported by others,[14–16,25,26] there were no significant differences in this domain between ATMPs and other biologicals. This observation indicates that manufacturing is challenging across all biological medicinal products. For ATMPs, these objections revealed themselves as mostly deficiencies in product characterization and specification, analytical tests and assays and their validation, microbiological controls, and, inevitably, comparability studies for process changes. For instance, some products were requested to undergo further characterization, such as for leukapheresis starting material and the viral vector in the case of the chimeric antigen receptor T cell product Kymriah.[27] For other products such as Provenge, Spherox, and Holoclar, it was requested to develop and validate rapid microbiological testing strategies to overcome the 14 days sterility testing issue, as duration of the test might not be suitable for products with a short shelf-life.[28] One important objection related to analytical methods was the potency assay that, ideally, should reflect the biological activity of the product.[16,29,30] For Kymriah and Yescarta, *in vitro* assays successfully revealed the biological activity of the product and the proposed MoA (e.g., level of interferon γ [IFN-γ] produced upon co-culture with the target cells).[16,27] However, potency testing based on surrogate indicators (e.g., cell surface markers expression) for products such as ChondroCelect, MACI, Spherox, and Provenge were more challenging, as meaningful correlations between the biological activity and the surrogate markers had to be established. Interestingly, we observed that more of these objections were solved through post-approval commitments in the case of ATMPs (Figure 3).

The evidence on the design, conduct, and outcome of clinical studies that were submitted by ATMP developers suffered from more objections when compared to other biologicals (Table 2). Clinical trials of ATMPs did not meet the same strict standards for clinical evidence that were applied to other biologicals submissions. Despite matching for the disease area and orphan status, ATMPs had more non-randomized, non-blinded trials and included significantly ($p = 0.009$) lower numbers of patients, raising serious doubts about the trial outcomes. In the case of study outcomes, the modest effect size in the primary endpoint (Provenge, Kymriah, Alofisel, Zalmoxis) or relying on secondary and sub-analyses to show the efficacy of the product (Glybera, Imlygic) represented the main share of objections. Addressing the urgency of patient needs and countering the spread of unproven therapeutic claims[31] has prompted regulatory bodies to launch products with limited clinical evidence.[6] Nevertheless, HTA agencies, including the National Institute for Health and Care Excellence (NICE) and the Institute for Clinical and Economic Review (ICER), acknowledge this flaw and encourage developers to generate additional evidence post-approval.[32–34] It is acknowledged that financial constraints faced by SMEs, which represent the majority of ATMP developers, can have implications on the ability to conduct large (multicenter) clinical trials. Company size has been shown to be a significant factor in a product's chances of approval; for example, for the period 2004–2007, large companies had an MA success rate of 89%, medium sized companies had 73%, whereas for small companies it was only 48%.[19] Moreover, in the case of fresh autologous products with a short shelf-life, challenges with manufacturing and logistics can limit the number of centers

that can be included in the trials.[4] Lastly, robust clinical trial designs with randomization and blinding for ATMPs addressing life-threatening or debilitating conditions might not be feasible. However, we showed in previous studies that nearly half of the currently marketed products, including products that were approved based on single-arm trials such as Kymriah and Yescarta, planned or already started controlled trials in the post-approval phase.[27,35] Such observations suggest that the submissions based on single-arm trials might be a strategic decision rather than being forced by limiting factors. These strategies for regulatory submissions can lower the motivation of the industry to attain robust trial designs at the time of the submission and reserve the larger, more financially demanding trials after securing the MA.

Divergence from the Annex I data requirements was not detected in the EPARs in the manufacturing and quality domain of either cohort. This may seem surprising, as this is the area where the use of a risk-based approach would be expected to be most evident. However, as mentioned previously, the details of this section of the dossier are, for the most part, confidential, and, consequently, the details in the EPAR are limited. Nevertheless, some of the shortcomings observed in the second and third domains, and accepted by regulators, were more prominent in ATMPs as compared to other biologicals. In the non-clinical data packages of ATMPs, the technical hurdles and the relevance of the animal models constituted the most observed divergence (Figure 2). Furthermore, developers of authorized ATMPs relied more on non-GLP studies in their submissions (Figure 2). It seems likely that this relates to difficulties in complying with GLP for such studies, since the reasons provided by developers were accepted. This issue has prompted the EMA to release a question and answer document in 2017.[36,37] Due to the high species specificity of gene therapies, there is a challenge in having animal models available that mimic the tissue tropism, immune response, as well as the cellular specificity in humans for toxicology and biodistribution studies.[38–40] In addition, the lack of clear primary pharmacological targets for some of the cellular therapies significantly complicates the design and the robustness of the proof-of-principle animal studies.[41]

Both clinical and non-clinical biodistribution and other pharmacokinetics as well as non-clinical toxicity studies led to the most divergence for approved and failed products, equally. Such divergence was understandably around twice as common for ATMPs than for other biologicals. *In vivo* cell tracking in animals can be technically difficult, with human subjects presenting an even greater challenge. As more experience is gained with certain cell types and vectors, some of these aspects might become addressable. Some developers may consider the possibility of bypassing traditional *in vivo* animal testing as a benefit; however, these limitations in the non-clinical dataset can pose a significant source of uncertainty, when considering the overall risks and benefits of the product. Properly designed non-clinical studies can reduce such uncertainty and support a positive risk/benefit ratio, while their absence can tip the risk/benefit ratio to the negative or might lead to a CMA with significant post-approval commitments. In our attempt to understand the degree to which a risk-based approach offered flexibility to developers or was accepted by the EMA, we observed only one EPAR, for Provenge, to include a clear statement on using such an approach to justify the extent of the non-clinical data. Two other EPARs referred to risk-based approaches for specific aspects, such as the selection of raw materials and shipping qualification. Consequently, it was challenging to draw such a correlation.

Finally, our results further showed that regulatory objections about the long-term safety and efficacy of ATMPs were addressed through post-approval commitments by performing new clinical trials and deposit data from real-world use into designated registries.[42] Note that ATMP approvals with limited evidence have led to an increased prevalence of exploratory trial designs required to be performed in the post-approval phase, which does not fully mimic the real-world settings.[35] By having many clinical and manufacturing objections for ATMPs addressed in the post-approval settings, developers are overwhelmed with regulatory requirements and commitments, which adds a significant financial, organizational, and administrative burden; in turn, this could impede the product performance and market access.

Conclusions

As of October 2019, 5 out of 14 approved ATMPs were withdrawn after approval. Considering that the first ATMP was approved in October 2009, this is particularly disappointing and warrants analysis such as ours to understand the reasons. As the first study to compare ATMPs to established biologicals, our results send a clear signal that regulations offer a reasonable degree of flexibility in order to bring such innovative therapies to the market. This flexibility comes with a caveat, however. ATMP submissions are authorized with more evidential shortcomings as compared to other biologicals, particularly in the submitted clinical outcomes and trial designs. Such observations, coupled with the high divergence in the non-clinical submission package, create a hurdle for regulators to conduct a well-informed benefit-risk assessment. This might challenge our understanding and confidence in the long-term safety and efficacy of these novel products and could also explain why five ATMPs were withdrawn after approval, approximately 5-fold higher than the matched biological cohort. Even though regulators are imposing extensive post-marketing measures on applicants to overcome these shortcomings, such an approach might impose more hurdles on ATMPs in the post-marketing phase. Our observations are a strong indicator that the scientific community needs to rethink the traditional development framework for such products, in order to mitigate potential evidence deficiencies that may jeopardize their market success. After all, the aim is to develop products that can achieve market sustainability and be available to patients in need.

MATERIALS AND METHODS

Search Strategy

Data on the authorized, rejected, and withdrawn MAA were obtained from the EMA database (https://www.ema.europa.eu/en/medicines/download-medicine-data) (data cutoff, July 1, 2019). Two separate

spreadsheets were obtained: one comprised all of the products that have an EPAR since they completed the evaluation process (authorized and refused), while the other datasheet contained withdrawn products that had a withdrawal assessment report. Screening of all the products presented in the datasheets was performed, and all ATMPs were identified. The corresponding administrative information about each product was then collected through accessing the product-specific profile on the EMA website available from the medicine search engine (https://www.ema.europa.eu/en/medicines). The small and medium-size status of the company was searched on the SME register database (https://fmapps.emea.europa.eu/SME/). When the company was not found, the relevant financial annual report for the year of the MA application submission was obtained and the criteria for SMEs as defined by the EMA were applied.[43]

Pair Matching ATMPs with Other Biologicals

ATMPs (authorized and failed) were matched to other biologicals to compare the differences in the evaluation process. The products were matched on selected confounding factors that can influence the sufficiency of evidence in the EPARs. The selected factors for matching included (1) the MA application outcome (authorized, refused, or withdrawn), (2) the targeted disease which may influence the availability of suitable animal models and the ability to conduct controlled clinical trials (e.g., in case of oncology treatments),[44] (3) the nature and rarity of orphan indications which can complicate the clinical trial design, and patient recruitment;[45] (4) whether products were approved under the CMA or authorization under exceptional circumstances provisions where the product dossiers may have deficiencies in their clinical evidence, and (5) the time at which the application was evaluated, since the regulatory policy, legislation, and guidelines evolve over time and, in turn, the data requirements for MA also evolve. Exact match on MA application outcome, orphan designations and the type of MA was initially conducted. A screening for all the resulted potential biologicals matches was then performed, and exact matching on the disease area was achieved. Afterward, a greedy nearest neighbor matching was used to match the date of MA application outcome for biological submissions, as described elsewhere.[46]

Defining the Data Requirements and the Evidence Domains for Comparison

The data requirements that should be submitted within the frame of an MA application were defined and retrieved from Annex I of Directive 2001/83/EC of the European Parliament and the council (Table S3).[10] Rather than attaining the traditional categorization of the data requirements that group them according to their source (manufacturing, non-clinical, and clinical data), we opted to categorize the data requirements according to their purpose in the scientific evaluation and the decision-making process. Accordingly, a value tree similar to that described in studies of multi-criteria decision analysis (MCDA) of the HTAs was formulated (Figure 1) (Table S3).[47–49]

Based on this approach, the data requirements can be clustered into four main domains manufacturing and quality testing, experimental design and conduct of studies, efficacy and MoA, and safety and toxicity. The first two domains are considered "confidence criteria" and the other two are considered "outcome criteria". The confidence criteria ensure that the manufacturing process itself does not introduce additional risks (e.g., impurities, contaminations, formulation) and is able to constantly produce a product with a defined set of physicochemical or biological characteristics. Furthermore, they also aim to ensure that the submitted studies were designed, conducted, and documented in the most proper way. Any issues in these criteria will affect the level of confidence in the reported outcome criteria. For instance, manufacturing data that indicate a high batch-to-batch variation will affect the level of confidence in the consistency of the presented preclinical and clinical evidence across the different studies. Also, an underpowered clinical trial affects the level of confidence in the benefits reported from such a trial and whether the results can be reproduced in real-world scenarios.

Definitions

Objections

During the evaluation of an MA application, the applicant receives a list of identified issues in the applications under two categories: first is "major objections," defined as critical issues that preclude a recommendation for MA;[50] second is "other concerns," defined as issues that do not preclude a recommendation of the MA, as it can be solved through modifying the summary of product characteristics, or implementation of risk minimization measures.[50] However, in case of failure to solve the other concerns, the product cannot be authorized. Since EPARs do not clearly differentiate between major objections and other concerns, all issues extracted from the EPAR are referred to in this article as objections.

Divergence

Any studies stated as a requirement for the MAA in Annex I of Directive 2001/83/EC and that have not been performed by the applicant should be justified. Justifications include the availability of specific guidelines that deem these studies unnecessary for this kind of therapy, through a rational justification from the applicant or by the application of a risk-based approach. We quantified the degree of divergence by collecting the number of studies that were omitted in the EPARs and accepted during the evaluation of the application.

Data Extraction and Statistical Analysis

Data were then collected, sorted, and coded by M.E and verified by M.A. Upon discrepancies regarding extracted text or sorting of the objections and divergence, discussions were conducted to reach an agreement. All of the data were coded and statistically analyzed using SPSS version 25. Means, ranges, and SDs were used for the descriptive statistics. Due to the small sample size, the matched design, and the exploratory nature of the analysis, a non-parametric statistical test was pre-defined. A Wilcoxon signed-rank test was used to estimate the differences in objections and divergence between the matched pairs. Two-tailed p values less than 0.05 were considered statistically significant. Figures were produced by SPSS version 25 and R studio (version 1.2.1335) using the tidyverse package (version 1.3.0).

SUPPLEMENTAL INFORMATION

Supplemental Information can be found online at https://doi.org/10.1016/j.omtm.2020.05.035.

AUTHOR CONTRIBUTIONS

Conceptualization, M.E. and MA.; Methodology, M.E., and M.A.; Formal Analysis, M.E., and M.A.; Investigation, M.E., C.A.B. and M.A..; Writing –Original Draft, M.E.,and M.A.; Writing – Review & Editing, M.E., C.A.B., A.K. and M.A..; Visualization, M.E.; Funding Acquisition, M.A.; Supervision, A.K., and M.A.

CONFLICTS OF INTEREST

The authors declare no competing interests.

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OMTM, Volume *18*

Supplemental Information

Mitigating Deficiencies in Evidence during Regulatory Assessments of Advanced Therapies: A Comparative Study with Other Biologicals

Magdi Elsallab, Christopher A. Bravery, Andreas Kurtz, and Mohamed Abou-El-Enein

Table 1 List of ATMP submissions

Commercial name	INN	Type	Cell source	Vector type	Indication	ICD 10 disease classification	Developer	SME	Initial evaluation	Type of MA	OD	OD date	Submission date	CHMP opinion date	Withdrawal date	E.C. decision date
Chondro-Celect [1]	Characterized viable autologous cartilage cells expanded *ex vivo* expressing specific marker proteins	TEP	Autologous	NA	Cartilage defects	Diseases of the musculoskeletal system and connective tissue	TiGenix N.V.	yes	Authorized	Full	no	NA	01.06.2007	25.06.2009	NA	05.10.2009
MACI[2]	Matrix-applied characterized autologous cultured chondrocytes	TEP	Autologous	NA	Cartilage defects	Diseases of the musculoskeletal system and connective tissue	Genzyme Europe	no	Authorized	Full	no	NA	01.09.2011	24.04.2013	NA	27.06.2013
Provenge [3]	Autologous peripheral blood mononuclear cells activated with prostatic acid phosphatase granulocyte-macrophage colony-stimulating factor (sipuleucel-T)	CTMP	Autologous	NA	Prostatic neoplasms	Malignant neoplasms except for lymphoid, hematopoietic and related tissue	Dendreon UK LTD	yes	Authorized	Full	no	NA	30.12.2011	27.06.2013	NA	06.09.2013
Spherox[4]	Spheroids of human autologous matrix-associated chondrocytes	TEP	Autologous	NA	Cartilage defects	Diseases of the musculoskeletal system and connective tissue	CO.DON AG	yes	Authorized	Full	no	NA	03.12.2012	15.05.2017	NA	10.07.2017
Imlygic [5]	Talimogene laherparepvec	GTMP	NA	herpes simplex virus type-1 (HSV-1)	Melanoma	Malignant neoplasms except for lymphoid, hematopoietic and related tissue	Amgen	no	Authorized	Full	no	NA	28.08.2014	22.10.2015	NA	16.12.2015
Strimvelis [6]	Autologous CD34+ enriched cell fraction that contains CD34+ cells transduced with retroviral vector that encodes for the human ADA cDNA sequence	GTMP	Autologous	Retroviral vector	ADA-SCID	Diseases of the blood and blood-forming organs	Glaxo SmithKline	No	Authorized	Full	yes	26.08.2005	01.05.2015	01.04.2016	NA	26.05.2016
Alofisel [7]	Darvadstrocel	CTMP	Allogeneic	NA	Anal fistula	Diseases of the digestive system	TiGenix N.V.	yes	Authorized	Full	yes	08.10.2009	02.03.2016	14.12.2017	NA	23.03.2018
Kymriah [8]	Tisagenlecleucel	GTMP	Autologous	Lentivirus	ALL DLBCL	Malignant neoplasms primary of lymphoid, hematopoietic and related tissue	Novartis	no	Authorized	Full	yes	26.04.2014	02.11.2017	28.06.2018	NA	22.08.2018
Yescarta [9]	Axicabtagene ciloleucel	GTMP	Autologous	Retrovirus	DLBCL	Malignant neoplasms of lymphoid hematopoietic and related tissue	Kite Pharma	yes	Authorized	Full	yes	16.11.2014	29.07.2017	28.06.2018	NA	23.08.2018

Commercial name	INN	Type	Cell source	Vector type	Indication	ICD 10 disease classification	Developer	SME	Initial evaluation	Type of MA	OD	OD date	Submission date	CHMP opinion date	Withdrawal date	E.C. decision date
Luxturna [10]	Voretigene neparvovec	GTMP	NA	Adeno-associated viral type 2 (AAV2)	retinal dystrophy	Diseases of the eye and adnexa	Spark Therapeutics	yes	Authorized	Full	yes	02.04.2012	29.07.2017	20.09.2018	NA	22.11.2018
Holoclar[11]	Ex vivo expanded autologous human corneal epithelial cells containing stem cells	TEP	Autologous	NA	Limbal stem-cell deficiency	Diseases of the eye and adnexa	Chiesi Farmaceutici	no	Authorized	Conditional	yes	07.11.2008	06.03.2013	18.12.2014	NA	17.02.2015
Zalmoxis[12]	Allogeneic T cells genetically modified with a retroviral vector encoding for a truncated form of the human low-affinity nerve growth factor receptor (ΔLNGFR) and the herpes simplex I virus thymidine kinase (HSV-TK Mut2)	CTMP	Allogeneic	Retrovirus	HSCT, blood cancer	Malignant neoplasms primary of lymphoid, hematopoietic and related tissue	MolMed SpA	yes	Authorized	Conditional	yes	20.10.2003	05.03.2014	23.06.2016	NA	18.08.2016
Zynteglo[13]	Autologous CD34+ cells encoding βA-T87Q-globin gene	GTMP	Autologous	Lentivirus	beta-thalassemia	Diseases of the blood and blood-forming organs	bluebird bio	yes	Authorized	Conditional	yes	24.01.2013	21.08.2018	28.03.2019	NA	29.05.2019
Glybera [14]	Alipogene tiparvovec	GTMP	NA	Adeno-associated virus type 1 (AAV1)	LPL deficiency	Endocrine nutritional and metabolic diseases	Amsterdam Molecular Therapeutics	yes	Authorized	Exceptional circumstances	yes	08.03.2004	23.12.2009	19.07.2012	NA	25.10.2012
Cerepro (2007) [15]	sitimagene ceradenovec	GTMP	NA	adenovirus serotype 5 (Ad 5)	high-grade glioma	Malignant neoplasms except lymphoid, hematopoietic and related tissue	Ark therapeutics	yes	Withdrawn	NA	yes	06.02.2002	04.10.2005	26.04.2007	13.07.2007	NA
Advexin [16]	contusugene ladenovec	GTMP	NA	adenovirus serotype 5 (Ad 5)	Li-Fraumeni cancer	Malignant neoplasms, except lymphoid hematopoietic and related tissue	Gendux Molecular Limited	yes	Withdrawn	NA	yes	23.10.2006	06.09.2007	NA	17.12.2008	NA
Contusugene Ladenovec Gendux (CLG) [17]	contusugene ladenovec	GTMP	NA	adenovirus serotype 5 (Ad 5)	squamous cell carcinoma	Malignant neoplasms except for lymphoid, hematopoietic and related tissue	Gendux Molecular Limited	yes	Withdrawn	NA	no	NA	02.07.2008	NA	12.06.2009	NA
Cerepro (2010) [18]	sitimagene ceradenovec	GTMP	NA	adenovirus serotype 5 (Ad 5)	high-grade glioma	Malignant neoplasms except for lymphoid, hematopoietic and related tissue	Ark therapeutics	yes	Withdrawn	NA	yes	06.02.2002	28.11.2008	17.12.2009	08.03.2010	NA

Commercial name	INN	Type	Cell source	Vector type	Indication	ICD 10 disease classification	Developer	SME	Initial evaluation	Type of MA	OD	OD date	Submission date	CHMP opinion date	Withdrawal date	E.C. decision date
Oranera [19]	multilayered cell-sheet of autologous oral mucosal epithelial cells	TEP	Autologous	NA	Limbal stem-cell deficiency	Diseases of the eye and adnexa	CellSeed Europe Ltd	yes	Withdrawn	NA	no	NA	01.06 .2011	NA	14.03 .2013	NA
Raligize	axalimogene filolisbac	GTMP	NA	NA	cervical cancer	Malignant neoplasms except for lymphoid, hematopoietic and related tissue	Advaxis Inc	no	Withdrawn	NA	no	NA	13.02 .2018	NA	10.07 .2018	NA
Hyalograft C autograft [20]	characterized viable autologous chondrocytes expanded in vitro, seeded and cultured on a hyaluronan-based scaffold	TEP	Autologous	NA	Cartilage defects	Diseases of the musculoskeletal system and connective tissue	Anika Therapeutics	yes	Withdrawn	NA	no	NA	28.02 .2012	NA	14.01 .2013	NA
Heparesc[21]	Human heterologous liver cells	CTMP	Allogeneic	NA	urea cycle disorders	Endocrine, nutritional and metabolic diseases	cytonet	yes	Rejected	NA	yes	14.09 .2007	05.12 .2013	22.10 .2015	NA	21.12 .2015

SME: small and medium-sized enterprise; O.D.: orphan designation; MA: marketing authorization; CHMP: Committee for Medicinal Products for Human Use; E.C.: European Commission; GTMP: gene therapy medicinal product; TEP: tissue-engineered product; CTMP: cell therapy medicinal product; LPL: lipoprotein lipase, HSCT: hematopoietic stem cell transplantation, ALL: acute lymphoblastic leukemia; DLBCL: diffuse large B-cell lymphoma; ADA-SCID: adenosine deaminase deficiency - severe combined immune deficiency.

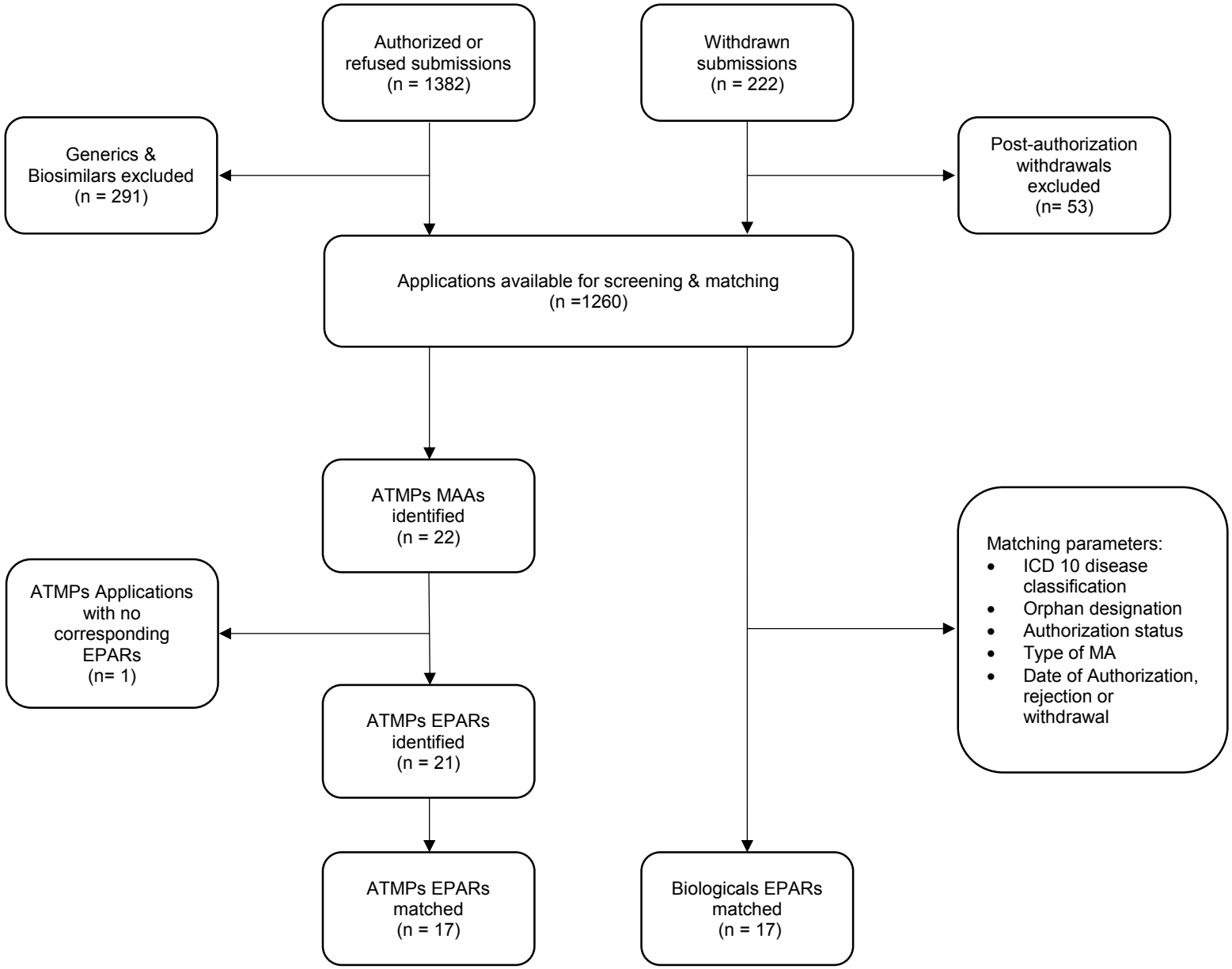

Figure 1 flow chart of the screening of the EMA database, data retrival and products matching.

ATMPs: advanced therapy medicinal products, MAA: marketing authorization application, MA: marketing authorization, EPAR: European public assessment report, ICD: International Classification of Diseases.

Table 2 list of the matched Biological Medicinal Products

Commercial name	INN	Type	Indication	Developer	SME	ICD 10 disease classification Indication	Initial MA status	Type of MA	OD	OD date	Submission date	CHMP opinion date	Withdrawal date	E.C. decision date	ATMP Match
Simponi [22]	Golimumab	monoclonal antibody	Rheumatoid arthritis, Psoriatic arthritis Axial, spondyloarthritis	Centocor B.V. currently (Janssen Biologics B.V.)	No	Diseases of the musculoskeletal system and connective tissue	Authorized	Full	No	NA	03.03 .2008	25.07 .2009	NA	01.10 .2009	Chondroc elect
Krystexxa [23]	Pegloticase	Recombinant Enzyme	Gouty arthritis	Savient Pharma	No	Diseases of the musculoskeletal system and connective tissue	Authorized	Full	No	NA	03.05 .2011	18.10 .2012	NA	08.01 .2013	MACI
Kadcyla [24]	Trastuzumab emtansine	monoclonal antibody (antibody-drug conjugate)	Advanced or metastatic breast cancer	Roche	No	Malignant neoplasms except for lymphoid, hematopoietic and related tissue	Authorized	Full	No	NA	30.08 .2012	19.09 .2013	NA	15.11 .2013	Provenge
Kevzara [25]	Sarilumab	monoclonal antibody	Chronic idiopathic arthritis	Sanofi-aventis group	No	Diseases of the musculoskeletal system and connective tissue	Authorized	Full	No	NA	24.06 .2016	21.04 .2017	NA	23.06 .2017	Spherox
Portrazza[26]	Necitumumab	monoclonal antibody	Squamous non-small cell lung cancer	Eli Lilly Netherlands	No	Malignant neoplasms except for lymphoid, hematopoietic and related tissue	Authorized	Full	No	NA	01.12 .2014	17.12 .2015	NA	15.02 .2016	Imlygic
Alprolix [27]	Eftrenonacog alfa	Recombinant coagulation factor (fusion protein)	Hemophilia B	Biogen Idec Ltd	No	Diseases of the blood and blood-forming organs	Authorized	Full	Yes	08.06 .2007	04.06 .2015	25.02 .2016	NA	12.05 .2016	Strimvelis
Revestive[28]	Teduglutide	Recombinant Hormone	Short bowel syndrome	Nycomed Denmark	No	Diseases of the digestive system	Authorized	Full	Yes	11.12 .2001	03.03 .2011	14.12 .2017	NA	03.08 .2012	Alofisel
Besponsa [29]	Inotuzumab ozogamicin	monoclonal antibody (antibody-drug conjugate)	Precursor Cell Lymphoblastic Leukemia-Lymphoma	Pfizer Limited	No	Malignant neoplasms of lymphoid, hematopoietic and related tissue	Authorized	Full	Yes	07.06 .2013	14.04 .2016	21.04 .2017	NA	28.06 .2017	Kymriah
Mylotarg [30]	Gemtuzumab ozogamicin	monoclonal antibody (antibody-drug conjugate)	Acute myeloid leukemia	Pfizer Limited	No	Malignant neoplasms of lymphoid, hematopoietic and related tissue	Authorized	Full	Yes	18.10 .2000	01.12 .2016	22.02 .2018	NA	19.04 .2018	Yescarta
Oxervate [31]	Cenegermin	Recombinant growth factor	Neurotrophic keratitis	Dompé farmaceutici	No	Diseases of the eye and adnexa	Authorized	Full	yes	NA	03.11 .2016	18.05 .2017	NA	06.07 .2017	Luxturna
Adcetris [32]	Brentuximab vedotin	monoclonal antibody (antibody-drug conjugate)	Hodgkin's lymphoma	Takeda Global Research	No	Malignant neoplasms of lymphoid, hematopoietic and related tissue	Authorized	Conditional approval	Yes	15.01 .2009	31.05 .2011	19.07 .2012	NA	25.10 .2012	Zalmoxis
Strensiq [33]	Asfotase alfa	Recombinant Enzyme (Fusion protein)	Hypophosphatasia	Alexion Europe	No	Endocrine, nutritional and metabolic diseases	Authorized	Exceptional circumstances	Yes	03.12 .2008	01.07 .2014	25.06 .2015	NA	28.08 .2015	Glybera
Theraloc [34]	Nimotuzumab	monoclonal antibody	High-grade glioma.	Oncoscience AG	Yes	Malignant neoplasms except for lymphoid, hematopoietic and related tissue	Withdrawn	NA	Yes	02.09 .2004	04.10 .2007	NA	01.12 .2008		Cerepro (2007)

Commercial name	INN	Type	Indication	Developer	SME	ICD 10 disease classification Indication	Initial MA status	Type of MA	OD	OD date	Submission date	CHMP opinion date	Withdrawal date	E.C. decision date	ATMP Match
Oncophage [35]	vitespen	Autologous Tumor-Derived Protein-Peptide Complex	Renal cell carcinoma	Antigenics Therapeutics Limited	No	Malignant neoplasms except for lymphoid, hematopoietic and related tissue	Withdrawn	NA	Yes	11.04 .2005	29.09 .2008	NA	23.11 .2009		Advexin
Zafiride [36]	Ngr-human tumor necrosis factor-alpha	Recombinant cytokine (Fusion protein)	Advanced malignant pleural mesothelioma	Molmed	Yes	Malignant neoplasms except for lymphoid, hematopoietic and related tissue	Withdrawn	NA	Yes	03.06 .2008	03.12 .2016	NA	01.06 .2017		Cerepro (2010)
Plivensia [37]	Sirukumab	monoclonal antibody	Rheumatoid arthritis	Janssen-Cilag	No	Diseases of the musculoskeletal system and connective tissue	Withdrawn	NA	No	NA	12.09 .2016	NA	26.10 .2017		Hyalograf t C autograft
Elelyso [38]	Taliglucerase alfa	Recombinant Enzyme	Type 1 Gaucher disease	Pfizer Ltd	No	Endocrine, nutritional and metabolic diseases	Rejected	NA	Yes	23.03 .2010	25.11 .2010	03.07 .2012	NA	25.10 .2012	Heparesc

SME: small and medium-sized Enterprise, OD: orphan designation, MA: marketing authorization, CHMP: Committee for Medicinal Products for Human Use, EC: European Commission

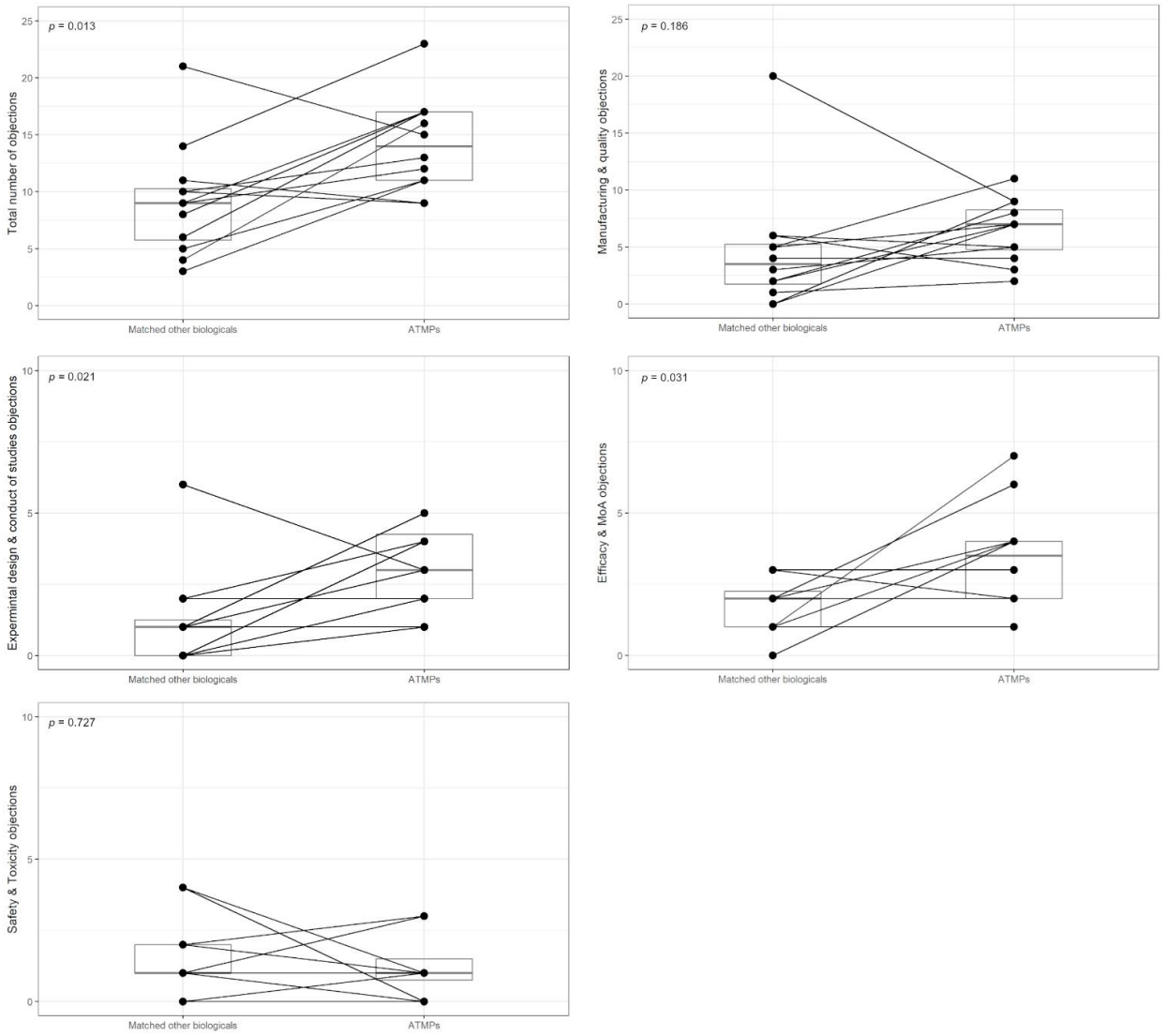

Figure 2 Paired dot plots and boxplots of objections in matched authorized ATMPs and biologicals submissions. ATMPs (authorized and failed) were matched to other biologicals via a matched-pair experimental design to compare the difference in the evaluation process between both. Regulatory objections were scored using quantitative assessment of the European public assessment reports (EPARs) of each product. The groups were compared statistically using two-tailed Wilcoxon signed-rank test. In the authorized cohorts ATMPs showed significantly higher differences in the total number of objections, the experimental design and conduct of the studies, and the efficacy and mode of Action (MoA) as depected in the figure. Statistical test: Wilcoxon signed-rank test.

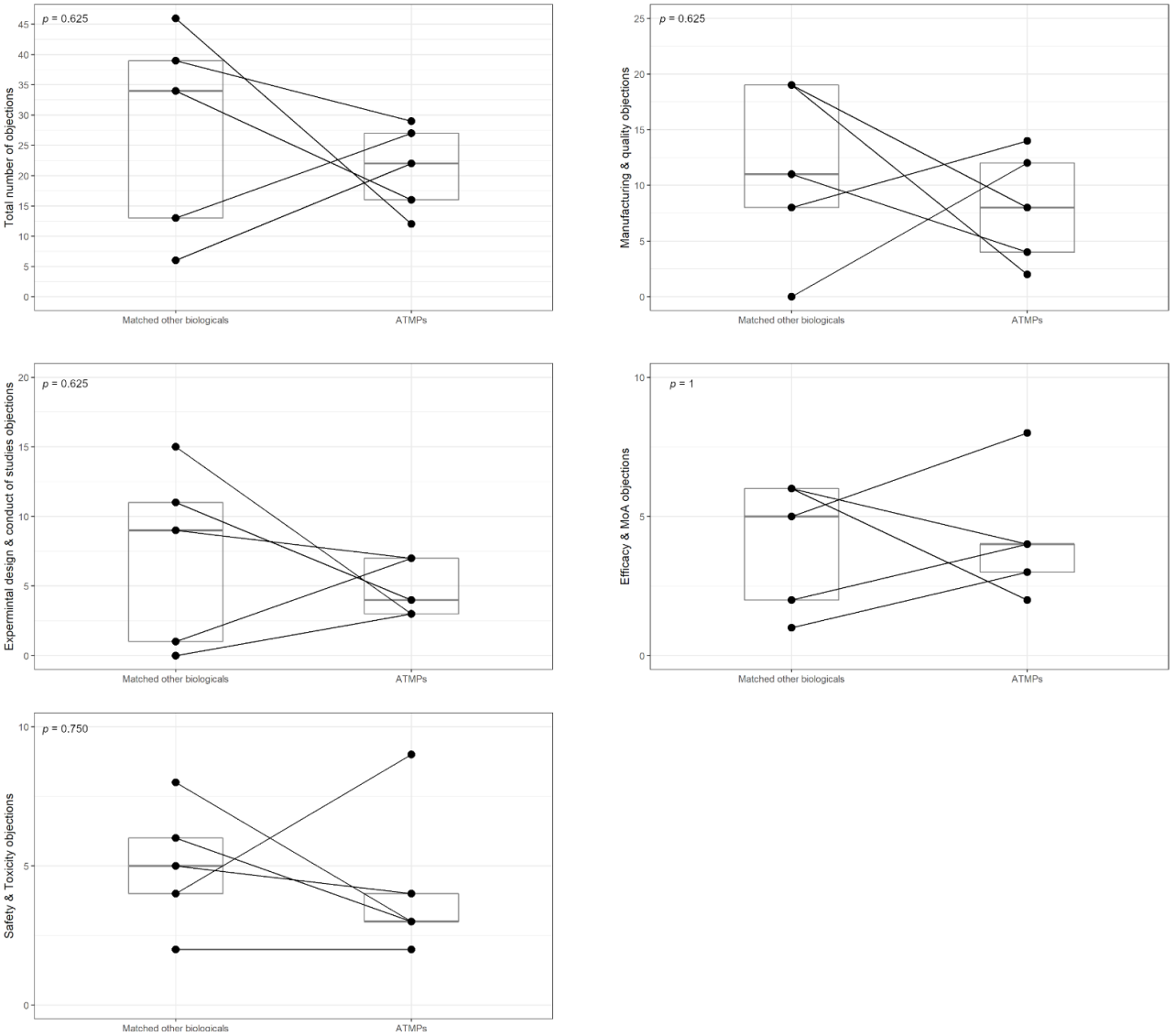

Figure 3 Paired dot plots and boxplots of objections in matched failed ATMPs and biologicals submissions. ATMPs (authorized and failed) were matched to other biologicals via a matched-pair experimental design to compare the difference in the evaluation process between both. Regulatory objections were scored using quantitative assessment of the European puplic assessment reports (EPARs) of each product. The groups were compared statistically using two-tailed Wilcoxon signed-rank test. In the failed cohorts no statistically significant difference were noted in any of the comparisons. Statistical test: Wilcoxon signed-rank test.

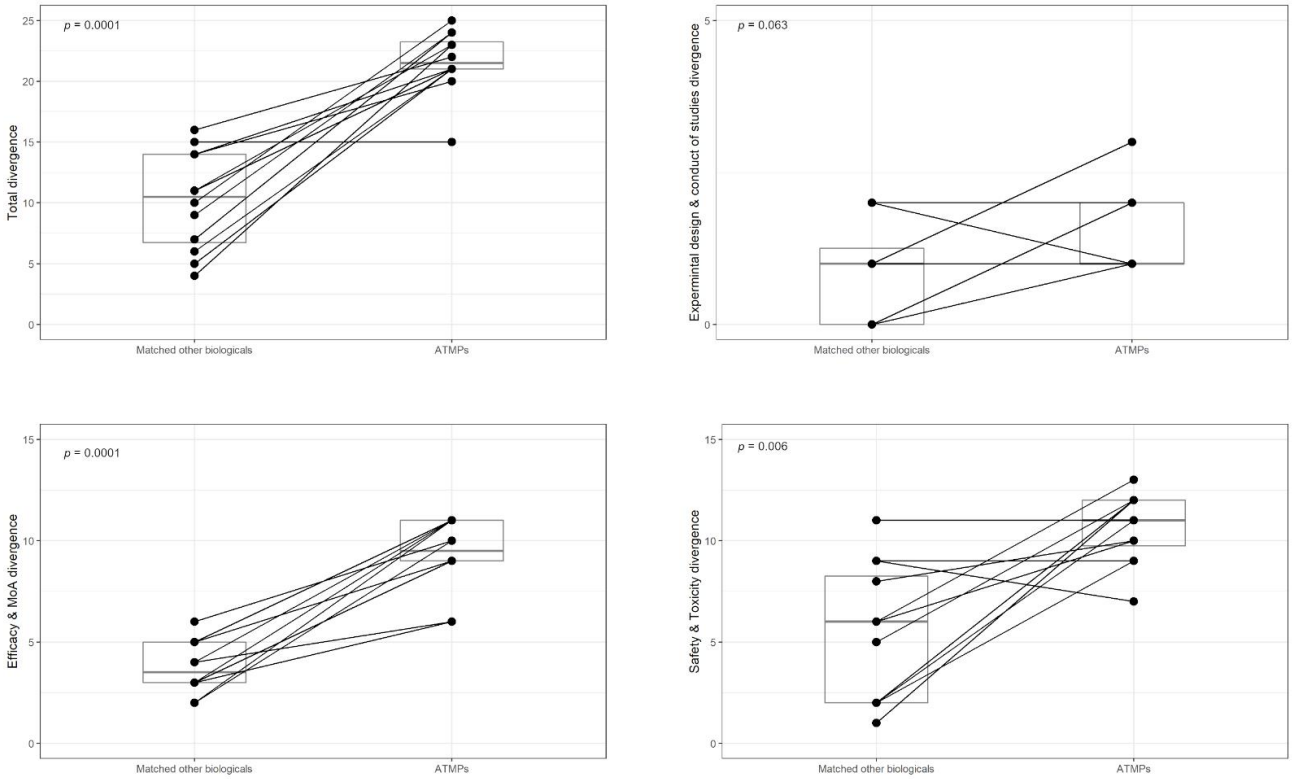

Figure 4 Paired dot plots and boxplots of divergence in matched authorized ATMPs and biologicals submissions. ATMPs (authorized and failed) were matched to other biologicals via a matched-pair experimental design to compare the difference in the evaluation process between both. Divergence from the regulatory requirments laid down in Annex I of Directive 2001/83/EC were measured using quantitative assessment of the omitted studies in the European puplic assessment reports (EPARs) of each product. The groups were compared statistically using two-tailed Wilcoxon signed-rank test. Significantly higher divergence were noted in the total numbers of divergence, the divergence in the efficacy and mode of action studies, as well as the divergence in safety and toxicity studies in the ATMPs cohort compared to the matched other biologicals. Statistical test: Wilcoxon signed-rank test.

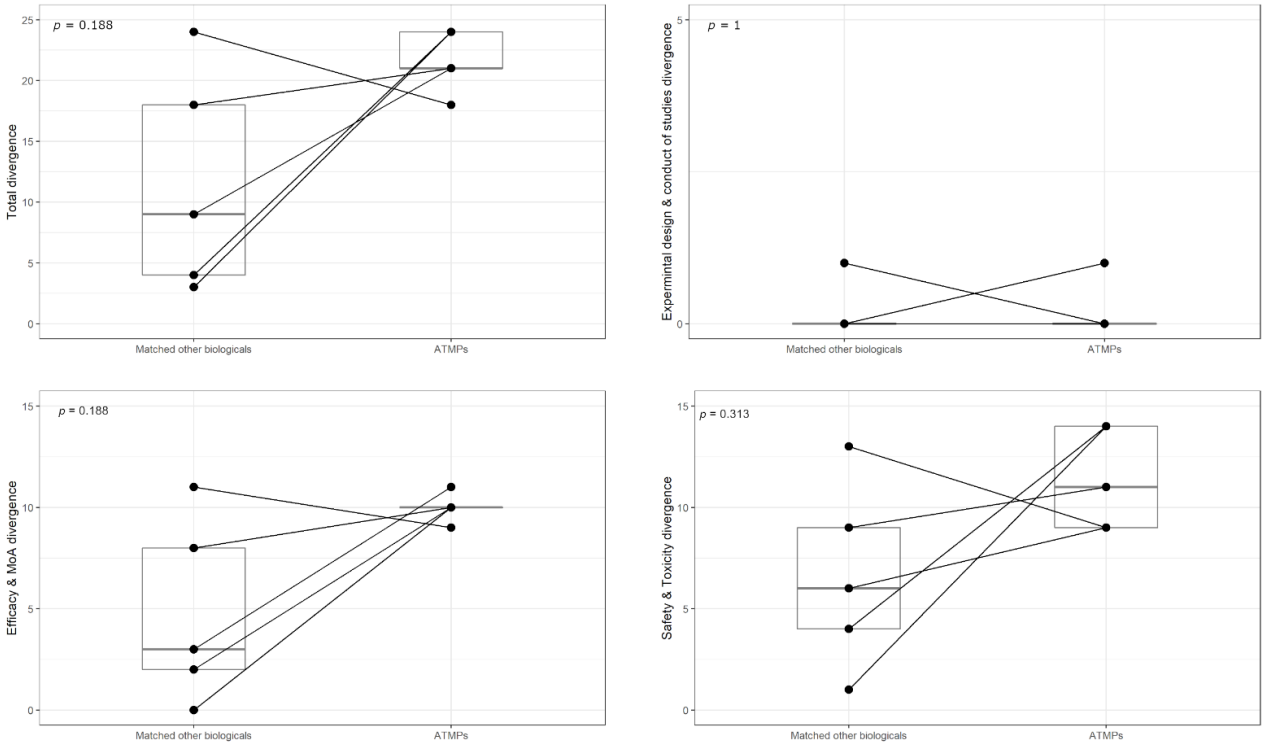

Figure 5 Paired dot plots and boxplots of divergence in matched failed ATMPs and biologicals submissions. ATMPs (authorized and failed) were matched to other biologicals via a matched-pair experimental design to compare the difference in the evaluation process between both. Divergence from the regulatory requirments laid down in Annex I of Directive 2001/83/EC were measured using quantitative assessment of the omitted studies in the European puplic assessment reports (EPARs) of each product. The groups were compared statistically using two-tailed Wilcoxon signed-rank test. No statsticall significant differences were reported in the divergence in the total numbers or the divergence in any of the evidence domains. Statistical test: Wilcoxon signed-rank test.

Table 3 evidence domains, data requirments, and definitions.

Evidence domains	Data requirments	Definition		reference
Quality of manufactured product	**Good manufacturing practice (GMP) compliance**	Compliance to the set of guidelines that ensure that the produced active pharmaceutical ingredients are consistent high quality. The guidelines include rules for quality management, personnel, building and facility, process equibment, documentation, material mangment, production, packaging, and storage.		[39]
	Control of materials (starting, raw, excipients)	Materials used in production of the active pharmaceutical ingredients and the final products. The quality of each material should be confirmed by an appropriate set of test methods and acceptance criteria (specification).		[39]
	Manufacturing process design & control stratgey	Manufacturing process should be clearly defined and controlled. Control strategy is defined as the planned set of controls that are derived from the current product and the understanding of the manufacturing process that assures process performance and product quality.		[39]
	Manufacturing process validation	Evidence that the manufacturing process when operated within defined parameters, can produce an intermediate or active pharmaceutical ingrediet with consistent set of predifiened specifications and quality attributes.		[39]
	Choice of Analytical methods (e.g., assays)	Suitability of the analytical methods used for process control, release testing and stability.		[40]
	Analytical methods validation	A documented program that provides a high degree of assurance that a specific process, method, or system will consistently produce a result meeting pre-determined acceptance criteria		[39,40]
	Comparability	The activities, including study design, conduct of studies, and evaluation of data, that are designed to investigate whether the products are comparable.		[41,42]
	Stability testing	Data on the stability of of the drug substance and drug product under different conditions that confirms the product remins within specifiction when stored or handled as intended.		[43]
	Product characterization, specification & acceptance criteria	Determination of physicochemical properties, biological activity, purity and impurities by appropriate analytical methods. The outcome of such studies are used to identify relevant test methods. Acceptance criteria are established from batch data, process characterisation and other studies.		[44]
Experimental design and conduct of the studies	**Non-clinical studies**	**GLP compliance**	Compliance to the set of rules and criteria laid down in Directive 2004/9/EC and Directive 2004/10/EC. GLP is a quality system concerned with the organisational process and the conditions under which non-clinical health and environmental safety studies are planned, performed, monitored, recorded, reported and archived.	[45]
		Animal models & experiments	*In vivo* and/or *in vitro* studies designed to explore the pharmacology, PK/PD and biodistribution, toxicity and other desirable or undesirable biological effects. Such studies aim to mimic the human disease and intended route of administration, dose and dosing schedule intended for humans.	[46]
	Clinical studies	**GCP & protocol compliance**	Compliance to the princibles of good clinical practice that insure that the design, conduct, recording and repoting of the clinical studies are of high quality. Deviation from such principles should be assessed for its impact on the quality and the integrity of the clinical studies.	[47]

		Clinical Study methodology	All the aspects related to the design of the main clinical study submitted for the marketing authorization. These aspects include the control arm of the trial, randomization, blinding, adequacy of the sample size and statistical methods.	[48]
		Study population	Data that show that the included population in the study is well-defined through clear inclusion and exclusion criteria which is crucial for assessing the target population and the intended indication.	[47]
		Choice of Endpoints	Study endpoints are the response variables that are chosen to assess drug effects that are related to pharmacokinetic parameters, pharmacodynamic measures, efficacy and safety. A primary endpoint(s) should reflect clinically relevant effects and is typically selected based on the principal objective of the study. Secondary endpoints assess other drug effects that may or may not be related to the primary endpoint. Endpoints and the plan for their analysis should be prospectively specified in the protocol.	[48]
Efficacy & mode of action	**Non-clinical evidence**	**Pharmacodynamics studies**	Primary non-clinical PD studies should address the mode of action (MoA) related to intended therapeutic use and provide knowledge on the interaction of the investigational medicinal product with the intended target as well as with related targets.	[49]
		Pharmacokinetics/Biodistribution studies (PK/BD)	Non-clinical part of the PK/BD that focus on the interaction of the investigation medicinal product with the taget action site, hence influencing the efficacy of the product. This either include the traditional Pharmacokinetic studies (absorbtion, distribution) or other BD assessments such as distribution, persistence of the drug product.	
	Clinical evidence	**Primary Pharmacodynamics studies**	Studies on the mode of action and/or effects of a substance in relation to its desired therapeutic target are primary pharmacodynamic studies. Evidence that can provide early estimates of activity and potential efficacy and may guide the dose and dosing regimen in later studies.	[48,50]
		Pharmacokinetics/Biodistribution (PK/BD)	See nonclinical PK/BD	
		Dose finding studies	A dose-finding study is a clinical trial that aims to outline the no-effect dose, the mean effective dose, and the maximal effective dose while taking tolerability into account to define an optimal dose.	[51]
		Clinical efficacy results	The degree to which a medicinal product produces a beneficial effects under ideal and controlled conditions. Usually obtained from the main study submitted in the marketing authorization application.	[52]
		Long-term clinical efficacy	The long-term benefits of the medicinal product	
		Indication	The disease(s) or condition(s) and population(s) that a medicine is intended to treat.	[53]
		Post-hoc analysis and meta-analysis and supportive studies	Any studies or analyses other than that of the main study that are conducted and included in the marketing authorization application to support the claims of the efficacy. These studies include post-hoc analyses, meta-analyses across studies, and other supportive studies.	
Safety & Toxicity	**Non-clinical evidence**	**Non-clinical Toxicity studies**	Non-clinical studies that measure functional indices of potential toxicity in animal studies. This include general toxicity studies, genotoxicity, tumorgenicity, immunotoxicity, and local tolerance.	[50]

		Pharmacokinetics/Bioditribution PK/BD	Non-clinical Part of the PK/BD that focus on the interaction of the drug product with sites other than the taget action site, hence influencing the safety of the product. This either include the traditional Pharmacokinetic studies (metabolism, and excreation) or other BD assessments such as mobilization, clearance, shedding, and off-target distribution of the biologicaly active substance.	
	Clinical evidence	**Adverse events**	Any untoward medical occurrence in a patient or clinical trial subject administered a medicinal product and which does not necessarily have a causal relationship with this treatment.	[48]
		Long term safety data	The long-term studies to identify any undesirable effects of the product	
		Secondary pharmacodynamics studies	Secondary pharmacodynamic studies (previously referred to as general pharmacology) can be defined as studies on the mode of action and/or effects of a substance not related to its desired therapeutic target.	[50]
		Pharmacokinetics/Bioditribution PK/BD	see nonclinical PK/BD	
	Risk-management plan		Risk management plans include: (1) the identification or characterisation of the safety profile of the medicinal product, with emphasis on important identified and important potential risks and missing information, and also on which safety concerns need to be managed proactively or further studied (the ‘safety specification’); 2. the planning of pharmacovigilance activities to characterise and quantify clinically relevant risks, and to identify new adverse reactions (the ‘pharmacovigilance plan’); 3. the planning and implementation of risk minimisation measures, including the evaluation of the effectiveness of these activities (the ‘risk minimisation plan’).	[54]

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4. CURRICULUM VITAE

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5. COMPLETE LIST OF PUBLICATIONS

Publications:	**Impact factor (2019)**
• **Elsallab, M.**, Levine, BL., Wayne, AS. and Abou-El-Enein, M. (2020). CAR T-cell product performance in haematological malignancies before and after marketing authorisation. Lancet Oncology. Volume: 21, Pages: e104–e116.	34.340
• **Elsallab, M.**, Bravery, CA., Kurtz, A. and Abou-El-Enein, M. (2020). Mitigating Deficiencies in Evidence during Regulatory Assessments of Advanced Therapies: A Comparative Study with Other Biologicals. Molecular Therapy. – Methods & Clinical Development. Volume: 18, Pages: 269–279.	4.533
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