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CAR-T CELL THERAPY: MANUFACTURING CHALLENGES AND COST CONSIDERATIONS

PABLO SOTOMAYOR[1]*, LUCIE PROCHAZKA[2], MATTHIAS BROCKHAUS[3]

Affiliation

[1]Department of Pharmaceutical Biotechnology, Navarre Plain Institute of Pharmacy, Pamplona, Spain
[2]Department of Biotechnology, Vysocina Institute of Pharmaceutical Chemistry, Jihlava, Czech Republic
[3]Department of Biotechnology, Neckar Valley Institute of Pharmaceutical Sciences, Heilbronn, Germany

*Corresponding author: Pablo Sotomayor, p.sotomayor@npip.es
ORCID iD: Pablo Sotomayor 0000-0001-0523-711X; Lucie Prochazka 0000-0001-4468-4430; Matthias Brockhaus 0000-0001-4561-3124

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Abstract

Chimeric antigen receptor (CAR) T-cell therapy has produced durable remissions in some patients with refractory B-cell cancers, yet each dose is a bespoke, living product made from the patient's own cells. This narrative review summarises published evidence on how autologous CAR-T products are manufactured, where the process fails or slows, how quality and regulation are handled, and what the products cost health systems. Collection, activation, gene transfer, expansion and release testing each carry technical and logistical limits, and published economic models suggest value only under specific assumptions. Automation, shorter processes and allogeneic products are promising but unproven at scale. This review reports no new data, and cost evidence is largely modelled.

1. INTRODUCTION

Few cancer treatments have changed the conversation as quickly as CAR-T cells. A patient's own T lymphocytes are collected, engineered in a laboratory to carry a receptor that recognises a tumour antigen, multiplied, and infused back after lymphodepleting chemotherapy [1, 2]. In children with relapsed B-cell acute lymphoblastic leukaemia and in adults with refractory large B-cell lymphoma, registration trials of tisagenlecleucel and axicabtagene ciloleucel reported remissions in cases that previously had few options [3, 4, 5]. Later products

extended the approach to mantle-cell lymphoma and multiple myeloma [6, 7]. The enthusiasm is tempered by practical reports from treating centres. Early adopters describe a demanding pathway in which patients must be identified, referred, assessed for fitness, apheresed, bridged with other therapy while the product is made, and then admitted for conditioning and infusion. Every step has a person, a schedule and a cost attached, and delay at any point can matter to a patient with aggressive disease [8, 9]. Response rates in these trials were high, but durability varied by disease and product, and the follow-up was short for many of the cohorts.

The medicine, however, is not a tablet or an infusion bag produced by the thousand. It is a patient-specific biological made in small batches, with starting material that varies from person to person, and a shelf life measured in hours or days. These facts shape almost everything about how the therapy is made, released and paid for. Manufacturing is not a back-office detail here; it determines who gets treated and when, and whether a patient whose disease is progressing can wait for the product [10, 11]. That dependence on a chain of events is also why the field speaks of an end-to-end supply chain and not just a production line. Cells travel by courier between hospital and factory, sometimes across continents, and the loss of a shipment or a missed slot in a facility has consequences that a delayed delivery of a conventional drug would not. Early reviews of the commercial model already pointed to scale-up, not efficacy, as the real test of the technology [2, 10, 12].

Interest in the second-line use of the therapy has grown, with phase 3 trials comparing axicabtagene ciloleucel and lisocabtagene maraleucel against standard salvage chemotherapy and transplantation in large B-cell lymphoma, and the findings widen the population who might be considered and therefore the pressure on capacity [13, 14]. This review follows the product from collection to infusion, covering production steps, quality control and regulation, and then costs. Evidence is gathered in table 1. It is a narrative synthesis of published papers and reports no new data. Two further questions follow from this background. First, which parts of the process most limit speed, success rate and reproducibility, and what has been tried to improve them? Second, what does the whole system cost, who pays, and how robust are the published judgements of value? The sections below treat these in order, and fig. 1 summarises how the evidence collected in table 1 is distributed across clinical, manufacturing, quality and economic themes.

2. CAR-T PRODUCT DESIGN

A CAR is a fusion protein that joins an extracellular antigen-recognition domain, usually a single-chain variable fragment from an antibody, to intracellular signalling domains from the T-cell receptor complex. First-generation constructs carried CD3-zeta alone and persisted poorly. Second-generation receptors add a costimulatory domain, CD28 or 4-1BB, and these underlie the licensed products [1, 15, 16]. The costimulatory choice affects expansion kinetics, persistence and the timing of toxicity, so the construct is part of the product's identity and of what must be controlled in manufacture. The choice of binding domain also carries risk. Mouse-derived fragments can be recognised by the immune system and may contribute to loss of persistence, which has led some developers to humanised or fully human binders. Affinity must be tuned, because too weak a binder fails to activate and too strong a binder may drive exhaustion or toxicity. These trade-offs are settled during development, but they set the specification that manufacturing must later meet [1, 2, 16]. Understanding these domains helps pharmacists and clinicians appreciate why products with similar targets are not interchangeable.

Gene transfer is usually carried out with replication-incompetent gamma-retroviral or lentiviral vectors, which integrate into the T-cell genome and give stable expression. The vector itself is a complex biological product that must be manufactured under good manufacturing practice, tested for replication-competent virus, and supplied in the quantities needed for each patient batch [10, 17]. Non-viral approaches, including transposon systems and messenger RNA

electroporation, offer cheaper or more transient alternatives, though most are still at early clinical stages [16, 18]. Vector supply is a recognised bottleneck. Producing clinical-grade lentivirus requires transfection of producer cells, purification and extensive testing, and a single vector lot may take months to prepare and cost a large sum. Shortages or delays in vector supply have limited the number of patients who could be treated in some programmes. For academic groups, access to vector manufacturing capacity is frequently the limiting factor, ahead of cell processing itself [9, 10, 19].

Table 1: Selected published evidence on CAR-T manufacturing and cost

Study or topic	Reported finding or feature	Ref.
Tisagenlecleucel paediatric leukaemia	High remission rates in refractory disease; some enrolled patients did not receive product.	[3]
Axicabtagene ciloleucel refractory lymphoma	Durable responses in a population with few alternatives; cytokine release and neurotoxicity were common.	[4, 13]
Second-line trials lymphoma	Randomised comparisons against salvage chemotherapy and transplantation widened potential eligibility.	[13, 14]
Starting material T-cell phenotype	Less differentiated cells in the apheresis product were associated with response in leukaemia.	[20]
Culture duration preclinical work	Shorter ex vivo culture preserved less-differentiated cells and improved antitumour activity.	[21]
Automated closed systems device-based	Single-device processes produced clinical-grade cells with less hands-on time.	[22, 23]
Regulatory guidance FDA and EMA	Expect control of vector, cell characterisation, potency, comparability and long-term follow-up.	[24, 25]
Toxicity grading ASTCT consensus	Shared scales for cytokine release syndrome and neurotoxicity support comparison across centres.	[26, 27]
Total cost of care US estimate	Costs of apheresis, admission and toxicity add substantially to the product price.	[28]
Cost-effectiveness models lymphoma and leukaemia	Favourable ratios depend on assumed long-term survival beyond trial follow-up.	[29, 30, 31]
Outpatient administration modelling	Lower modelled cost than inpatient care in selected patients.	[32]

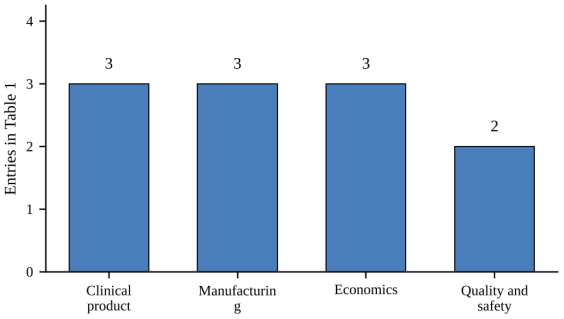


Fig. 1: Number of entries in table 1 by theme

Choice of the target antigen shapes the clinical plan. CD19 is present on normal and malignant B cells, which makes it a useful target and also explains the expected loss of normal B cells and the hypogammaglobulinaemia that follows. B-cell maturation antigen is the target in myeloma. Escape through loss of antigen is a recognised mechanism of relapse, so dual-targeting constructs and combinations are under study, and each makes manufacturing and release testing a little more complicated [7, 15, 20]. Safety features are being designed into constructs, including suicide genes, switchable receptors and logic gates that require two antigens to activate the cell. These additions can improve control but complicate characterisation and may increase the number of tests required at release. Each extra element has to be shown to function in every batch, which is why many developers keep designs as simple as the clinical question allows [1, 15, 16]. Practical consequences include the need for validated antigen testing of the tumour before treatment.

3. MANUFACTURING STEPS AND BOTTLENECKS

Production starts with leukapheresis at a clinical site. White cells are collected over a few hours, shipped cold to the manufacturing

facility, and checked on arrival. This step looks simple but is a major source of variability: lymphocyte counts differ widely, and patients heavily pretreated with chemotherapy yield fewer and less fit T cells [10, 11]. Apheresis centres also differ in practice, which is one reason standardised collection protocols have been proposed [11, 18]. Timing of collection relative to previous therapy matters. Cytotoxic drugs, steroids and some targeted agents may reduce the number or function of T cells, and many centres impose a washout period before apheresis. At the same time, patients with active disease cannot wait indefinitely, so clinicians must balance cell quality against clinical urgency. The more heavily pretreated the patient, the greater the difficulty [10, 11, 20].

T cells are then enriched or selected, often by magnetic beads, and activated with antibodies against CD3 and CD28 or with equivalent reagents. Contaminating monocytes and granulocytes have been shown to impair T-cell expansion, which is why cell selection and the composition of the starting material matter more than they might seem [18, 33]. After activation the cells are transduced with the vector, typically within one or two days, and the efficiency of transduction partly determines the dose that can be achieved [10, 17]. Process engineers have tried to remove monocytes by elutriation, by adherence or by selecting for CD4 and CD8 cells directly, and in some programmes a defined CD4 to CD8 ratio is the target. Whether a defined composition gives better outcomes than a bulk mixture has not been settled in randomised comparisons, so the added complexity of selection is justified more by consistency than by proof of clinical gain [11, 18, 33]. Reagents of this kind must themselves be sourced to quality standards, adding another supplier to audit.

Expansion takes place in culture bags, gas-permeable flasks or bioreactors, usually for a week or more, with cytokines such as interleukin-2, interleukin-7 or interleukin-15. A longer culture produces more cells but also drives differentiation towards less persistent phenotypes. Ghassemi and colleagues showed in preclinical work that shortening ex vivo culture preserved less-differentiated cells and improved antileukaemic activity, a finding that has prompted interest in rapid manufacturing processes [21]. The balance between dose and quality remains a design decision [18, 22]. Different expansion platforms suit different scales. Static bags are simple and cheap but need frequent manual handling. Rocking-motion bioreactors give better gas exchange and allow higher densities, and stirred-tank or hollow-fibre systems have been explored for larger runs. Because each autologous batch is small, bigger vessels offer little benefit, and the economics favour many small parallel processes rather than one big one [12, 17, 18].

Harvest, formulation and cryopreservation follow. Cells are washed, concentrated, mixed with cryoprotectant, frozen in a controlled-rate freezer and kept in the vapour phase of liquid nitrogen until shipment. Release testing for identity, viability, purity, potency, sterility, mycoplasma and endotoxin must be completed before the product can travel, and the test turnaround time adds directly to the wait. Some tests, such as sterility by culture, take days, so rapid alternatives are under development [11, 17, 19]. Cryopreservation is convenient but not free of cost. Freezing and thawing may reduce viability and alter function, and the logistics of cryogenic shipping and storage at treatment sites add both expense and risk. Some groups have asked whether fresh products could be infused directly, shortening the process, but fresh cells need tight scheduling and cannot be tested as fully before use [11, 12, 19]. Local expertise in handling these tests is therefore as important as the equipment.

Throughput is the broader challenge. Traditional manufacturing is open and manual, requiring skilled operators and cleanroom space, and each patient's product occupies a suite and a team for days. Automated closed systems, such as the CliniMACS Prodigy, combine selection, activation, transduction and expansion in a single disposable set, reducing hands-on time and contamination risk, and published runs have yielded clinically relevant doses [22, 23]. Even so, the machine does not remove the need for quality oversight [9, 19]. Process

analytical technology and real-time monitoring are slowly entering the field. Measurements of metabolites in the culture medium, cell counts by in-line sensors and automated sampling can show deviations earlier and lower the burden on operators. They also generate data that allow a manufacturer to learn which early signals predict a good final product, an approach that has worked in biologics but is only beginning here [9, 22, 23]. Training of operators is a continuing cost.

Failure of manufacture is a patient-level event rather than a batch deviation. In registration trials a minority of patients who were enrolled and underwent apheresis did not receive the product, because of insufficient expansion, failed release criteria, or disease progression or death during the wait [3, 4, 5]. These dropouts are counted in intention-to-treat analyses of some trials but hidden in others that report only treated patients, which can flatter outcomes [14, 34]. Readers should check which denominator a trial uses. Product failures may also arise from the patient's cells themselves. Leukaemic blasts in the apheresis material, contamination with tumour cells and infection are recognised causes of problems, and an apheresis product with very few T cells may not support manufacture at all. Reported failure rates in trials were low in the lymphoma studies but more visible in some leukaemia and myeloma series [3, 7, 11].

4. QUALITY CONTROL AND REGULATION

Quality assurance of a living product with a very small batch size is an uncomfortable fit for classical pharmaceutical thinking. Destructive tests consume part of a dose that cannot be remade, retained samples are limited, and the usual idea of a lot representing thousands of identical units does not apply. Manufacturers therefore rely on in-process controls and a carefully defined control strategy, and release specifications are set on the basis of relatively few clinical batches [11, 19, 35]. Some of the controls are specific to gene-modified cells. Vector copy number per cell is measured to confirm that the cells carry enough but not excessive copies, and tests for replication-competent virus, usually on the vector and sometimes on the final cells, are required because a replicating virus would be a serious hazard. Residual beads, cytokines and reagents are also controlled, since these enter the infusion bag with the cells [19, 24, 25].

Potency is the hardest attribute. A useful assay should predict clinical activity, yet cytotoxicity assays against target cells, cytokine release and flow cytometry of transduced cells each capture only part of it. Regulators ask for a potency assay that reflects the mechanism of action, which in practice means a combination of tests whose correlation with response is still being learned [20, 24, 35]. In the absence of such correlation, specification limits may be conservative or arbitrary. Variation between facilities is a regulatory topic in its own right. A commercial product may be made at several sites, each of which must show that it can produce cells meeting specification. Multi-site production helps resilience but multiplies the work of demonstrating comparability, while single-site production is simpler and exposes supply to a single point of failure, such as a contamination event or equipment fault [10, 19, 35]. Retention of vial samples for later testing is limited, so every assay must earn its place.

Regulatory frameworks treat these products as gene therapy medicinal products, or advanced therapy medicinal products in Europe. The European guideline on medicinal products containing genetically modified cells and the US guidance on chemistry, manufacturing and controls for gene therapy investigational applications describe expectations on vector quality, cell characterisation, comparability, stability and testing for adventitious agents [24, 25]. Manufacturers must also show comparability when a process is changed, which is demanding because clinical data are scarce and a small change can alter cell phenotype [11, 35]. Accreditation of clinical centres, such as the Joint Accreditation Committee of the International Society for Cellular Therapy and EBMT, is another layer of quality assurance. It sets standards for cell collection, processing and clinical care, and many authorities now expect accredited or comparable programmes to administer

commercial CAR-T products. Hospitals must invest in staff, training and facilities before they treat their first patient, a hidden cost that rarely appears in price comparisons [8, 35].

Safety systems extend beyond the factory. Cytokine release syndrome and neurological toxicity are the main acute risks, and consensus grading by the American Society for Transplantation and Cellular Therapy gives the field a shared language for severity [26, 27, 36]. Management with tocilizumab and corticosteroids is well described, and best-practice recommendations from European societies set out expectations for centres, including access to intensive care and trained staff [8, 27]. Treatment centres are therefore part of the supply chain, and accreditation takes time and money. Post-authorisation obligations continue for years. Regulators ask for follow-up of treated patients for up to fifteen years to detect delayed adverse events such as secondary malignancy, and registries run by transplant organisations collect outcome data across centres. This long-term surveillance, together with early reports of rare T-cell malignancies in other contexts, keeps attention on vector integration and on the need for traceable product records [24, 25, 35]. Cost of such preparedness is carried by the hospital rather than the manufacturer.

Chain of identity and custody is the quality feature unique to autologous products. A bag labelled with the wrong patient's name could be fatal, so manufacturers use barcodes, tamper-evident seals and electronic tracking from collection to infusion, and sites audit the process. These systems add cost but are not optional. The same infrastructure supports traceability for long-term follow-up, which regulators require for gene-modified cells because of insertional risks [19, 24, 25]. Educating staff in the recognition of toxicity is an important quality element, and the wider team includes nurses, pharmacists and intensivists. Pharmacy services handle receipt of frozen product, thawing close to the bedside, verification of identity and documentation, and preparation of tocilizumab for immediate availability. Experience from large centres suggests that these protocols reduce errors, but the evidence is descriptive [8, 27, 36].

5. COSTS AND ECONOMIC EVALUATION

List prices for the first licensed products were in the range of several hundred thousand US dollars per infusion, and those prices cover only the product. Hernandez and colleagues estimated that total costs of care, including apheresis, conditioning, hospitalisation and management of adverse events, were considerably higher than the list price, and that the cost of managing severe toxicity could be substantial [28]. Estimates from a single country do not carry over to systems with different price structures [28, 32]. Because the first products were priced similarly across manufacturers, comparing them on cost alone was never likely to settle choices; payers have been more interested in whether prices are justified by outcomes. Critics point out that public funding contributed to the early science, and that a higher price for a therapy that depends on a small number of treated patients is difficult to defend without transparent cost data [28, 37]. Prices also differ between countries, and discounts negotiated with payers are usually confidential.

Where does the money go? Manufacturing costs include the vector, reagents, disposables, cleanroom space, quality testing and highly trained staff. Modelling of production costs in an academic non-profit setting suggested that decentralised manufacturing could be done at a fraction of the commercial price, though such estimates exclude research and development costs, regulatory fees and the risk borne by a company that must also pay for failures [9, 37]. Manufacturers argue that prices reflect the economics of a complex product with a small market. Local production can change the picture. In the academic non-profit model described by Ran and colleagues, the manufacturing cost for a decentralised process was a fraction of the commercial price, although the figure depends on assumptions about staff time, throughput and failure rates. A hospital contemplating its own programme must also budget for quality systems, validation, inspections and vector supply, which are easily underestimated [9,

37]. Such caveats should travel with any quoted figure.

Cost-effectiveness analyses have been broadly favourable under stated assumptions. For children with relapsed leukaemia, a US model suggested that tisagenlecleucel could be cost-effective if a proportion of patients are long-term survivors, and for adults with large B-cell lymphoma, models of axicabtagene ciloleucel versus chemotherapy reported incremental cost-effectiveness ratios within commonly cited thresholds [29, 30, 31]. The key assumption is long-term survival, which is extrapolated well beyond the follow-up of the trials. Table 1 lists these studies together with the clinical data on which they rely. Sensitivity analyses in the published models consistently show that the answer depends on the plateau in survival and on the price. If the long-term cure fraction is lower than assumed, or the product price is higher, the ratio moves above conventional willingness-to-pay thresholds. The authors of these models say so openly, and independent assessments have called for caution in applying the results beyond the settings they were built for [29, 30, 31].

Site of care matters. Lyman and colleagues modelled outpatient administration in selected patients with large B-cell lymphoma and found lower costs than inpatient care, mostly through avoiding hospital days, although outpatient care needs close monitoring and rapid access to admission when toxicity arises [32]. Whether such savings can be realised outside well-resourced centres is uncertain. In low- and middle-income settings, the costs of a single product may exceed the annual health budget per head by many multiples [10, 38]. Hospital costs deserve specific attention. A stay on a specialised unit, admission to intensive care for severe toxicity, and the drugs used to manage it can together add substantially to the bill, and the rate of such events differs between products and patient groups. Reducing severe toxicity through earlier treatment of cytokine release is therefore both a clinical and an economic aim [27, 28, 36]. Such arrangements need staffing, bed capacity and reliable community support.

Payment models are evolving. Outcomes-based agreements, in which the manufacturer is paid in full only if the patient responds, and instalment payments spread over several years, have been proposed to reduce the risk to payers [28, 30]. Offshore or regional manufacturing may shift costs, and one modelling study explored how producing in a lower-cost location would change the aggregate cost of goods, although shipping and regulatory considerations limit the savings [38]. Policy makers face an unresolved question about fairness, as one-off prices are hard to compare with chronic therapy. Competing demands within a health system make the decision harder. A one-off expenditure of several hundred thousand dollars for one patient has to be set against other uses of the same funds, and equity concerns arise if only wealthy patients or regions can reach treatment. Some authors argue for national purchasing and a limited number of designated centres, to concentrate expertise and share the load [31, 32, 38].

6. LIMITATIONS AND FUTURE DIRECTIONS

The literature has gaps that matter to this topic. Manufacturing details are largely proprietary, so published information on yields, failure rates and the actual cost of goods is sparse and often derived from modelling or academic-scale processes. Most economic models rest on single-arm trials and on extrapolation of survival. Patients with the poorest fitness or very rapid disease are underrepresented, and real-world outcomes may be less favourable than trial results [4, 28, 34]. Follow-up in the registration trials was short relative to the long horizon over which cost-effectiveness is judged, so the key uncertainty for payers is also the one the data cannot yet resolve. Long-term remission rates and late toxicities will clarify the value, and until they do, every published economic analysis should be read as conditional on stated assumptions [3, 4, 30].

Several technical developments might ease the problem. Automation and closed systems, shorter culture times, point-of-care production in hospital facilities and rapid release tests all aim to reduce time and cost [9, 21, 22, 23]. Allogeneic products from healthy donors, using gene-edited cells to avoid graft-versus-host disease, would allow

batches to be made in advance and shared among patients, and early clinical results in leukaemia are encouraging but very limited [16, 39]. Whether they will match the persistence of autologous cells is not known. Gene-edited universal cells, off-the-shelf products from induced pluripotent stem cells and in vivo gene delivery to T cells are some of the more ambitious proposals, but all remain experimental. They face the same quality, safety and regulatory questions, plus new ones about immune rejection and off-target editing, and their economics are hypothetical until real batches and real outcomes are available [16, 25, 39]. Ethical review of such trials will have to consider long-term follow-up in people who may be cured.

Finally, a word about access. Without regional manufacturing, technology transfer and sustainable payment, the therapy will stay confined to a handful of high-income centres. Academic and non-profit manufacturing is one route towards wider availability, and hospital-based programmes show that it can be done, but they require regulatory capacity and investment that many health systems lack [9, 35, 37]. A final limitation of this review is that it relies on published literature, which lags the pace of industry development. Process changes, new facilities and commercial pricing decisions may not be described in journals until years later, and readers should consult current regulatory documents for the position today.

7. CONCLUSION

CAR-T therapy shows that a living, engineered cell product can produce lasting responses in cancers that resist conventional treatment, and the evidence for this is strong in B-cell leukaemia and lymphoma [3, 4, 5]. The same literature makes plain that the process of making each dose is a major determinant of who benefits. Variable starting cells, viral vector supply, long cultures, manual operations and slow release tests all lengthen the wait and raise the price, while patients with fast-moving disease have little time to spare [10, 11, 21].

Costs follow from this. Published economic models can find the treatment good value, but those results hinge on extrapolated survival and on assumptions about the site of care and the price, and the total cost of treating a patient far exceeds the sticker price [28, 29, 30]. Practical progress is likely to come from closed automated manufacturing, faster release testing, outcomes-based payment and, if they prove durable, allogeneic products [22, 38, 39]. Until then, pharmacists, clinicians and payers must weigh a strong clinical signal against real constraints of capacity and money.

8. ABBREVIATIONS

ASTCT-American Society for Transplantation and Cellular Therapy, BCMA-B-cell maturation antigen, CAR-Chimeric antigen receptor, CRS-Cytokine release syndrome, EMA-European Medicines Agency, FDA-US Food and Drug Administration, GMP-Good manufacturing practice, ICER-Incremental cost-effectiveness ratio, IL-Interleukin, TALEN-Transcription activator-like effector nuclease

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Nil

10. AUTHORS CONTRIBUTIONS

All the authors have contributed equally.

11. CONFLICT OF INTERESTS

Declared none

12. REFERENCES

1. June CH, O'Connor RS, Kawalekar OU, Ghassemi S, Milone MC. CAR T cell immunotherapy for human cancer. Science 2018;359:1361–5.

2. Fesnak AD, June CH, Levine BL. Engineered T cells: the promise and challenges of cancer immunotherapy. Nat Rev Cancer 2016;16:566–81.

3. Maude SL, Laetsch TW, Buechner J, Rives S, Boyer M, Bittencourt H, *et al.* Tisagenlecleucel in children and young adults with B-cell lymphoblastic leukemia. N Engl J Med 2018;378:439–48.

4. Neelapu SS, Locke FL, Bartlett NL, Lekakis LJ, Miklos DB, Jacobson CA, *et al.* Axicabtagene ciloleucel CAR T-cell therapy in refractory large B-cell lymphoma. N Engl J Med 2017;377:2531–44.

5. Schuster SJ, Bishop MR, Tam CS, Waller EK, Borchmann P, McGuirk JP, *et al.* Tisagenlecleucel in adult relapsed or

refractory diffuse large B-cell lymphoma. N Engl J Med 2019;380:45–56.

6. Wang M, Munoz J, Goy A, Locke FL, Jacobson CA, Hill BT, *et al.* KTE-X19 CAR T-cell therapy in relapsed or refractory mantle-cell lymphoma. N Engl J Med 2020;382:1331–42.
7. Munshi NC, Anderson LD, Shah N, Madduri D, Berdeja J, Lonial S, *et al.* Idecabtagene vicleucel in relapsed and refractory multiple myeloma. N Engl J Med 2021;384:705–16.
8. Hayden PJ, Roddie C, Bader P, Basak GW, Bonig H, Bonini C, *et al.* Management of adults and children receiving CAR T-cell therapy: 2021 best practice recommendations of the European Society for Blood and Marrow Transplantation (EBMT) and the Joint Accreditation Committee of ISCT and EBMT (JACIE) and the European Haematology Association (EHA). Ann Oncol 2022;33:259–75.
9. Abou-El-Enein M, Elsallab M, Feldman SA, Fesnak AD, Heslop HE, Marks P, *et al.* Scalable manufacturing of CAR T cells for cancer immunotherapy. Blood Cancer Discov 2021;2:408–22.
10. Levine BL, Miskin J, Wonnacott K, Keir C. Global manufacturing of CAR T cell therapy. Mol Ther Methods Clin Dev 2017;4:92–101.
11. Roddie C, O'Reilly M, Dias Alves Pinto J, Vispute K, Lowdell M. Manufacturing chimeric antigen receptor T cells: issues and challenges. Cytotherapy 2019;21:327–40.
12. Heathman TRJ, Nienow AW, McCall MJ, Coopman K, Kara B, Hewitt CJ. The translation of cell-based therapies: clinical landscape and manufacturing challenges. Regen Med 2015;10:49–64.
13. Locke FL, Miklos DB, Jacobson CA, Perales MA, Kersten MJ, Oluwole OO, *et al.* Axicabtagene ciloleucel as second-line therapy for large B-cell lymphoma. N Engl J Med 2022;386:640–54.
14. Kamdar M, Solomon SR, Arnason J, Johnston PB, Glass B, Bachanova V, *et al.* Lisocabtagene maraleucel versus standard of care with salvage chemotherapy followed by autologous stem cell transplantation as second-line treatment in patients with relapsed or refractory large B-cell lymphoma (TRANSFORM): results from an interim analysis of an open-label, randomised, phase 3 trial. Lancet 2022;399:2294–308.
15. Jackson HJ, Rafiq S, Brentjens RJ. Driving CAR T-cells forward. Nat Rev Clin Oncol 2016;13:370–83.
16. Sadelain M, Rivière I, Riddell S. Therapeutic T cell engineering. Nature 2017;545:423–31.
17. Wang X, Rivière I. Clinical manufacturing of CAR T cells: foundation of a promising therapy. Mol Ther Oncolytics 2016;3:16015.
18. Vormittag P, Gunn R, Ghorashian S, Veraitch FS. A guide to manufacturing CAR T cell therapies. Curr Opin Biotechnol 2018;53:164–81.
19. Gee AP. GMP CAR-T cell production. Best Pract Res Clin Haematol 2018;31:126–34.
20. Fraietta JA, Lacey SF, Orlando EJ, Pruteanu-Malinici I, Gohil M, Lundh S, *et al.* Determinants of response and resistance to CD19 chimeric antigen receptor (CAR) T cell therapy of chronic lymphocytic leukemia. Nat Med 2018;24:563–71.
21. Ghassemi S, Nunez-Cruz S, O'Connor RS, Fraietta JA, Patel PR, Scholler J, *et al.* Reducing ex vivo culture improves the antileukemic activity of chimeric antigen receptor (CAR) T cells. Cancer Immunol Res 2018;6:1100–9.
22. Kaiser AD, Assenmacher M, Schröder B, Meyer M, Orentas R, Bethke U, *et al.* Towards a commercial process for the manufacture of genetically modified T cells for therapy. Cancer Gene Ther 2015;22:72–8.
23. Lock D, Mockel-Tenbrinck N, Drechsel K, Barth C, Mauer D, Schaser T, *et al.* Automated manufacturing of potent CD20-directed chimeric antigen receptor T cells for clinical use. Hum Gene Ther 2017;28:914–25.
24. US Food and Drug Administration. Chemistry, manufacturing, and control (CMC) information for human gene therapy investigational new drug applications (INDs): guidance for industry. Silver Spring: FDA; 2020.
25. European Medicines Agency. Guideline on quality, non-clinical and clinical aspects of medicinal products containing genetically modified cells. London: EMA; 2012.
26. Lee DW, Santomasso BD, Locke FL, Ghobadi A, Turtle CJ, Brudno JN, *et al.* ASTCT consensus grading for cytokine release syndrome and neurologic toxicity associated with immune effector cells. Biol Blood Marrow Transplant 2019;25:625–38.

27. Neelapu SS, Tummala S, Kebriaei P, Wierda W, Gutierrez C, Locke FL, *et al.* Chimeric antigen receptor T-cell therapy – assessment and management of toxicities. Nat Rev Clin Oncol 2018;15:47–62.

28. Hernandez I, Prasad V, Gellad WF. Total costs of chimeric antigen receptor T-cell immunotherapy. JAMA Oncol 2018;4:994–6.

29. Whittington MD, McQueen RB, Ollendorf DA, Tice JA, Chapman RH, Pearson SD, *et al.* Long-term survival and cost-effectiveness associated with axicabtagene ciloleucel vs chemotherapy for treatment of B-cell lymphoma. JAMA Netw Open 2019;2:e190035.

30. Whittington MD, McQueen RB, Ollendorf DA, Tice JA, Chapman RH, Pearson SD, *et al.* Long-term survival and value of chimeric antigen receptor T-cell therapy for pediatric patients with relapsed or refractory leukemia. JAMA Pediatr 2018;172:1161–8.

31. Roth JA, Sullivan SD, Lin VW, Bansal A, Purdum AG, Navale L, *et al.* Cost-effectiveness of axicabtagene ciloleucel for adult patients with relapsed or refractory large B-cell lymphoma in the United States. J Med Econ 2018;21:1238–45.

32. Lyman GH, Nguyen A, Snyder S, Gitlin M, Chung KC. Economic evaluation of chimeric antigen receptor T-cell therapy by site of care among patients with relapsed or refractory large B-cell lymphoma. JAMA Netw Open 2020;3:e202072.

33. Stroncek DF, Ren J, Lee DW, Tran M, Frodigh SE, Sabatino M, *et al.* Myeloid cells in peripheral blood mononuclear cell concentrates inhibit the expansion of chimeric antigen receptor T cells. Cytotherapy 2016;18:893–901.

34. Abramson JS, Palomba ML, Gordon LI, Lunning MA, Wang M, Arnason J, *et al.* Lisocabtagene maraleucel for patients with relapsed or refractory large B-cell lymphomas (TRANSCEND NHL 001): a multicentre seamless design study. Lancet 2020;396:839–52.

35. Hartmann J, Schüßler-Lenz M, Bondanza A, Buchholz CJ. Clinical development of CAR T cells – challenges and opportunities in translating innovative treatment concepts. EMBO Mol Med 2017;9:1183–97.

36. Brudno JN, Kochenderfer JN. Toxicities of chimeric antigen receptor T cells: recognition and management. Blood 2016;127:3321–30.

37. Ran T, Eichmüller SB, Schmidt P, Schlander M. Cost of decentralized CAR T-cell production in an academic nonprofit setting. Int J Cancer 2020;147:3438–45.

38. Harrison RP, Zylberberg E, Ellison S, Levine BL. Chimeric antigen receptor-T cell therapy manufacturing: modelling the effect of offshore production on aggregate cost of goods. Cytotherapy 2019;21:224–33.

39. Qasim W, Zhan H, Samarasinghe S, Adams S, Amrolia P, Stafford S, *et al.* Molecular remission of infant B-ALL after infusion of universal TALEN gene-edited CAR T cells. Sci Transl Med 2017;9:eaaj2013.