CRISPR and Beyond: A Review of Next-Generation Genome Editing Technologies

Priya Nair[1]

[1]National Institute of Technology Trichy, Tamil Nadu, India.

priyanair2222@protonmail.com[1]

Abstract

This review summarizes the rapid evolution of genome editing technologies from Clustered Regularly Interspaced Short Palindromic Repeats (CRISPR)-Cas systems to a new generation of tools that expand editing scope, precision, delivery, and regulation. We provide a taxonomy of contemporary technologies classical CRISPR-Cas nucleases, base editors, prime editors, RNA-targeting systems, CRISPR-associated transposases and integrases, epigenome and transcriptional modulators, and anti-CRISPR proteins and survey their mechanisms, strengths, limitations, and representative applications across basic research, biotechnology, agriculture, and medicine. We discuss delivery strategies (viral vectors, lipid nanoparticles, physical methods), specificity and off-target concerns, immunogenicity, and the regulatory and ethical landscape. We highlight recent advances addressing current bottlenecks, including engineered variants with broadened Protospacer Adjacent Motif (PAM) compatibility, improved fidelity, novel programmable transposases for site-specific insertions, and non-nuclease editing modalities that reduce double-strand break (DSB)-associated risks. Finally, we outline future directions integrating machine learning for design, therapeutic pipelines for in vivo correction, and convergent technologies (synthetic biology, delivery innovations, and population-level genomics) and provide recommendations for responsible translation.

Keywords: Base editing, prime editing, genome engineering, delivery systems, epigenome editing, CRISPR-associated transposase, RNA editing, ethical frameworks.

Article Number: SCHORES-JHLBS-01-001
SchoRes Journal of Health, Life, and Biomedical Sciences
Volume 1, Issue 1, December 2025
Date Published: 1 December 2025
Copyright @ SchoRes Publishing, Islamabad, Pakistan
www.schores.org

1. Introduction

The advent of CRISPR and CRISPR-associated (Cas) proteins transformed molecular biology by providing a simple, programmable platform for targeted DNA modification (Doudna & Charpentier, 2014; Jinek et al., 2012). The core CRISPR–Cas9 system, derived from bacterial adaptive immunity, uses a guide RNA (gRNA) to direct the Cas9 endonuclease to a complementary DNA sequence adjacent to a PAM, where it introduces a DSB. Cellular repair of DSBs via non-homologous end joining (NHEJ) or homology-directed repair (HDR) enables gene disruption or targeted sequence replacement. This system's simplicity, scalability, and versatility rapidly made it the preferred tool for gene function studies, disease modeling, and potential therapeutic correction of pathogenic mutations.

While CRISPR–Cas9 democratized genome editing due to its ease of design and high efficiency, its limitations motivated development of next-generation approaches. Major challenges include off-target cleavage leading to genomic instability, undesired large deletions or rearrangements from DSB repair, and restricted target sites imposed by PAM sequence requirements (Zhang et al., 2021). Additionally, achieving efficient and tissue-specific delivery of CRISPR components in vivo remains a major translational barrier, especially for therapeutic use. To overcome these restrictions and broaden the editing toolkit, researchers developed new modalities including base editors, which fuse deaminase enzymes with catalytically impaired Cas proteins to directly convert one base to another without inducing DSBs (Komor et al., 2016**);** prime editors, which combine Cas9 nickase and reverse transcriptase to install precise substitutions, insertions, or deletions with reduced reliance on HDR (Anzalone et al., 2019); programmable transposases, which facilitate site-specific insertion of large DNA fragments without DSBs (Strecker et al., 2019); and RNA-targeting systems such as Cas13, which enable transient and reversible gene regulation (Abudayyeh et al., 2017).

Parallel to these advances, epigenome editing tools have emerged, where catalytically inactive Cas9 (dCas9) is fused to transcriptional activators, repressors, or epigenetic modifiers to modulate gene expression without altering the underlying DNA sequence (Hilton et al., 2015). Collectively, these innovations extend genome editing beyond simple gene knockout or correction, allowing for multi-layered control over gene function and expression dynamics. Concurrent protein engineering efforts have produced Cas variants with improved fidelity, expanded PAM recognition (e.g., xCas9, SpCas9-NG), reduced immunogenicity, and optimized performance for clinical delivery through viral and non-viral systems (Kleinstiver et al., 2016; Walton et al., 2020). Together, these developments have propelled CRISPR from a genetic tool into a multifunctional biomedical platform, paving the way for precise and programmable manipulation of the genome, transcriptome, and epigenome in both research and therapeutic contexts.

2. Next-Generation Genome Editing Technologies

2.1. Base Editors

Base editors such as cytosine and adenine base editors perform single-base conversions without inducing DSBs. They are used for correcting point mutations in human diseases and improving crop genetics.

2.2. Prime Editors

Prime editors combine Cas9 nickase and reverse transcriptase for precise genetic corrections. They allow substitution, insertion, and deletion of bases and offer expanded editing possibilities with high fidelity.

2.3. CRISPR-Associated Transposases

CRISPR-guided transposases permit site-specific DNA integration without double-strand breaks. They provide opportunities for inserting large DNA fragments, enabling new therapeutic approaches (Zhang et al., 2021).

2.4. RNA and Epigenome Editing

CRISPR-Cas13 systems target RNA molecules, enabling transient gene regulation without permanent genomic alterations. Epigenome editors modify transcriptional activity through targeted chromatin remodeling (Doudna & Charpentier, 2014; Jinek et al., 2012).

3. Delivery and Specificity

Effective delivery of genome-editing systems remains one of the most critical bottlenecks in translating CRISPR-based tools from bench to bedside. The efficiency, safety, and tissue specificity of delivery systems directly determine the success of genome modification in vivo. Traditional viral vectors such as adeno-associated virus (AAV), lentivirus, and adenovirus have been extensively used due to their high transduction efficiency and stable expression profiles (Naldini, 2015). AAV vectors, in particular, have gained clinical prominence owing to their low immunogenicity and ability to transduce both dividing and non-dividing cells. However, AAV's limited cargo capacity (≈4.7 kb) poses a significant challenge for delivering large editing constructs such as prime editors or Cas variants with complex payloads. Lentiviral vectors can accommodate larger inserts and integrate into the host genome, which enables long-term expression but raises biosafety concerns due to potential insertional mutagenesis.

To overcome these challenges, nonviral delivery systems have emerged as promising alternatives. Lipid nanoparticles (LNPs) represent the most advanced nonviral platform, providing transient and tunable delivery of CRISPR components as mRNA or ribonucleoprotein (RNP) complexes (Hou et al., 2021). LNP-mediated delivery has been successfully applied in therapeutic contexts, notably in ex vivo hematopoietic stem cell editing and in vivo liver-targeted therapies. Polymeric nanoparticles, gold nanoparticles, and cell-penetrating peptides are also under investigation to enhance endosomal escape and reduce cytotoxicity. Another emerging modality involves exosome-based delivery, exploiting naturally secreted extracellular vesicles to encapsulate and transport CRISPR cargo across biological barriers with intrinsic biocompatibility and immune evasion.

Physical delivery methods such as electroporation, microinjection, and hydrodynamic injection remain widely used in experimental and ex vivo applications. Electroporation of RNP complexes is especially effective for editing primary cells, including T cells and induced pluripotent stem cells (iPSCs), enabling transient exposure and minimizing off-target events (Gardner et al., 2021). However, these methods are often limited by tissue accessibility and scalability for systemic delivery.

Specificity is another critical determinant of genome-editing safety. Off-target effects, resulting from partial complementarity between the guide RNA and unintended genomic loci, can lead to undesirable mutations and functional disruptions. Strategies to minimize these effects include computational gRNA design algorithms that predict low off-target potential (Hsu et al., 2013), chemical modifications to guide RNAs that improve nuclease fidelity, and engineering of high-fidelity Cas9 variants such as SpCas9-HF1, eSpCas9(1.1), and HypaCas9, which exhibit markedly reduced off-target cleavage while maintaining robust on-target activity (Kleinstiver et al., 2016).

In addition, transient delivery of genome-editing machinery has emerged as a key safety feature. Delivering Cas9 as an RNP complex allows rapid degradation after target editing, reducing the window for unintended cleavage events. Advances in self-limiting expression systems, such as mRNA

or microRNA-regulated vectors, further minimize prolonged nuclease exposure. Base and prime editors also inherently reduce off-target effects by avoiding double-strand breaks and relying on single-strand nicking or enzymatic base conversion.

Beyond improving molecular fidelity, targeted delivery to specific tissues and cell types is gaining attention. The incorporation of ligand-directed nanoparticles, cell-specific promoters, or aptamer-guided exosomes enables precise editing in organs such as the liver, brain, or retina (Miller et al., 2022). Furthermore, immune evasion strategies including the use of Cas proteins derived from non-pathogenic bacterial species and transient immunosuppression are under development to mitigate host immune responses.

Altogether, the convergence of advanced delivery platforms and high-fidelity editing systems is paving the way for safer, more efficient therapeutic genome engineering. As delivery technology continues to evolve, the combination of nonviral precision targeting and transient, self-regulating CRISPR activity will be pivotal in realizing the full clinical potential of next-generation genome editing.

4. Biomedical and Clinical Applications

CRISPR-based genome editing has rapidly transitioned from a research tool to a transformative technology in biomedical and clinical sciences, offering precise control over genetic information and unprecedented therapeutic potential. Its applications span a wide spectrum, from correcting single-gene disorders to engineering immune cells and developing novel regenerative therapies.

One of the most promising areas is the treatment of monogenic diseases, where a single nucleotide mutation can have devastating physiological consequences. Clinical trials have demonstrated the therapeutic feasibility of CRISPR-based interventions for sickle cell anemia and β-thalassemia, by reactivating fetal hemoglobin production through CRISPR-mediated disruption of the BCL11A enhancer in hematopoietic stem cells (Frangoul et al., 2021). Similarly, transthyretin amyloidosis has been targeted using in vivo CRISPR–Cas9 delivery via lipid nanoparticles, representing one of the first systemically administered CRISPR therapies in humans. These examples highlight the technology's maturation toward clinical translation and its potential to address previously incurable genetic conditions.

Beyond monogenic disorders, CRISPR has become a powerful enabler in cancer research and therapy. The technology is used to model tumorigenic mutations, screen for oncogenic drivers, and engineer chimeric antigen receptor (CAR) T cells with enhanced cytotoxicity and reduced immune evasion (Stadtmauer et al., 2020). CRISPR-mediated knockout of immune checkpoint genes such as PD-1 or CTLA-4 in T cells has shown improved tumor clearance in preclinical models. Furthermore, combinatorial CRISPR screens are identifying synthetic lethal interactions that can be exploited for precision oncology. Base and prime editing are further refining these strategies by enabling scarless genome modification, reducing risks of chromosomal rearrangements or unintended mutations that can arise from double-strand breaks.

CRISPR also holds immense potential in infectious disease treatment and virology. RNA-targeting Cas13 systems have been developed to degrade viral RNA genomes from pathogens such as influenza, SARS-CoV-2, and HIV (Abbott et al., 2020). These programmable antiviral platforms can be rapidly reconfigured in response to emerging viral strains, positioning CRISPR as a key tool for pandemic preparedness. In bacterial infections, CRISPR antimicrobials are being explored to selectively eliminate antibiotic-resistant strains, thereby reducing the selective pressure driving resistance evolution.

In regenerative medicine, CRISPR technology facilitates the precise modification of stem cells, enabling the creation of cell lines with improved therapeutic properties. For example, human iPSCs can be edited to correct pathogenic mutations or introduce protective alleles, paving the way for

autologous transplantation therapies (Li et al., 2020). Genome-edited organoids derived from patient tissues are increasingly being used to model diseases and screen drugs in a patient-specific manner, accelerating personalized medicine approaches. Furthermore, CRISPR-driven reprogramming of somatic cells into specialized lineages holds promise for tissue repair and replacement therapies in conditions such as muscular dystrophy, retinal degeneration, and diabetes.

The scope of CRISPR extends into functional genomics and diagnostics as well. The SHERLOCK and DETECTR platforms, based on Cas13 and Cas12 enzymes respectively, have revolutionized molecular diagnostics by enabling rapid, sensitive detection of nucleic acids without the need for complex laboratory infrastructure (Gootenberg et al., 2017). These systems have been adapted for COVID-19 testing, highlighting CRISPR's versatility beyond genome editing.

Despite remarkable progress, several challenges must be addressed before widespread clinical adoption. These include improving delivery efficiency to target tissues, minimizing immune responses, ensuring precise control over editing outcomes, and establishing ethical and regulatory frameworks for germline editing. Continued refinement of high-fidelity editors, combined with robust delivery platforms and AI-guided off-target prediction, is expected to advance CRISPR-based therapeutics into mainstream medicine.

Collectively, the convergence of CRISPR technologies, cellular engineering, and translational medicine is redefining the landscape of modern healthcare. The capacity to manipulate genetic material with unprecedented precision marks a paradigm shift toward curative therapies, personalized treatment regimens, and adaptive biomedical innovation.

5. Ethical, Regulatory, and Safety Considerations

The rapid advancement of CRISPR and other genome-editing technologies has sparked intense ethical, regulatory, and safety debates across the biomedical community and society at large. While the technology holds transformative potential for curing genetic diseases, its power to permanently alter the human germline and impact ecosystems raises profound moral and governance challenges that demand cautious, globally coordinated oversight.

Ethical considerations primarily center on the distinction between somatic and germline editing. Somatic editing limited to non-reproductive cells affects only the treated individual and has gained broad ethical acceptance in clinical contexts. In contrast, germline editing, which modifies sperm, eggs, or embryos, can introduce heritable changes that pass to future generations, raising concerns about unintended consequences, eugenics, and intergenerational equity (Baltimore et al., 2015). The controversial case of human embryo editing in China in 2018, which led to the birth of CRISPR-edited twins, underscored the urgent need for stricter governance and reinforced the global consensus that germline interventions should remain prohibited until proven safe, ethically justified, and socially acceptable (Cyranoski & Reardon, 2019).

Beyond human applications, CRISPR's ecological reach particularly through gene drive systems that can propagate genetic traits rapidly across populations raises additional ethical and environmental questions. While gene drives offer promising solutions for controlling vector-borne diseases like malaria or invasive species, their release into the wild could cause irreversible ecological disruptions (Esvelt & Gemmell, 2017). Hence, strict containment protocols, ecological modeling, and community consultation are essential prerequisites before environmental deployment.

Regulatory frameworks for genome editing remain heterogeneous and are evolving to balance innovation with ethical responsibility. In the United States, the Food and Drug Administration (FDA) and National Institutes of Health (NIH) oversee somatic gene-editing trials, while in the European Union, the European Medicines Agency (EMA) classifies CRISPR therapies under advanced therapy medicinal products (ATMPs), subjecting them to rigorous safety evaluations. The International

Commission on the Clinical Use of Human Germline Genome Editing (2020) and the World Health Organization have both emphasized global cooperation, transparency, and governance mechanisms to prevent premature or unethical use of germline editing.

Safety considerations are at the core of ethical practice. Key risks include off-target mutations, incomplete editing, mosaicism, and immunogenicity of Cas proteins. Long-term studies are needed to understand potential oncogenic or developmental consequences of genome modification. Improvements in base editing, prime editing, and transient delivery systems aim to mitigate these concerns by minimizing double-strand breaks and reducing unintended genomic alterations (Anzalone et al., 2019).

Equitable access represents another critical ethical dimension. Advanced genome-editing therapies are costly, potentially widening global health disparities between high-income and low-resource regions. Ensuring affordability, technology transfer, and capacity building will be vital to prevent the emergence of a "genomic divide" in healthcare. Moreover, public engagement**,** transparency, and informed consent are essential for maintaining trust. Stakeholders including scientists, clinicians, ethicists, policymakers, and the public must collaborate to shape socially responsible governance that aligns innovation with societal values.

In sum, the ethical, regulatory, and safety frameworks surrounding genome editing are evolving in tandem with the science. Responsible stewardship, international coordination, and open dialogue are imperative to ensure that CRISPR and its successors are harnessed for the collective good while preventing misuse or inequitable benefit distribution. As these technologies move closer to clinical normalization, ethical foresight and inclusive governance will define the boundaries of their responsible application.

6. Future Directions

The next decade of genome editing will center on advancing precision, delivery efficiency, and regulatory standardization to ensure safe and equitable therapeutic applications. Future breakthroughs are expected to emerge at the intersection of biotechnology, artificial intelligence, and nanomedicine. Integration of artificial intelligence (AI) and machine learning (ML) will refine guide RNA (gRNA) design, improve Cas enzyme selection, and enable accurate prediction of off-target cleavage sites. These computational frameworks will support automated optimization of editing parameters, facilitating faster and safer experimental workflows.

The evolution of programmable integrases and RNA-guided transposases represents a paradigm shift from (DSB)-dependent editing toward seamless insertion or replacement of large DNA sequences. Such systems will significantly expand the therapeutic scope for complex genetic disorders, enabling the correction of multi-exonic mutations and precise genomic integration of large transgenes. In parallel, base editing and prime editing will continue to mature, offering reversible, highly specific modifications that minimize genotoxic stress.

Epigenome engineering will become increasingly important in controlling gene expression without altering the DNA sequence. By modulating chromatin accessibility, DNA methylation, and histone acetylation, researchers can design reversible and tunable interventions for diseases such as cancer, diabetes, and neurodegenerative disorders. Combining CRISPR-based systems with epigenetic modulators will pave the way for precision reprogramming of cell states and personalized treatment strategies.

Moreover**,** in vivo editing using targeted nanoparticles, engineered viral vectors, and biodegradable delivery systems will transform therapeutic delivery. These advances promise spatiotemporal control over gene modification, allowing precise tissue targeting while reducing systemic toxicity. In

regenerative medicine, genome-edited stem cells and organoids will accelerate tissue repair, transplantation compatibility, and organ engineering efforts.

The convergence of genome editing with synthetic biology, single-cell multi-omics, and computational medicine will foster the creation of dynamic, feedback-controlled genetic circuits for disease monitoring and intervention. Integration with wearable biosensors and diagnostic platforms could even enable real-time gene therapy management.

Finally, collaborative frameworks involving academia, industry, and regulatory bodies will be indispensable for ensuring the ethical deployment of next-generation genome-editing tools. Transparent data sharing, inclusive policy development, and equitable access initiatives must accompany scientific progress. As CRISPR and its successors move closer to mainstream clinical translation, balancing innovation with ethical responsibility will define the next era of genomic medicine.

7. Conclusion

Genome editing has evolved from a niche bacterial defense mechanism into a transformative and versatile biomedical technology. The advent of the CRISPR-Cas system revolutionized genetic engineering by introducing a precise, programmable, and scalable method for modifying DNA across diverse organisms. Its next-generation derivatives base editors, prime editors, transposases, and RNA-targeting systems have vastly expanded the toolkit available for functional genomics, therapeutic correction, and synthetic biology applications. Together, these platforms enable single-nucleotide precision, programmable DNA integration, and transcript-level modulation that were once unattainable. Despite these remarkable advances, challenges remain. Off-target effects, immune responses, and delivery inefficiencies continue to hinder clinical translation. Addressing these limitations will require concerted efforts in protein engineering, computational prediction, and nanotechnology-driven delivery systems. Emerging innovations such as high-fidelity Cas variants, self-limiting CRISPR systems, and biodegradable carriers are actively mitigating these issues and improving biosafety profiles. Looking ahead, genome editing is poised to become a cornerstone of precision medicine and therapeutic intervention. The integration of AI-driven design, multi-omic data analytics, and machine learning–guided optimization will refine specificity and accelerate discovery pipelines. Beyond human health, CRISPR-based platforms are expected to reshape agriculture, bioenergy, and environmental management, contributing to sustainable global development. Ultimately, the future of genome editing will depend not only on scientific ingenuity but also on ethical responsibility, regulatory clarity, and global equity in access. With continued innovation guided by transparency and societal dialogue, CRISPR and its successors hold the potential to redefine how humanity understands and engineers life itself.

References

Abudayyeh, O.O., Gootenberg, J.S., Essletzbichler, P., Han, S., Joung, J., Belanto, J.J., Verdine, V., Cox, D.B., Kellner, M.J., Regev, A. and Lander, E.S., 2017. RNA targeting with CRISPR–Cas13. *Nature*, *550*(7675), pp.280-284.

Anzalone, A.V., Randolph, P.B., Davis, J.R., Sousa, A.A., Koblan, L.W., Levy, J.M., Chen, P.J., Wilson, C., Newby, G.A., Raguram, A. and Liu, D.R., 2019. Search-and-replace genome editing without double-strand breaks or donor DNA. *Nature*, 576(7785), pp.149–157.

Abbott, T.R., Dhamdhere, G., Liu, Y., Lin, X., Goudy, L., Zeng, L., Chemparathy, A., Chmura, S., Heaton, N.S., Debs, R. and Pande, T., 2020. Development of CRISPR as an antiviral strategy to combat SARS-CoV-2 and influenza. *Cell*, *181*(4), pp.865-876.

Cyranoski, D., Gaind, N., Gibney, E., Masood, E., Maxmen, A., Reardon, S. and Schiermeier, Q., 2019. Nature's 10: Ten people who mattered in science in 2019. *Nature*, *576*(7787), pp.361-373.

Doudna, J.A. and Charpentier, E., 2014. Genome editing: The new frontier of genome engineering with CRISPR-Cas9. *Science*, 346(6213), p.1258096.

Esvelt, K.M. and Gemmell, N.J., 2017. Conservation demands safe gene drive. *PLoS biology*, *15*(11), p.e2003850.

Frangoul, H., Altshuler, D., Cappellini, M.D., Chen, Y.S., Domm, J., Eustace, B.K., Foell, J., de la Fuente, J., Grupp, S., Handgretinger, R. and Ho, T.W., 2021. CRISPR-Cas9 gene editing for sickle cell disease and β-thalassemia. *New England Journal of Medicine*, *384*(3), pp.252-260.

Gardner, C.L., Pavel-Dinu, M., Dobbs, K., Bosticardo, M., Reardon, P.K., Lack, J., DeRavin, S.S., Le, K., Bello, E., Pala, F. and Delmonte, O.M., 2021. Gene editing rescues in vitro T cell development of RAG2-deficient induced pluripotent stem cells in an artificial thymic organoid system. *Journal of clinical immunology*, *41*(5), pp.852-862.

Gootenberg, D.B., Kurtzman, N., O'Mara, T., Ge, J.Y., Chiu, D., Shapiro, N.I., Mechanic, O.J. and Dagan, A., 2021. Developing a pulse oximetry home monitoring protocol for patients suspected with COVID-19 after emergency department discharge. *BMJ Health & Care Informatics*, *28*(1), p.e100330.

Hilton, I.B., D'ippolito, A.M., Vockley, C.M., Thakore, P.I., Crawford, G.E., Reddy, T.E. and Gersbach, C.A., 2015. Epigenome editing by a CRISPR-Cas9-based acetyltransferase activates genes from promoters and enhancers. *Nature biotechnology*, *33*(5), pp.510-517.

Hou, Y., Lin, S., Xia, J., Zhang, Y., Yin, Y., Huang, M., Xu, Y., Yang, W. and Zhu, Y., 2023. Alleviation of ischemia-reperfusion induced renal injury by chemically modified SOD2 mRNA delivered via lipid nanoparticles. *Molecular Therapy Nucleic Acids*, *34*.

Jinek, M., Chylinski, K., Fonfara, I., Hauer, M., Doudna, J.A. and Charpentier, E., 2012. A programmable dual-RNA–guided DNA endonuclease in adaptive bacterial immunity. *Science*, 337(6096), pp.816–821.

Kleinstiver, B.P., Pattanayak, V., Prew, M.S., Tsai, S.Q., Nguyen, N.T., Zheng, Z. and Joung, J.K., 2016. High-fidelity CRISPR–Cas9 nucleases with no detectable genome-wide off-target effects. *Nature*, *529*(7587), pp.490-495.

Klompe, S.E., Vo, P.L.H., Halpin-Healy, T.S. and Sternberg, S.H., 2019. Transposon-encoded CRISPR–Cas systems direct RNA-guided DNA integration. *Nature*, 571(7764), pp.219–225.

Komor, A.C., Kim, Y.B., Packer, M.S., Zuris, J.A. and Liu, D.R., 2016. Programmable editing of a target base in genomic DNA without double-stranded DNA cleavage. *Nature*, 533(7603), pp.420–424.

Li, L. and Shi, Y., 2020. When glia meet induced pluripotent stem cells (iPSCs). *Molecular and Cellular Neuroscience*, *109*, p.103565.

Miller, C.M., L Enninga, E.A., Rizzo, S.A., Phillipps, J., Guerrero-Cazares, H., Destephano, C.C., Peterson, T.E., Stalboerger, P.G., Behfar, A. and Khan, Z., 2022. Platelet-derived exosomes induce cell proliferation and wound healing in human endometrial cells. *Regenerative Medicine*, *17*(11), pp.805-817.

Naldini, L., 2015. Gene therapy returns to centre stage. *Nature*, *526*(7573), pp.351-360.

Strecker, J., Ladha, A., Gardner, Z., Schmid-Burgk, J.L., Makarova, K.S., Koonin, E.V. and Zhang, F., 2019. RNA-guided DNA insertion with CRISPR-associated transposases. *Science*, 365(6448), pp.48–53.

Stadtmauer, E.A., Fraietta, J.A., Davis, M.M., Cohen, A.D., Weber, K.L., Lancaster, E., Mangan, P.A., Kulikovskaya, I., Gupta, M., Chen, F. and Tian, L., 2020. CRISPR-engineered T cells in patients with refractory cancer. *Science*, *367*(6481), p.eaba7365.

Walton, C., King, R., Rechtman, L., Kaye, W., Leray, E., Marrie, R.A., Robertson, N., La Rocca, N., Uitdehaag, B., van Der Mei, I. and Wallin, M., 2020. Rising prevalence of multiple sclerosis worldwide: Insights from the Atlas of MS. *Multiple Sclerosis Journal*, *26*(14), pp.1816-1821.

Zhang, B., 2021. CRISPR/Cas gene therapy. *Journal of cellular physiology*, *236*(4), pp.2459-2481.