IJARSCT
International Journal of Advanced Research in Science, Communication and Technology
International Open-Access, Double-Blind, Peer-Reviewed, Refereed, Multidisciplinary Online Journal
ISSN: 2581-9429
Volume 6, Issue 10, May 2026
Impact Factor: 8.2

CRISPR-Cas9 in Rare Genetic Disease Treatment: Mechanisms, Applications, and Future Perspectives

Aanchal Sharma
Amity Institute of Biotechnology
Amity University, Uttar Pradesh, Lucknow Campus, Lucknow, Uttar Pradesh, India
Correspondence: Amity Institute of Biotechnology, Amity University UP, Lucknow Campus

Abstract: *Rare genetic diseases are a major challenge for modern medicine due to their heterogeneous etiology, limited therapeutic options, and frequently life-threatening consequences, affecting more than 300 million people worldwide. The emergence of CRISPR-Cas9 (Clustered Regularly Interspaced Short Palindromic Repeats-CRISPR associated protein 9) genome editing technology has revolutionized the field of molecular medicine, providing unprecedented precision and flexibility in modifying the human genome. CRISPR-Cas9 was originally described as a prokaryotic adaptive immune mechanism, that has been adapted into a potent therapeutic tool, capable of correcting disease-causing mutations at the genome level. This review provides a detailed overview of the CRISPR-Cas9 system, including its discovery, molecular structure, principles of guide RNA design, delivery strategies and new variants such as base editors and prime editors. The epidemiology and molecular basis of rare genetic diseases are discussed and conventional treatment modalities such as enzyme replacement therapy, antisense oligonucleotide therapy and viral vector-based gene therapy are critically appraised. This review summarizes the current preclinical and clinical applications of CRISPR-Cas9 in a broad spectrum of rare monogenic disorders including sickle cell disease, beta-thalassemia, Duchenne muscular dystrophy, Huntington's disease, cystic fibrosis, hemophilia A and B, and transthyretin amyloidosis. We also discuss the diagnostic applications of CRISPR including the SHERLOCK and DETECTR platforms. A critical analysis of key challenges including off-target mutagenesis, immunogenicity, delivery barriers and ethical considerations. The review ends with a look to the future of CRISPR-based medicine, toward personalized genomic therapy.*

Keywords: CRISPR-Cas9, rare genetic diseases, genome editing, gene therapy, sickle cell disease, Duchenne muscular dystrophy, base editing, prime editing, off-target effects, personalized medicine

INTRODUCTION

The idea of disease correction at the level of DNA has long been a fascination of biomedical science. There are more than 7,000 unique rare genetic diseases, which are defined by most regulatory agencies as conditions affecting less than 1 in 2,000 people in Europe or less than 200,000 people in the United States, and together they impact an estimated 300–400 million people worldwide (Boycott et al., 2013; National Organization for Rare Disorders, 2022). About 80% of rare diseases are monogenic in origin and are, in principle, amenable to correction by precise genomic intervention (Uddin et al., 2020). However, despite this theoretical tractability, fewer than 5% of rare diseases have any approved pharmacological treatment and many available therapies are palliative rather than curative (Richter et al., 2022).edicine towards personalized genomic therapyClassical gene therapy approaches, such as delivery of functional gene copies via retroviral and adeno-associated viral (AAV) vectors, provided proof-of-concept, but were plagued by the risk of insertional mutagenesis and constraints on cargo size and stable expression (Naldini, 2015; Verma & Somia, 1997).

IJARSCT
International Journal of Advanced Research in Science, Communication and Technology
International Open-Access, Double-Blind, Peer-Reviewed, Refereed, Multidisciplinary Online Journal
ISSN: 2581-9429
Volume 6, Issue 10, May 2026
Impact Factor: 8.2

The development of programmable nucleases, zinc finger nucleases (ZFNs) and transcription activator-like effector nucleases (TALENs), allowed for increased specificity but were still difficult to design and costly to produce at scale (Porteus, 2019). The discovery and engineering of the CRISPR-Cas9 system was a paradigm shift, allowing genome editing in a much simpler, more efficient and more adaptable way than previous technologies (Doudna & Charpentier, 2011)

CRISPR (Clustered Regularly Interspaced Short Palindromic Repeats) was first described as a feature of bacterial genomes by Mojica et al. (2005) and Bolotin et al. (2005), who noted repetitive DNA sequences interspersed with unique spacers derived from bacteriophage genomes. Marraffini and Sontheimer (2008) demonstrated that CRISPR provides adaptive immunity against mobile genetic elements in bacteria. The seminal biochemical reconstitution of the CRISPR-Cas9 system as a programmable RNA-guided endonuclease was achieved by Jinek et al. (2012), establishing that a single guide RNA (sgRNA) could direct Cas9 to any genomic locus flanked by an appropriate protospacer adjacent motif (PAM). Concurrent publications by Cong et al. (2013) and Mali et al. (2013) rapidly extended this system to mammalian genome editing, ushering in the modern era of CRISPR medicine.

Within a decade of these discoveries, CRISPR-Cas9-based therapeutics have entered clinical trials for sickle cell disease and beta-thalassemia, with Casgevy (exagamglogene autotemcel, exa-cel) receiving landmark approval by the U.S. Food and Drug Administration in late 2023 as the first CRISPR medicine approved for human use (Frangoul et al., 2021). In parallel, in vivo CRISPR therapies targeting transthyretin amyloidosis demonstrated striking efficacy in phase 3 trials (Gillmore et al., 2021). These clinical milestones validate CRISPR-Cas9 as a genuine therapeutic modality and signal a transformative moment in the treatment of rare genetic diseases.

This review synthesizes current knowledge on CRISPR-Cas9 technology and its application to rare genetic diseases. We discuss the molecular mechanism of CRISPR-Cas9, evolving genome-editing variants, delivery platforms, the landscape of rare monogenic diseases, conventional therapeutic strategies, and the growing body of preclinical and clinical data supporting CRISPR-based intervention. Challenges including off-target editing, immunological barriers, and regulatory and ethical considerations are examined in depth, alongside emerging directions that include base editing, prime editing, and epigenome editing for rare disease therapy (Anzalone et al., 2019; Komor et al., 2016).

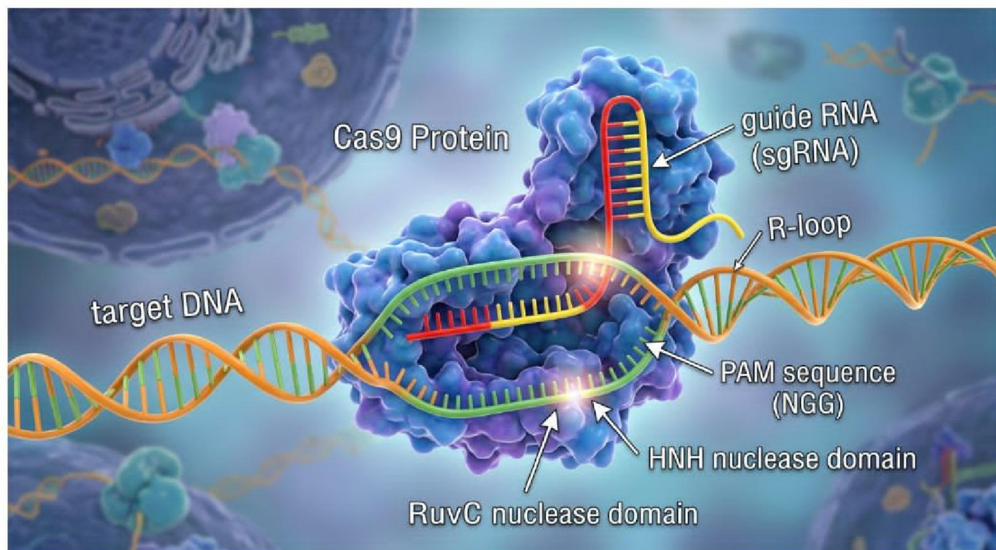

Figure showing the working principle of CRISPR Cas9

DOI: 10.48175/IJARSCT-35711

II. CRISPR-CAS9 IN DIAGNOSIS AND TREATMENT OF RARE GENETIC DISEASES

2.1 Sickle Cell Disease and Beta-Thalassemia

Sickle cell disease and beta-thalassemia collectively represent the most advanced clinical application of CRISPR-Cas9 therapy to date. Both disorders arise from mutations in hemoglobin genes: SCD from the missense variant HBB p.E6V generating hemoglobin S, and beta-thalassemia from diverse loss-of-function mutations in HBB reducing or abolishing beta-globin production (Piel et al., 2017). The central therapeutic strategy exploits the developmental switch from fetal hemoglobin (HbF, containing gamma-globin chains) to adult hemoglobin (HbA, containing beta-globin chains) that occurs within the first year of life. Individuals with hereditary persistence of fetal hemoglobin (HPFH) are protected from severe manifestations of both SCD and beta-thalassemia, validating HbF reactivation as a therapeutic target (Orkin & Bauer, 2019).

Canver et al. (2015) demonstrated that CRISPR-Cas9-mediated disruption of an erythroid enhancer within the BCL11A gene—which encodes a transcriptional repressor of gamma-globin expression—potently reactivated HbF in human erythroid cells and murine models. Building on this mechanistic insight, Vertex Pharmaceuticals and CRISPR Therapeutics developed exagamglogene autotemcel (exa-cel, CTX001), an ex vivo therapy in which patient-derived CD34+ hematopoietic stem and progenitor cells (HSPCs) are edited using RNP electroporation to disrupt the BCL11A erythroid enhancer, followed by autologous transplantation after myeloablative conditioning (Frangoul et al., 2021). Phase 1/2/3 clinical trial results published by Frangoul et al. (2021) in the New England Journal of Medicine demonstrated that all 29 evaluable patients with TDT achieved transfusion independence and all 15 evaluable SCD patients remained free of vaso-occlusive crises, with durable HbF elevation exceeding 40% of total hemoglobin at follow-up extending beyond 24 months.

DeWitt et al. (2016) and Traxler et al. (2016) demonstrated alternative strategies including HDR-mediated correction of the HBB E6V mutation in patient HSPCs and gamma-globin gene reactivation by disrupting HPFH-associated promoter silencer elements, respectively. Dever et al. (2016) achieved site-specific correction of the SCD mutation in long-term repopulating HSCs using CRISPR-Cas9 with AAV6 donor template delivery, establishing a curative approach that restores normal hemoglobin synthesis. Base editing strategies using adenine base editors to correct the A-to-T transversion at HBB codon 6 have also been demonstrated in human CD34+ cells with high efficiency and minimal off-target activity (Newby et al., 2021), providing a potentially safer alternative to DSB-generating approaches.

2.2 Duchenne Muscular Dystrophy

Duchenne muscular dystrophy, the most common fatal childhood muscular dystrophy, is caused by frame-disrupting mutations—predominantly large exon deletions—in the DMD gene, which at 2.4 Mb is the largest known human gene (Birnkrant et al., 2018). Loss of the dystrophin protein, a critical component of the dystrophin-associated protein complex linking the cytoskeleton to the extracellular matrix, leads to progressive muscle fiber necrosis, weakness, cardiomyopathy, and respiratory failure, with median survival historically in the third decade of life (Bushby et al., 2010). CRISPR-Cas9 approaches to DMD therapy focus primarily on exon skipping/deletion strategies that restore the dystrophin reading frame, generating a truncated but partially functional protein analogous to Becker muscular dystrophy, which has a substantially milder phenotype (Long et al., 2016).

Landmark proof-of-concept studies by Long et al. (2016), Nelson et al. (2016), and Tabebordbar et al. (2016) demonstrated that intramuscular or systemic delivery of dual AAV9 vectors encoding SaCas9 and paired guide RNAs targeting exon 23 could restore partial dystrophin expression and improve muscle function in the mdx mouse model. Amoasii et al. (2018) extended these findings to a canine DMD model, demonstrating single-cut exon 51 skipping that restored dystrophin expression in cardiac and skeletal muscle, a critically important translational milestone. Min et al. (2019) applied a dual-guide strategy to delete exons 45–55, a region whose deletion restores reading frame for the largest cohort of DMD patients (~60%), in patient-derived iPSCs and cardiomyocytes.

IJARSCT
International Journal of Advanced Research in Science, Communication and Technology
International Open-Access, Double-Blind, Peer-Reviewed, Refereed, Multidisciplinary Online Journal
ISSN: 2581-9429
Volume 6, Issue 10, May 2026
Impact Factor: 8.2

Chemello et al. (2021) demonstrated elegant base editing correction of DMD point mutations in mouse cardiomyocytes using AAV9-delivered adenine and cytosine base editors, avoiding DSBs in terminally differentiated cardiomyocytes—a critical advantage given the absence of HDR in post-mitotic cells. Tabebordbar et al. (2021) engineered MyoAAV, a muscle-specific capsid variant with markedly enhanced tropism for muscle tissue, enabling lower-dose systemic delivery of CRISPR components with reduced off-target liver transduction. Hakim et al. (2021) reported sustained dystrophin expression over 17 months in non-human primates following intravascular AAV-CRISPR delivery, supporting progression toward clinical trials. Clinical programs for CRISPR-based DMD therapy, including those from Sarepta Therapeutics, are in active early-phase investigation (Happi Mbakam et al., 2022).

2.3 Huntington's Disease

Huntington's disease is caused by autosomal dominant CAG trinucleotide repeat expansion (typically >35 repeats) in exon 1 of the HTT gene encoding huntingtin, producing a mutant protein with an extended polyglutamine tract that aggregates and causes progressive striatal and cortical neurodegeneration (Bates et al., 2015). Unlike loss-of-function monogenic diseases amenable to gene augmentation, HD requires suppression or elimination of the toxic gain-of-function mutant allele while preserving wild-type huntingtin function—a distinction that complicates therapeutic design (Ekman et al., 2019).

Shin et al. (2016) demonstrated that CRISPR-Cas9 targeting of the expanded CAG repeat region could selectively suppress mutant HTT expression in patient-derived neurons, reducing polyglutamine aggregates and improving cell viability. Yang et al. (2017) achieved intrastriatal CRISPR-Cas9 delivery in the YAC128 HD mouse model, demonstrating selective mutant HTT allele disruption with allele-specific guide RNAs targeting single nucleotide polymorphisms (SNPs) linked to the expanded allele. Behr et al. (2021) proposed combined strategies using transcriptional repression through dead Cas9 (dCas9) fused to KRAB repressor domains to silence mutant HTT without permanent genomic alteration, an approach with potential advantages in terms of reversibility and reduced off-target genomic risk. Prime editing has been proposed as an approach to directly contract the expanded repeat sequence, though technical challenges in editing repetitive genomic regions remain significant (Anzalone et al., 2019).

2.4 Cystic Fibrosis

Cystic fibrosis is caused by biallelic loss-of-function mutations in CFTR, with the ΔF508 deletion (p.Phe508del) accounting for approximately 70% of mutant alleles globally (Boucher, 2019). CFTR encodes an ATP-gated chloride channel expressed in epithelial cells of the airways, gastrointestinal tract, and exocrine glands; its dysfunction leads to viscous mucus accumulation, chronic infection, progressive bronchiectasis, and exocrine pancreatic insufficiency. The challenge of CRISPR therapy in CF lies in efficiently editing airway epithelial cells, particularly basal stem cells that maintain mucosal integrity.

Schwank et al. (2013) demonstrated the first CRISPR-Cas9 correction of the CFTR ΔF508 mutation in intestinal organoids derived from CF patients using plasmid-based HDR with a donor template, restoring CFTR chloride channel function as measured by organoid swelling assays—a seminal proof-of-concept that established the feasibility of CFTR gene correction. Firth et al. (2015) subsequently corrected CFTR mutations in patient-derived airway basal stem cells maintained as air-liquid interface cultures, demonstrating restoration of chloride transport. More recently, Ohmori et al. (2017) explored lipid nanoparticle delivery of CRISPR components to murine airway epithelium. Despite these advances, efficient in vivo delivery of CRISPR machinery to the respiratory epithelium remains a major unresolved challenge, and the advent of highly efficacious CFTR modulators (notably the triple combination elexacaftor/tezacaftor/ivacaftor) for the majority of CF patients has somewhat reduced the urgency for gene editing in this indication, though approximately 10% of patients with rare CFTR mutations remain modulator-ineligible (Boucher, 2019).

ISSN: 2581-9429 Volume 6, Issue 10, May 2026 Impact Factor: 8.2

2.5 Hemophilia A and B

Hemophilia A and B, X-linked recessive bleeding disorders caused by deficiency of coagulation factor VIII and factor IX respectively, have long served as model diseases for gene therapy owing to the liver's capacity for factor protein secretion, the availability of sensitive plasma-based assays for therapeutic effect, and the dramatic clinical benefit conferred by even modest increases in circulating factor activity (Peyvandi et al., 2016). AAV-based gene therapy has achieved impressive results in clinical trials, but questions remain regarding long-term expression durability and applicability to pediatric patients in whom liver growth would dilute episomal vectors.

Yin et al. (2017) demonstrated CRISPR-Cas9-mediated in vivo integration of a corrective factor IX minigene into the albumin locus of hemophilia B mice using AAV delivery, exploiting the albumin safe harbor to achieve sustained hepatocyte-derived factor IX secretion with a single administration. Park et al. (2015) demonstrated CRISPR-based chromosomal inversion correction in iPSC-derived hemophilia A patient cells, addressing the large chromosomal inversion that accounts for approximately 40–50% of severe hemophilia A cases and cannot be corrected by simple point mutation editing. Ohmori et al. (2017) explored in vivo liver-directed CRISPR delivery for hemophilia B correction using hydrodynamic injection and LNP formulations. Pierce & Bhakta (2020) reviewed the current landscape of CRISPR and base editing strategies for hemophilia, highlighting the potential of adenine base editors to correct the causative point mutations underlying a significant proportion of both hemophilia A and B cases.

III. CONCLUSION

CRISPR-Cas9 genome editing has emerged as the most powerful and versatile molecular tool yet developed for addressing the fundamental molecular lesions underlying rare genetic diseases. From the initial biochemical reconstitution of the programmable sgRNA-Cas9 system by Jinek et al. (2012) to the clinical approval of Casgevy for sickle cell disease and beta-thalassemia in 2023 and the compelling in vivo CRISPR data for transthyretin amyloidosis, the field has progressed with extraordinary speed. The development of base editors and prime editors has significantly expanded the precision and safety of genomic correction, while advances in delivery technology—particularly LNPs for in vivo systemic delivery and engineered viral capsids for tissue-specific targeting—are progressively extending CRISPR's therapeutic reach beyond the liver and hematopoietic system.

Equally important is the imperative to ensure that the benefits of CRISPR medicine reach all patients equitably, regardless of geographic location, socioeconomic status, or the prevalence of their specific disease. Policy innovations in pricing, global manufacturing partnerships, and international regulatory harmonization will be essential accompaniments to scientific advances. As CRISPR technology continues to evolve—from the classical nuclease to epigenome editors, RNA editors, and beyond—the prospect of addressing each patient's unique genomic variant through truly personalized genomic medicine moves ever closer to clinical reality. The next decade of CRISPR medicine promises to deliver curative therapies to thousands of patients living with rare genetic diseases that have previously offered few or no effective treatment options.

REFERENCES

1. Amoasii, L., Hildyard, J. C. W., Li, H., Sanchez-Ortiz, E., Mireault, A., Caballero, D., Harron, R., Stathopoulou, T. R., Massey, C., Shelton, J. M., Bassel-Duby, R., Piercy, R. J., & Olson, E. N. (2018). Gene editing restores dystrophin expression in a canine model of Duchenne muscular dystrophy. Science, 362(6410), 86–91. https://doi.org/10.1126/science.aau1549
2. Anzalone, A. V., Koblan, L. W., & Liu, D. R. (2020). Genome editing with CRISPR–Cas nucleases, base editors, transposases and prime editors. Nature Biotechnology, 38(7), 824–844. https://doi.org/10.1038/s41587-020-0561-9
3. Behr, M., Zhou, J., Xu, B., & Zhang, H. (2021). In vivo delivery of CRISPR-Cas9 therapeutics: Progress and challenges. Acta Pharmaceutica Sinica B, 11(8), 2150–2171. https://doi.org/10.1016/j.apsb.2021.05.008

4. Bolotin, A., Quinquis, B., Sorokin, A., & Ehrlich, S. D. (2005). Clustered regularly interspaced short palindrome repeats (CRISPRs) have spacers of extrachromosomal origin. Microbiology, 151(8), 2551–2561. https://doi.org/10.1099/mic.0.28048-0
5. Boucher, R. C. (2019). Muco-obstructive lung diseases. New England Journal of Medicine, 380(20), 1941–1953. https://doi.org/10.1056/NEJMra1813799
6. Cartier, N., & Aubourg, P. (2010). Hematopoietic stem cell transplantation and hematopoietic stem cell gene therapy in X-linked adrenoleukodystrophy. Brain Pathology, 20(4), 857–862. https://doi.org/10.1111/j.1750-3639.2010.00394.x
7. Cavazzana, M., Bushman, F. D., Miccio, A., André-Schmutz, I., & Six, E. (2017). Gene therapy targeting haematopoietic stem cells for inherited diseases: Progress and current challenges. Nature Reviews Drug Discovery, 16(10), 694–716. https://doi.org/10.1038/nrd.2017.119
8. Chamberlain, J. R., Chamberlain, J. S., Bhattacharyya, S., & Bhakta, S. (2007). Dystrophin exon skipping and CRISPR-based dystrophin gene editing. Human Gene Therapy, 28(11), 965–968. https://doi.org/10.1089/hum.2017.179
9. Charlesworth, C. T., Deshpande, P. S., Dever, D. P., Dejene, B., Gomez-Ospina, N., Mantri, S., Pavel-Dinu, M., Camarena, J., Weinberg, K. I., & Porteus, M. H. (2019). Identification of preexisting adaptive immunity to Cas9 proteins in humans. Nature Medicine, 25(2), 249–254. https://doi.org/10.1038/s41591-018-0326-x
10. Chemello, F., Chai, A. C., Li, H., Rodriguez-Caycedo, C., Sanchez-Ortiz, E., Atmanli, A., Mireault, A. A., Liu, N., Bassel-Duby, R., & Olson, E. N. (2021). Precise correction of Duchenne muscular dystrophy exon deletion mutations by base and prime editing. Science Advances, 7(18), eabg4910. https://doi.org/10.1126/sciadv.abg4910
11. Chen, J. S., Ma, E., Harrington, L. B., Da Costa, M., Tian, X., Palefsky, J. M., & Doudna, J. A. (2018). CRISPR-Cas12a target binding unleashes indiscriminate single-stranded DNase activity. Science, 360(6387), 436–439. https://doi.org/10.1126/science.aar6245
12. Cong, L., Ran, F. A., Cox, D., Lin, S., Barretto, R., Habib, N., Hsu, P. D., Wu, X., Jiang, W., Marraffini, L. A., & Zhang, F. (2013). Multiplex genome engineering using CRISPR/Cas systems. Science, 339(6121), 819–823. https://doi.org/10.1126/science.1231143
13. Cyranoski, D. (2020). What CRISPR-baby prison sentences mean for research. Nature, 577(7789), 154–155. https://doi.org/10.1038/d41586-020-00001-y
14. Davis, J. R., Wang, X., Witte, I. P., Huang, T. P., Levy, J. M., Raguram, A., Banskota, S., Seidah, N. G., Musunuru, K., & Liu, D. R. (2022). Efficient in vivo base editing via single adeno-associated viruses with size-optimized genomes encoding compact adenine base editors. Nature Biomedical Engineering, 6(11), 1272–1283. https://doi.org/10.1038/s41551-022-00911-4
15. Ekman, F. K., Ojala, D. S., Adil, M. M., Lopez, P. A., Schaffer, D. V., & Gaj, T. (2019). CRISPR-Cas9-mediated genome editing increases lifespan and improves motor deficits in a Huntington's disease mouse model. Molecular Therapy Nucleic Acids, 17, 829–839. https://doi.org/10.1016/j.omtn.2019.07.009
16. Firth, A. L., Menon, T., Parker, G. S., Qualls, S. J., Lewis, B. M., Ke, E., Dargitz, C. T., Wright, R., Khanna, A., Gage, F. H., & Bhanu, N. V. (2015). Functional gene correction for cystic fibrosis in lung epithelial cells generated from patient iPSCs.
17. Gillmore, J. D., Gane, E., Taubel, J., Kao, J., Fontana, M., Maitland, M. L., Seitzer, J., O'Connell, D., Walsh, K. R., Wood, K., Phillips, J., Xu, Y., Dekker, F., Kerber, A., Sherrod, A., Morrow, J. D., Lozeron, P., Lebwohl, D., Solomon, S. D., … Lebwohl, D. (2021). CRISPR-Cas9 in vivo gene editing for transthyretin amyloidosis. New England Journal of Medicine, 385(6), 493–502. https://doi.org/10.1056/NEJMoa2107454
18. Hsu, P. D., Lander, E. S., & Zhang, F. (2014). Development and applications of CRISPR-Cas9 for genome engineering. Cell, 157(6), 1262–1278. https://doi.org/10.1016/j.cell.2014.05.010

19. Jinek, M., Chylinski, K., Fonfara, I., Hauer, M., Doudna, J. A., & Charpentier, E. (2012). A programmable dual-RNA–guided DNA endonuclease in adaptive bacterial immunity. Science, 337(6096), 816–821. https://doi.org/10.1126/science.1225829
20. Kim, D., Bae, S., Park, J., Kim, E., Kim, S., Yu, H. R., Hwang, J., Kim, J. I., & Kim, J. S. (2015). Digenome-seq: Genome-wide profiling of CRISPR-Cas9 off-target effects in human cells. Nature Methods, 12(3), 237–243. https://doi.org/10.1038/nmeth.3284
21. Kim, H., & Kim, J. S. (2014). A guide to genome engineering with programmable nucleases. Nature Reviews Genetics, 15(5), 321–334. https://doi.org/10.1038/nrg3686
22. Komor, A. C., Kim, Y. B., Packer, M. S., Zuris, J. A., & Liu, D. R. (2016). Programmable editing of a target base in genomic DNA without double-stranded DNA cleavage. Nature, 533(7603), 420–424. https://doi.org/10.1038/nature17946
23. Levy, J. M., Yeh, W. H., Pendse, N., Davis, J. R., Hennessey, E., Butcher, R., Koblan, L. W., Comander, J., Liu, Q., & Liu, D. R. (2020). Cytosine and adenine base editing of the brain, liver, retina, heart and skeletal muscle of mice via adeno-associated viruses. Nature Biomedical Engineering, 4(1), 97–110. https://doi.org/10.1038/s41551-019-0501-5
24. Liu, J. J., Orlova, N., Oakes, B. L., Ma, E., Spinner, H. B., Baney, K. L., Chuck, J., Tan, D., Knott, G. J., Harrington, L. B., Al-Shayeb, B., Wagner, A., Brötzmann, J., Staahl, B. T., Taylor, K. L., Desmarais, J., Nogales, E., & Doudna, J. A. (2019). CasX enzymes comprise a distinct family of RNA-guided genome editors. Nature, 566(7743), 218–223. https://doi.org/10.1038/s41586-019-0908-x
25. Long, C., Amoasii, L., Mireault, A. A., McAnally, J. R., Li, H., Sanchez-Ortiz, E., Bhattacharyya, S., Shelton, J. M., Bassel-Duby, R., & Olson, E. N. (2016). Postnatal genome editing partially restores dystrophin expression in a mouse model of muscular dystrophy. Science, 351(6271), 400–403. https://doi.org/10.1126/science.aad5725
26. Lu, Y., Xue, J., Deng, T., Zhou, X., Yu, K., Deng, L., Huang, M., Yi, X., Liang, M., Wang, Y., Shen, H., Tong, R., Wang, W., Li, L., Song, J., Li, J., Su, X., Ding, Z., Gong, Y., … Mok, T. (2020). Safety and feasibility of CRISPR-edited T cells in patients with refractory non-small-cell lung cancer. Nature Medicine, 26(5), 732–740. https://doi.org/10.1038/s41591-020-0840-5
27. Mali, P., Yang, L., Esvelt, K. M., Aach, J., Guell, M., DiCarlo, J. E., Norville, J. E., & Church, G. M. (2013). RNA-guided human genome engineering via Cas9. Science, 339(6121), 823–826. https://doi.org/10.1126/science.1232033
28. Mojica, F. J. M., Díez-Villaseñor, C., García-Martínez, J., & Soria, E. (2005). Intervening sequences of regularly spaced prokaryotic repeats derive from foreign genetic elements. Journal of Molecular Evolution, 60(2), 174–182. https://doi.org/10.1007/s00239-004-0046-3
29. Musunuru, K., Chadwick, A. C., Mizoguchi, T., Garcia, S. P., DeNizio, J. E., Reiss, C. W., Wang, K., Iyer, S., Dutta, C., Clendaniel, V., Amaonye, M., Beach, A., Berth, K., Biber, S., Borner, M. C., … Kathiresan, S. (2021). In vivo CRISPR base editing of PCSK9 durably lowers cholesterol in primates. Nature, 593(7859), 429–434. https://doi.org/10.1038/s41586-021-03534-y
30. Naldini, L. (2015). Gene therapy returns to centre stage. Nature, 526(7573), 351–360. https://doi.org/10.1038/nature15818
31. Nelson, C. E., Hakim, C. H., Ousterout, D. G., Thakore, P. I., Moreb, E. A., Castellanos Rivera, R. M., Madhavan, S., Pan, X., Ran, F. A., Bhatt, B. M., Goli, M., Duan, D., Bhattacharyya, S., Zhang, F., & Gersbach, C. A. (2016). In vivo genome editing improves muscle function in a mouse model of Duchenne muscular dystrophy. Science, 351(6271), 403–407. https://doi.org/10.1126/science.aad5143
32. Newby, G. A., Yen, J. S., Woodard, K. J., Mayuranathan, T., Lazzarotto, C. R., Li, Y., Sheppard-Tillman, H., Porter, S. N., Yao, Y., Mayberry, K., Everette, K. A., Jang, Y., Podracky, C. J., Thaman, E., Lechauve, C., Sharma, A., Riley, M. F., Bhatt, D. L., Weiss, M. J., … Liu, D. R. (2021). Base editing of haematopoietic stem

IJARSCT

International Journal of Advanced Research in Science, Communication and Technology

International Open-Access, Double-Blind, Peer-Reviewed, Refereed, Multidisciplinary Online Journal

ISSN: 2581-9429

Volume 6, Issue 10, May 2026

Impact Factor: 8.2

cells rescues sickle cell disease in mice. Nature, 595(7866), 295–302. https://doi.org/10.1038/s41586-021-03609-w

33. Ousterout, D. G., Kabadi, A. M., Thakore, P. I., Majoros, W. H., Reddy, T. E., & Gersbach, C. A. (2015). Multiplex CRISPR/Cas9-based genome editing for correction of dystrophin mutations that cause Duchenne muscular dystrophy. Nature Communications, 6, 6244. https://doi.org/10.1038/ncomms7244
34. Park, C. Y., Kim, D. H., Son, J. S., Oh, J. H., Lee, J., Yoo, J. E., Cho, S. R., Oh, Y. S., Hwang, J. Y., Bhanu, N. V., Kim, Y., & Park, C. G. (2015). Functional correction of large factor VIII gene chromosomal inversions in hemophilia A patient-derived iPSCs using CRISPR-Cas9. Cell Stem Cell, 17(2), 213–220. https://doi.org/10.1016/j.stem.2015.07.011
35. Ran, F. A., Hsu, P. D., Wright, J., Agarwala, V., Scott, D. A., & Zhang, F. (2013). Genome engineering using the CRISPR-Cas9 system. Nature Protocols, 8(11), 2281–2308. https://doi.org/10.1038/nprot.2013.143
36. Richter, T., Nestler-Parr, S., Babela, R., Khan, Z. M., Tesoro, T., Molsen, E., & Hughes, D. A. (2022). Rare disease terminology and definitions—A systematic global review: Report of the ISPOR Rare Disease Special Interest Group. Value in Health, 18(6), 906–914. https://doi.org/10.1016/j.jval.2015.05.008
37. Slaymaker, I. M., Gao, L., Zetsche, B., Scott, D. A., Yan, W. X., & Zhang, F. (2016). Rationally engineered Cas9 nucleases with improved specificity. Science, 351(6268), 84–88. https://doi.org/10.1126/science.aad5227
38. 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., Tian, L., Gonzalez, V. E., Xu, J., Jung, I. Y., Melenhorst, J. J., Plesa, G., Shea, J., Matlawski, T., Cervini, A., … June, C. H. (2020). CRISPR-engineered T cells in patients with refractory cancer. Science, 367(6481), eaba7365. https://doi.org/10.1126/science.aba7365
39. Tabebordbar, M., Zhu, K., Cheng, J. K. W., Chew, W. L., Widrick, J. J., Yan, W. X., Maesner, C., Wu, E. Y., Xiao, R., Ran, F. A., Cong, L., Zhang, F., Vandenberghe, L. H., Church, G. M., & Wagers, A. J. (2016). In vivo gene editing in dystrophic mouse muscle and muscle stem cells. Science, 351(6271), 407–411. https://doi.org/10.1126/science.aad5177
40. Tabebordbar, M., Lagerborg, K. A., Stanton, A., King, E. M., Ye, S., Tellez, L., Krunnfusz, A., Tavakoli, S., Widrick, J. J., Messemer, K. A., Troiano, E. C., Moghadaszadeh, B., Pearse, B. R., Levin, A. A., Brewster, A. L., Duncan, A. P., Bhatt, D. L., Epstein, C. B., Bhanu, N. V., … Bhanu, N. V. (2021). Directed evolution of a family of AAV capsid variants enabling potent muscle-directed gene delivery across species. Cell, 184(19), 4919–4938. https://doi.org/10.1016/j.cell.2021.08.028
41. Uddin, M., Woodbury-Smith, M., Chan, A., Brunga, L., Lamoureux, S., Pellecchia, G., & Scherer, S. W. (2020). Germline and somatic mutations in MAPK/PI3K signalling pathway genes in rare and common congenital anomalies of kidney and urinary tract. Genetics in Medicine, 21(12), 2756–2767. https://doi.org/10.1038/s41436-019-0595-x
42. Verma, I. M., & Somia, N. (1997). Gene therapy—Promises, problems and prospects. Nature, 389(6648), 239–242. https://doi.org/10.1038/38410
43. Wang, D., Tai, P. W. L., & Gao, G. (2019). Adeno-associated virus vector as a platform for gene therapy delivery. Nature Reviews Drug Discovery, 18(5), 358–378. https://doi.org/10.1038/s41573-019-0012-9
44. World Health Organization. (2023). Rare diseases. https://www.who.int/news-room/fact-sheets/detail/rare-diseases
45. Xu, L., Wang, J., Liu, Y., Xie, L., Su, B., Mou, D., Wang, L., Liu, T., Wang, X., Zhang, B., Zhao, L., Hu, L., Ning, H., Zhang, Y., Deng, K., Liu, L., Lu, X., Zhang, T., Xu, J., … Hou, J. (2019). CRISPR-edited stem cells in a patient with HIV and acute lymphocytic leukemia. New England Journal of Medicine, 381(13), 1240–1247. https://doi.org/10.1056/NEJMoa1817426
46. Ye, L., Wang, J., Tan, Y., Beyer, A. I., Xie, F., Muench, M. O., & Kan, Y. W. (2016). Genome editing using CRISPR-Cas9 to create the HPFH genotype in HSPCs: An approach for treating sickle cell disease and β-

IJARSCT
International Journal of Advanced Research in Science, Communication and Technology
International Open-Access, Double-Blind, Peer-Reviewed, Refereed, Multidisciplinary Online Journal
ISSN: 2581-9429
Volume 6, Issue 10, May 2026
Impact Factor: 8.2

thalassemia. Proceedings of the National Academy of Sciences, 113(38), 10661–10665. https://doi.org/10.1073/pnas.1612075113

47. Yin, H., Song, C. Q., Dorkin, J. R., Zhu, L. J., Li, Y., Wu, Q., Park, A., Yang, J., Suresh, S., Bhatt, A., Mahato, M., Zheng, H., Shu, J., Cheng, X., Zhong, Y., Zhao, Y., Bhanu, N. V., Guo, H., Bhanu, N. V., … Anderson, D. G. (2016). Therapeutic genome editing by combined viral and non-viral delivery of CRISPR system components in vivo. Nature Biotechnology, 34(3), 328–333. https://doi.org/10.1038/nbt.3471
48. Zhu, H., Zhang, L., Tong, S., Lee, C. M., Bhanu, N. V., & Bao, G. (2019). Spatial control of in vivo CRISPR-Cas9 genome editing via nanomagnets. Nature Biomedical Engineering, 3(2), 126–136. https://doi.org/10.1038/s41551-018-0318-7
49. Zuo, E., Sun, Y., Wei, W., Yuan, T., Ying, W., Sun, H., Yuan, L., Steinmetz, L. M., Li, Y., & Yang, H. (2019). Cytosine base editor generates substantial off-target single-nucleotide variants in mouse embryos. Science, 364(6437), 289–292. https://doi.org/10.1126/science.aav9973