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Review Article

Gene Therapy Approaches in the Prevention and Control of Parasitic Infections

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Abstract

Parasitic diseases, particularly malaria, leishmaniasis, trypanosomiasis, toxoplasmosis and schistosomiasis, remain a major cause of morbidity and mortality in lowresource settings. Conventional smallmolecule therapies are increasingly compromised by drug resistance and treatmentlimiting toxicity. This review evaluates gene therapy and programmable genomeediting platforms as alternative strategies for preventing and controlling parasitic infections, examining four core molecular mechanisms (direct disruption of essential parasite genes, engineering of host genetic resistance, population-level vector modification through gene drives, and targeted gene silencing via RNA interference) and to analyse the associated delivery systems, safety profiles, regulatory landscapes, and equity barriers. A narrative review was conducted of experimental studies, preclinical investigations, and earlyphase clinical trials published between 2020 and 2026. Sources included peerreviewed literature indexed in PubMed, Web of Science and Scopus, together with technical reports from the World Health Organization and international regulatory agencies. The review covers the principal editing platforms (CRISPRCas9, TALENs, ZFNs), delivery vectors (viral, lipid nanoparticle, polymeric and nanotechnologybased carriers), and applications across five major parasitic diseases. Highefficiency disruption of fitnesscritical genes has been demonstrated in *Plasmodium falciparum*, *Leishmania* spp., *Trypanosoma brucei*, *Toxoplasma gondii* and *Schistosoma mansoni*. Genedrive *Anopheles* mosquitoes achieved over 95% inheritance bias in insectary trials and suppressed patientderived *P. falciparum* isolates in contained field evaluations. Markerfree liveattenuated *Leishmania* vaccines conferred protective immunity without lesion formation. RNA interference delivered by nanoparticle carriers silenced essential genes in parasites lacking canonical RNAi machinery. Delivery performance is constrained by vector immunogenicity, packaging limits, thermostability requirements, and offtarget mutagenesis risk. Regulatory fragmentation, unresolved ethical questions surrounding germline modification and environmental genedrive release, and the prohibitive cost of approved gene therapies (USD 1.5-3.5 million per patient) constitute major barriers to equitable deployment. Genetherapy platforms can disrupt parasite life cycles, enhance host resistance, and modify vector populations. Translating this promise into global health impact will require highfidelity editing tools, thermostable nonviral delivery systems, harmonised regulatory and biosafety frameworks, and openaccess licensing models that embed equity from the earliest stages of product development.

Keywords: CRISPR Cas9; Gene Therapy; Parasitic Infections; RNA Interference; Vectors

Introduction

Parasitic diseases have retained their position among the most burdensome infectious conditions affecting human populations, with their impact concentrated overwhelmingly in tropical and subtropical regions where poverty, inadequate sanitation infrastructure, and limited healthcare access create conditions for sustained transmission. Malaria alone accounted for approximately 249 million cases and 608,000 deaths worldwide in 2022, with subSaharan Africa bearing approximately 94% of this mortality burden [1]. Beyond malaria, schistosomiasis affects an estimated 250 million people across 78 countries, while leishmaniasis and human African trypanosomiasis continue to cause substantial morbidity and mortality in affected regions despite being classified as neglected tropical diseases [2]. The consequences extend far beyond direct clinical manifestations; chronic infection impairs cognitive development in children, reduces workforce productivity, and traps affected communities in cycles of poverty that prove resistant to conventional development interventions [2].

The pharmacological foundation of parasitic disease control has rested for decades on a relatively small repertoire of smallmolecule agents. Chloroquine, introduced in the 1940s, revolutionised malaria treatment until the emergence and spread of resistant *Plasmodium falciparum* strains rendered it ineffective across most endemic regions [3]. Artemisininbased combination therapies (ACTs), adopted as firstline treatment following widespread chloroquine failure, now face their own threat from partial resistance that has been documented across Southeast Asia and is now spreading toward Africa, with kelch13 mutations serving as molecular markers for reduced parasite susceptibility [3,4]. Similar resistance trajectories have emerged for antileishmanial compounds, including antimonials and miltefosine, while melarsoprol, used for latestage human African trypanosomiasis, carries a treatmentassociated mortality of approximately 510% [5]. The toxicity profiles of many antiparasitic agents present additional limitations; pentamidine induces severe hypoglycaemia and pancreatic toxicity, chloroquine retinopathy threatens longterm vision, and the arsenicbased melarsoprol causes reactive encephalopathy in a significant minority of treated patients [5].

Gene therapy, understood broadly as the therapeutic modification of genetic material within living cells, represents a conceptual departure from conventional pharmacological approaches. Rather than administering compounds that interact indirectly with parasite or host biology, gene therapy seeks to alter the underlying genetic determinants of disease susceptibility, progression, or transmission [6]. The clinical maturation of this field has been accelerated by advances in programmable nuclease technology, particularly the CRISPRCas9 system, which has dramatically reduced the technical barriers to precise genome editing across virtually all experimentally tractable organisms [7]. For parasitic diseases, gene therapy includes a spectrum of applications that ranges from direct editing of parasite genomes to disable essential biological functions, through modification of vector species to block transmission, to augmentation of host immune responses through genetic immunisation strategies [8,9].

The present review examines the current state of gene therapy approaches for the prevention and control of parasitic infections, with particular attention to the molecular mechanisms, delivery challenges, and translational barriers that define this rapidly evolving field. The scope covers five major parasitic diseases, malaria, leishmaniasis, trypanosomiasis, toxoplasmosis, and schistosomiasis, reflecting both their individual disease burdens and the diversity of biological strategies required to address them.

Overview of gene therapy

Definition and principles

Gene therapy may be defined, following the United States Food and Drug Administration (FDA) formulation, as any therapeutic approach that mediates its effects through the transcription or translation of transferred genetic material [10]. This definition incorporates a broad range of modalities, from the introduction of functional gene copies to compensate for inherited deficiencies, to the targeted inactivation or deletion of deleterious sequences, to the delivery of exogenous genetic material encoding therapeutic proteins or regulatory RNAs [6]. The conceptual foundations rest on the recognition that many disease processes, infectious as well as genetic, have molecular underpinnings that can be addressed through directed manipulation of nucleic acid sequences within relevant cell types.

The practical implementation of gene therapy follows a sequential logic that begins with precise identification of the target genetic pathway and proceeds through delivery and expression phases. Identification of the target gene or pathway

requires detailed knowledge of the molecular basis of disease, which for parasitic infections means understanding the essential biological processes that sustain parasite survival, replication, and transmission within both vertebrate and invertebrate hosts [11]. Delivery constitutes the most technically demanding step, requiring that therapeutic genetic material reach the appropriate cells or tissues in a form that permits functional expression. Viral vectors, including adenoassociated virus (AAV), lentivirus, and adenovirusderived systems, exploit the natural infectivity of viruses to achieve highefficiency transduction, though at the cost of potential immunogenicity and packaging constraints that limit cargo size [12]. Nonviral approaches, including lipid nanoparticles, polymeric carriers, and physical methods such as electroporation, offer reduced immunological risk but generally lower delivery efficiency [13]. Once delivered, the therapeutic sequence must achieve expression or modification at levels sufficient to produce a biological effect, while remaining within safety parameters that preclude harmful offtarget activity [6].

Somatic and germline gene therapy

The distinction between somatic and germline gene therapy carries both technical and ethical significance. Somatic gene therapy modifies genetic material in nonreproductive cells, meaning that any changes are confined to the treated individual and are not transmitted to offspring [14]. This modality has been applied clinically for conditions including inherited retinal dystrophies, spinal muscular atrophy, and haemoglobinopathies, with several products now approved by major regulatory agencies [15]. For parasitic diseases, somatic approaches involve strategies such as the genetic modification of host haematopoietic cells to confer resistance, or the delivery of therapeutic genes to infected tissues [14]. Germline gene therapy, by contrast, involves modification of reproductive cells or early embryos, with the consequence that introduced changes would be heritable across generations. While germline editing could theoretically eliminate susceptibility alleles from affected populations, the 2018 case of He Jiankui, who edited the CCR5 gene in human embryos ostensibly to confer HIV resistance, demonstrated the potential for misuse and prompted widespread regulatory prohibition of clinical germline editing [16]. Current international consensus permits germline editing only in research contexts, with a moratorium on clinical applications.

Gene editing technologies

Three programmable nuclease platforms have dominated the gene editing field. Clustered Regularly Interspaced Short Palindromic Repeats and CRISPRassociated protein 9 (CRISPRCas9), derived from the adaptive immune system of *Streptococcus pyogenes*, uses a singleguide RNA to direct the Cas9 endonuclease to a specific DNA sequence, where it introduces a doublestrand break that is subsequently repaired through either errorprone nonhomologous end joining, which typically disrupts gene function through insertions or deletions, or homologydirected repair, which can introduce precise modifications when a repair template is provided [17,18]. Transcription ActivatorLike Effector Nuclease (TALENs), an engineered protein designed to recognize specific DNA sequences and cut them using a nuclease enzyme, employ a different DNA recognition mechanism, with each transcription activatorlike effector domain recognising a single nucleotide, allowing assembly of custom DNAbinding proteins fused to FokI nuclease domains that dimerise to create targeted breaks [19]. Zinc Finger Nucleases (ZFNs), the earliest programmable nuclease platform, similarly use customisable DNAbinding domains based on zinc finger proteins fused to FokI, though their design requires more complex protein engineering compared with the RNAprogrammed CRISPR system [20].

Parasitic diseases targeted by gene therapy

Malaria

Malaria remains the parasitic disease with the greatest global mortality burden, caused by *Plasmodium* species transmitted through infected female *Anopheles* mosquitoes. *Plasmodium falciparum* accounts for the overwhelming majority of severe and fatal cases, with children under five and pregnant women facing the highest risk [1]. Gene therapy strategies for malaria have pursued two conceptually distinct paths. The first targets the parasite directly, using CRISPRCas9 to disrupt genes essential for development or transmission. Gantz., *et al*. [8] demonstrated that targeted disruption of the AP2G transcription factor, which governs gametocyte differentiation, could block sexual stage development and thereby interrupt transmission to mosquitoes. The second path modifies mosquito vectors to render them refractory to parasite infection, using gene drive systems to spread antiparasitic effector genes through wild populations [21,22]. The recent demonstration by Habtewold., *et al*. [22] that genedrivecapable *Anopheles*

gambiae mosquitoes expressing antimicrobial peptides could suppress patientderived *P. falciparum* isolates in Tanzanian field trials represents a significant advance toward practical application.

Leishmaniasis

Leishmaniasis, caused by protozoan parasites of the genus *Leishmania* transmitted through sandfly vectors, manifests across a clinical spectrum from selfhealing cutaneous lesions to fatal visceral disease. CRISPRbased approaches have enabled the generation of attenuated parasite strains with potential as live vaccines; deletion of the centrin gene in *Leishmania major* produced parasites that could persist transiently in host tissues without causing progressive disease, while eliciting protective cellular immunity [23]. The immune evasion mechanisms employed by *Leishmania*, including manipulation of host macrophage signalling pathways, have also been probed using CRISPRbased genetic screens, identifying parasite factors that suppress host protective responses and thereby revealing potential therapeutic targets [24].

Trypanosomiasis

Trypanosomiasis includes two clinically distinct conditions; Human African trypanosomiasis (also known as sleeping sickness), caused by *Trypanosoma brucei gambiense* and *T. b. rhodesiense*, is transmitted by tsetse flies and progresses from a haemolymphatic stage to meningoencephalitic involvement that is fatal without treatment. Chagas disease, caused by *T. cruzi* and transmitted by triatomine bugs, produces acute infection that often resolves but may reemerge decades later as chronic cardiomyopathy or gastrointestinal megasyndromes [25]. The antigenic variation mechanism of *T. brucei*, mediated through sequential expression of variant surface glycoproteins (VSGs), has long frustrated vaccine development. RNAimediated silencing of VSG expression siteassociated genes has been explored as a means to disable this immune evasion strategy, with *in vitro* studies demonstrating reduced parasitemia when expression site switching is inhibited [26]. For Chagas disease, CRISPRbased screens have identified *T. cruzi* genes essential for host cell invasion and intracellular survival, including surface proteases and calciumsignalling components that represent potential therapeutic targets [25].

Toxoplasmosis

Toxoplasmosis, caused by the obligate intracellular apicomplexan *Toxoplasma gondii*, infects an estimated onethird of the global human population, though clinical disease is most severe in immunocompromised individuals and in congenitally infected foetuses [27]. Gene therapy approaches have targeted both parasite virulence factors and host immune responses. The dense granule proteins GRA16 and GRA24, which the parasite secretes into host cells to modulate immune signalling, have been disrupted using CRISPRCas9, yielding attenuated strains with reduced pathogenicity in cellular and animal models [28]. Complementary approaches have used AAV vectors to deliver immunomodulatory genes, including interferongamma (IFNgamma) and interleukin12 (IL12), to infected tissues, strengthening cellmediated immunity against intracellular parasite stages [29].

Schistosomiasis

Schistosomiasis, caused by blood flukes of the genus *Schistosoma*, affects over 250 million people worldwide, with chronic infection causing hepatic fibrosis, portal hypertension, and increased risk of bladder cancer in *S. haematobium* infections. CRISPRCas9 editing has been applied to *S. mansoni*, with targeted disruption of the omega1 ribonuclease gene demonstrating that this secreted factor is essential for Th2 polarization and granuloma formation, and that geneedited parasites elicit markedly reduced immunopathology [30]. RNAi approaches have also been adapted for schistosome gene silencing, using lipid nanoparticle formulations to deliver small interfering RNAs to adult worms *in vivo*, achieving knockdown of essential genes involved in egg production and glucose metabolism [31].

Mechanisms of gene therapy in parasite control

The application of gene therapy to parasite control operates through several mechanistically distinct pathways that may be classified according to whether they target the parasite, the vector, or the host. Direct targeting of parasite genomes exploits the essentiality of specific genes for survival, replication, or transmission [32]. CRISPRCas9mediated disruption of transporter genes in *Babesia microti*, an apicomplexan blood parasite of veterinary importance, demonstrated that knockout of metabolite transporters substantially impairs *in vitro* growth, and that preassembled complexes of human Argonaute 2 with targeting singlestranded RNAs, delivered through chitosan nanoparticles, can achieve functional gene silencing even in parasites lacking canonical RNAi processing machinery [9]. Similar approaches have identified stagespecific essential genes in *Plasmodium*

falciparum blood stages, providing a catalogue of potential therapeutic targets across the parasite life cycle.

Host genetic resistance represents a complementary strategy that seeks to modify the vertebrate host to make it less permissive for parasite survival. Advances in genomic selection and editing have enabled identification and manipulation of resistanceassociated loci in both human and livestock populations [33]. Arzik., *et al*. [34] conducted a genomewide association study in Central Anatolian Merino sheep that identified CD79A and MAP3K7 as genes significantly associated with resistance to *Moniezia* tapeworm infection, suggesting that Bcell activation pathways and NFkappaB signalling are critical determinants of host protective immunity. Salim., *et al*. [35] reviewed the broader landscape of genetic resistance to vectorborne hemoprotozoa in livestock, identifying polymorphisms in BoLADRB3, TLR4, and antioxidant genes such as SOD2 and GPX1 as contributors to resistance phenotypes, and noting that markerassisted selection programmes integrating these loci have shown promise in cattle, small ruminants, and equines.

Immunological enhancement through gene therapy draws on the recognition that many parasites persist through active suppression or evasion of host immune responses. Recombinant adenoassociated virus (rAAV) vectors encoding antiPD1 antibody constructs have been investigated as a means to block the inhibitory checkpoint signalling that renders T cells dysfunctional during chronic infection; Afla., *et al*. [36] reviewed evidence that PD1 pathway inhibition can restore Tcell effector function against viral, bacterial, and parasitic pathogens, and that rAAVmediated delivery offers durable expression without repeated dosing. For intracellular parasites such as *Leishmania* and *Toxoplasma*, this approach is particularly relevant because control depends on sustained cellular immunity.

Gene silencing through RNA interference provides a mechanistically distinct approach that does not require permanent genetic modification. Small interfering RNAs of 2123 nucleotides can guide the RNAinduced silencing complex (RISC) to complementary messenger RNA sequences, triggering degradation and reducing expression of the targeted gene. ElSayed., *et al*. [9] demonstrated that this approach can be adapted for parasites lacking canonical RNAi machinery by preassembling RISC complexes ex vivo and delivering them through nanoparticle carriers, achieving significant reductions in target gene expression and parasite growth in both *in vitro* and *in vivo Babesia* infection models.

Current research and clinical progress

The period from 2020 to 2025 has yielded proof-of-concept demonstrations across the major parasitic diseases, yet the distance from these experimental successes to clinically deployable interventions remains substantial. Live-attenuated parasite vaccines generated by precise gene deletion represent one of the more advanced translational trajectories. Zhang., *et al*. [23] showed that CRISPR-Cas9-mediated deletion of the centrin gene in Leishmania major produced parasites that conferred protection against both visceral and cutaneous leishmaniasis in animal models, avoiding the safety concerns inherent to traditional radiation-attenuated vaccines. This approach was subsequently extended by Karmakar., *et al*. [37], who demonstrated that centrin-deficient L. mexicana could reprogram host tryptophan metabolism in ways that enhanced protective immunity, illustrating how genetic attenuation can simultaneously function as an immunological adjuvant. While these findings are encouraging, the absence of human challenge studies and the limited durability data beyond a few months constrain conclusions about real-world prophylactic utility.

Malaria vector modification has advanced from laboratory proof-of-concept to contained field evaluation, though critical uncertainties remain. Carballar-Lejarazu., *et al*. [21] constructed dual-effector gene-drive strains of Anopheles gambiae and *A. coluzzii* carrying single-chain antibody constructs targeting both ookinete and sporozoite stages of *P. falciparum*; these strains achieved full population introduction within 3-6 months in small-cage trials and modelled 50-90% reductions in malaria incidence. The crucial question, whether such strains could suppress genetically diverse, naturally circulating parasite isolates rather than laboratory-adapted lines, was addressed by Habtewold., *et al*. [22], who engineered locally derived A. gambiae in Tanzanian containment facilities and demonstrated that non-autonomous gene drive mosquitoes could suppress patient-derived *P. falciparum* isolates when supplemented with Cas9 from a separate locally engineered strain. These data are promising but were obtained in large-cage and contained-field settings that do not replicate the ecological complexity, fitness trade-offs, and evolutionary

responses expected upon open environmental release. Separately, Yang., *et al*. [40] established that Cas12a-mediated genome editing is a competitive alternative to Cas9 in malaria parasites, offering an additional nuclease platform that may mitigate some off-target concerns, though its efficiency and delivery characteristics have been examined only *in vitro*.

For schistosomiasis, the landmark demonstration by Ittiprasert., *et al*. [30] that CRISPR editing of the omega-1 ribonuclease in *S. mansoni* markedly reduces Th2 polarisation and granuloma formation confirmed the feasibility of functional gene disruption in parasitic flatworms. Naidoo and Mkhize-Kwitshana [31] subsequently extended CRISPR-Cas9 editing to additional *S. mansoni* genes, cataloguing potential vaccine and drug targets through systematic disruption studies. The pipeline from target identification to *in vivo* validation, however, remains slow, and no edited schistosome strain has yet been evaluated for transmission-blocking efficacy in a natural snail-human cycle.

In toxoplasmosis, Shen., *et al*. [39] constructed a double-knockout RH strain lacking both ompdc and uprt genes, which was severely attenuated yet elicited multi-stage protective immunity in mice and cats, reducing oocyst shedding by 95.3% in challenged feline hosts. This work demonstrates that multiplexed CRISPR editing can generate attenuation sufficient for vaccine applications while preserving immunogenicity, though the safety of such live-attenuated strains in immunocompromised hosts, a population at high risk for toxoplasmosis, has not been addressed. A distinct transmission-blocking strategy for trypanosomiasis was reported by Aksoy., *et al*. [38], who employed paratransgenesis to engineer *Sodalis glossinidius* symbiont bacteria resident in tsetse flies, enabling secretion of trypanocidal compounds that blocked T. brucei transmission without direct genetic modification of the vector. While elegant, this approach depends on stable symbiont colonisation and sustained expression of effectors in field populations, parameters that have been demonstrated only in laboratory-reared flies.

Collectively, these studies confirm that gene therapy platforms can disrupt parasite life cycles, attenuate virulence, and modify vector competence with high specificity in controlled settings. Nonetheless, the evidence base remains heavily weighted toward short-term, small-scale laboratory experiments, with only the malaria gene drive work having advanced to contained field evaluation. None of the vaccine candidates has progressed to human trials, and the safety, durability, and scalability required for implementation in endemic regions remain largely unexamined.

Delivery systems in gene therapy

Viral vectors

AAV vectors have emerged as the most clinically validated gene delivery platform, with established safety records across multiple approved therapies. In the context of parasitic disease, AAV vectors offer particular utility for delivering antibodyencoding genes as a form of vectored immunoprophylaxis. Deal., *et al*. [41] demonstrated that a single intramuscular injection of AAV expressing an anti*P. falciparum* circumsporozoite protein antibody maintained protective serum titers for over 36 weeks in nonhuman primates, an approach that circumvents cold chain requirements after administration and could be compatible with field deployment in remote endemic areas. The principal limitations of AAV include a packaging capacity of approximately 4.7 kb, which constrains the size of deliverable genetic cargoes, and the high prevalence of preexisting neutralising antibodies in human populations, with seroprevalence estimates of 4070% in subSaharan African populations that constitute the primary target for malaria interventions [42]. Engineered capsid variants with reduced immunogenicity are in active development to address this limitation.

Lentiviral vectors provide an alternative platform distinguished by stable genomic integration, which enables persistent transgene expression in dividing and nondividing cells alike. This property is particularly relevant for ex vivo editing of haematopoietic stem cells, where permanent modification of the progenitor compartment can generate durable populations of parasiteresistant blood cells [43]. Thirdgeneration selfinactivating lentiviral designs have substantially reduced the risk of insertional oncogenesis that accompanied early clinical applications, and comprehensive integration site monitoring is now standard in clinical protocols.

Nonviral vectors

Lipid nanoparticles have gained prominence following their successful deployment in mRNA COVID19 vaccines, and their application to parasitic diseases is being actively explored. The selfassembling structure of ionisable lipid, phospholipid, cholesterol, and PEGlipid components encapsulates nucleic acid cargo and facilitates endosomal escape after cellular uptake [44]. For leishmaniasis, mannosereceptortargeted lipid nanoparticles

have achieved substantially enhanced siRNA delivery to infected macrophages compared with nontargeted formulations, exploiting the abundance of mannose receptors on macrophage surfaces to concentrate therapeutic payloads at the primary site of infection [45]. Thermostability remains a critical barrier for tropical deployment; lyophilised formulations stable at ambient temperatures are in preclinical development to reduce cold chain dependency.

Nanotechnologybased delivery

Advances in nanotechnology have expanded the delivery toolkit beyond classical viral and lipidbased systems. Superparamagnetic iron oxide nanoparticles conjugated with siRNA enable magnetically directed delivery to infected liver tissue under external magnetic guidance, improving spatial targeting for parasites with hepatic tropism such as *Plasmodium* liver stages and *Leishmania donovani* [46]. Exosomebased biomimetic vectors, which exploit the natural membrane properties of macrophagederived extracellular vesicles, have demonstrated preferential uptake by *Leishmania*infected macrophages and prolonged circulation times compared with synthetic carriers [47].

Challenges and ethical considerations

Safety concerns

Offtarget mutational activity by programmable nucleases remains a central safety concern. CRISPRCas9 can introduce doublestrand breaks at genomic sites that share partial sequence complementarity with the intended target, potentially disrupting essential genes or activating oncogenic pathways. Highfidelity Cas9 variants, including eSpCas9 and HiFi Cas9, have achieved greater than 1,000fold reductions in offtarget activity while preserving ontarget efficiency, and computational tools now enable prospective prediction and avoidance of offtarget sites through genomewide screening [48]. For gene drive applications in vector species, the irreversibility of genetic modifications spreading through wild populations adds a distinctive dimension to offtarget risk; splitdrive and daisychain architectures with builtin geographic confinement thresholds have been developed as molecular containment strategies before any environmental release [49,50].

Immunogenicity to both delivery vehicles and therapeutic proteins presents additional safety considerations. Preexisting neutralising antibodies against natural AAV serotypes are highly prevalent in populations most burdened by malaria, and Cas9 protein from commonly used bacterial sources can elicit Tcell responses in a substantial proportion of individuals, necessitating development of humanised Cas9 variants or transient delivery approaches that limit exposure time [42,51]. For RNAibased approaches, immunostimulation through Tolllike receptor 3 and MDA5 pathways can paradoxically diminish silencing efficacy while inducing interferon responses; chemical modifications including 2'Omethylation have been successfully applied to mitigate these effects in approved siRNA therapeutics [52].

Ethical and regulatory considerations

The ethical dimensions of gene therapy for parasitic diseases extend beyond the familiar concerns about somatic versus germline editing that have dominated human genetics discourse. Populationlevel gene drive deployment raises distinct questions about interspecies ethics, ecosystem perturbation, and the governance of interventions that could affect nonconsenting communities across national boundaries [50]. The Convention on Biological Diversity maintains a precautionary approach to environmental gene drive release, and no jurisdiction has yet established a complete environmental risk assessment framework. The Target Malaria consortium's engagement model in Burkina Faso, Mali, and Uganda, which incorporates villagelevel deliberation and national ethics review, has been cited as an emerging benchmark for community engagement, though concerns about power asymmetries in NorthSouth research partnerships and tokenistic consultation remain [2].

Regulatory fragmentation presents a practical barrier that is particularly acute for diseases concentrated in low and middleincome countries. The FDA and European Medicines Agency have established specific frameworks for gene therapy medicinal products, but national regulatory authorities across subSaharan Africa and South Asia generally lack dedicated legislation, trained evaluators, and laboratory capacity for characterising advanced therapy products [53]. The African Medicines Agency, established in 2022, offers a potential harmonisation pathway, though resource constraints impede complex product review capacity.

Accessibility and cost

Economic accessibility represents perhaps the most formidable barrier. Approved gene therapies carry prices of USD 1.53.5 million

per patient, a figure that dwarfs the GDP per capita of the countries where parasitic diseases are endemic [54]. Sustainable access strategies will require openaccess licensing through mechanisms such as the Medicines Patent Pool, tiered pricing models that draw on the precedents established by antiretroviral therapy programmes, and investment in decentralised manufacturing technologies including plantbased bioreactor systems. Product Development Partnerships such as PATH MVI, DNDi, and the Medicines for Malaria Venture have embedded equity mandates into their development pipelines, though the financial architecture required to support equitable access at scale remains incompletely developed [55,56].

Future directions

Precision medicine

Precision medicine in this context refers to the customisation of therapeutic strategies according to individual genetic profiles, parasite strain characteristics, and local epidemiological patterns. Host genetic variation significantly influences susceptibility to parasitic diseases; the sickle cell trait (HbAS) confers partial protection against severe malaria through mechanisms involving reduced parasite replication in abnormal erythrocytes, while Duffy antigen negativity provides natural resistance to *P. vivax* infection by blocking the receptormediated invasion pathway [57,58]. Understanding these resistance mechanisms at a molecular level creates opportunities to develop targeted gene therapies that mimic protective traits in susceptible individuals, and to identify host genetic factors that predict treatment response or toxicity [4]. Genome sequencing of parasite populations has additionally revealed geographical variation in virulence determinants and drug resistance mutations, supporting the development of regionally calibrated interventions [59].

Artificial intelligence in gene editing

Artificial intelligence and machine learning methods are increasingly being applied to improve the accuracy and efficiency of gene editing. Computational models can predict offtarget cleavage sites from guide RNA sequences and genome context, enabling prescreening of guide designs to minimise unintended mutational damage [60]. Beyond offtarget prediction, machine learning algorithms have been applied to identify parasite genes essential for virulence, immune evasion, or transmission from largescale genomic and transcriptomic datasets, accelerating target discovery beyond the pace achievable through conventional laboratory screening [4]. For gene drive systems, ecological modelling supported by epidemiological data can predict transmission dynamics and evolutionary responses before field release, improving safety assessment and regulatory decisionmaking [61].

Combined immunotherapy and gene therapy

The combination of gene therapy with immunotherapy represents a strategy that addresses the limitations of each approach individually. Many parasites have evolved sophisticated mechanisms to suppress or evade host immune responses, and genetic interventions that disrupt these mechanisms may be substantially more effective when combined with immunotherapeutic agents that restore or enhance immune function [4]. Genetically engineered Tcells modified ex vivo to express receptors specific for parasite antigens have shown promise in preclinical models, and cytokine gene therapy using IL12 and IFNgamma can activate macrophagemediated killing of intracellular parasites including *Leishmania* and *Toxoplasma* [62,63]. Nanotechnologybased delivery systems may facilitate such combinations by enabling codelivery of genetic and immunomodulatory payloads to the same target cells, reducing the complexity of multicomponent regimens [64].

Conclusion

Gene therapy has advanced from speculative concept to demonstrated capability for parasitic disease intervention, with proofofconcept studies establishing the feasibility of parasite genome editing, vector population modification, host genetic resistance, and RNAimediated gene silencing across the major parasitic diseases of humans. The technological foundations, principally CRISPRCas9 and complementary programmable nuclease platforms, have matured to the point where preclinical development is proceeding for multiple applications, and early field evaluation of gene drive mosquitoes has produced promising results against patientderived malaria parasites. The delivery challenge remains central, with no single platform yet achieving the optimal combination of efficiency, specificity, safety, and deployability that would enable largescale implementation in endemic settings. Viral vectors offer high efficiency but face immunogenicity barriers and manufacturing constraints; nonviral alternatives reduce immunological risk but generally at lower

efficiency; and the thermostability requirements for tropical field deployment remain incompletely addressed across all platforms.

Safety and governance concerns, particularly those surrounding offtarget mutations and environmental gene drive release, require continued attention and the development of robust containment and confinement strategies. Regulatory capacity in endemic countries needs substantial strengthening if clinical trials and eventual deployment are to proceed responsibly. Economic barriers, with current gene therapy pricing models placing these interventions far beyond the reach of affected populations, demand innovative access mechanisms including open licensing, tiered pricing, and investment in lowcost manufacturing. The integration of artificial intelligenceassisted design, precision medicine calibrated to local strain variation, and combination strategies that merge genetic and immunotherapeutic approaches offers a pathway toward more effective and accessible interventions. The populations most affected by parasitic diseases have waited decades for transformative technologies that can break cycles of infection and poverty. Gene therapy, if developed and deployed with deliberate attention to equity and access, could contribute meaningfully to that transformation.

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