AnimalSci Publisher®
Animal Molecular Breeding, 2024, Vol.14, No.6, 345-353
http://animalscipublisher.com/index.php/amb

Review Article Open Access

A Review of Gene Editing Technologies in Poultry Breeding: Focus on CRISPR/Cas9

Yanlin Wang, Qiqi Zhou, Jia Chen
Tropical Animal Resources Research Center, Hainan Institute of Tropical Agricultural Resources, Sanya, 572000, Hainan, China
Corresponding author: jia.chen@hitar.org
Animal Molecular Breeding, 2024, Vol.14, No.6 doi: 10.5376/amb.2024.14.0036
Received: 03 Nov., 2024
Accepted: 05 Dec., 2024
Published: 16 Dec., 2024

Preferred citation for this article:
Wang Y.L., Zhou Q.Q., and Chen J., 2024, A review of gene editing technologies in poultry breeding: focus on CRISPR/Cas9, Animal Molecular Breeding, 14(6): 345-353 (doi: 10.5376/amb.2024.14.0036)

Abstract This study reviews various gene editing techniques, including CRISPR/Cas9, TALENs, and ZFNs, and emphasizes their advantages over traditional breeding. It analyzes CRISPR/Cas9 in detail, explores the use of gene editing to improve the growth, meat quality, disease resistance, and reproductive traits of poultry, reviews the ethical considerations, regulatory challenges, and safety assessments of gene edited poultry, and provides practical insights into the potential and limitations of using CRISPR/Cas9 to enhance disease resistance. Finally, it discusses future innovations in gene editing tools, integration of multi omics methods, and industry application prospects. This study aims to emphasize the transformative potential of gene editing in poultry breeding and call for ongoing research to optimize the technology and address regulatory and ethical challenges.
Keywords Poultry breeding; Gene editing; CRISPR/Cas9; TALEN; ZFN

1 Introduction

Poultry breeding has long been a cornerstone of agricultural practices, aimed at enhancing desirable traits such as growth rate, feed efficiency, disease resistance, and meat quality. Traditional breeding methods have relied on selective breeding and crossbreeding to achieve genetic improvements. However, these methods are often time-consuming and limited by the genetic variability present within the breeding population. The advent of molecular genetics and biotechnology has introduced new avenues for accelerating genetic gains in poultry, offering more precise and efficient tools for genetic improvement (Khwatenge and Nahashon, 2021).

The emergence of gene editing technologies, particularly the CRISPR/Cas9 system, has revolutionized the field of genetic engineering in poultry. CRISPR/Cas9 allows for precise, cost-effective, and user-friendly genome editing, enabling researchers to modify gene functions, target specific genetic loci, and introduce or regulate genetic information in poultry genomes (Vilela et al., 2020). This technology has been successfully applied to enhance disease resistance, such as rendering chickens resistant to avian leukosis virus subgroup J through targeted gene deletions (Koslová et al., 2020). Additionally, CRISPR/Cas9 has been utilized to develop recombinant vaccines and improve host-virus interactions, showcasing its potential in advancing poultry health and productivity.

This study explores the progress of applying CRISPR/Cas9 to poultry species, evaluates its potential benefits and limitations, discusses its impact on the poultry industry, and analyzes the current status and future prospects of gene editing technology in poultry breeding. This study aims to emphasize the transformative impact of CRISPR/Cas9 on poultry farming and its role in addressing industry challenges.

2 Gene Editing Technologies in Poultry Breeding

2.1 Overview of gene editing tools (CRISPR/Cas9, TALEN, ZFN)

Gene editing technologies have revolutionized the field of poultry breeding, offering precise and efficient methods to modify the avian genome. The most prominent tools include CRISPR/Cas9, TALEN (Transcription Activator-Like Effector Nucleases), and ZFN (Zinc Finger Nucleases) (Gupta et al., 2019). Among these, CRISPR/Cas9 stands out due to its simplicity, cost-effectiveness, and versatility. CRISPR/Cas9 allows for targeted gene modifications by utilizing a guide RNA to direct the Cas9 nuclease to specific DNA sequences, enabling precise cuts and subsequent gene editing (Khwatenge and Nahashon, 2021). TALEN and ZFN, while also effective, are more complex and less user-friendly compared to CRISPR/Cas9. TALENs use engineered proteins to bind and cut specific DNA sequences, whereas ZFNs employ zinc finger domains to achieve similar outcomes. Despite their differences, all three technologies have been successfully applied in various organisms, including poultry, to achieve targeted genetic modifications (Figure 1) (Islam et al., 2020).

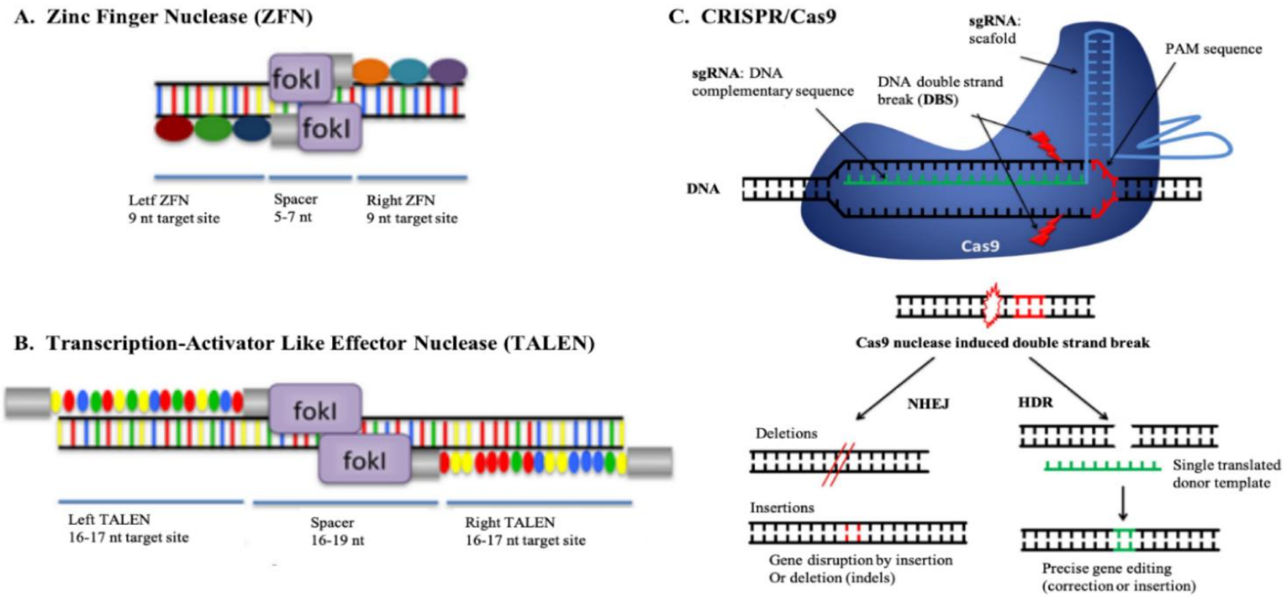

Figure 1 Nuclease-based genome editors (Adopted from Islam et al., 2020)

Image caption: (A). Zinc Finger Nuclease (B). Transcription-Activator Like Effector Nuclease (TALEN). (C). Schematic diagram showing genome editing using CRISPR/Cas9 system. The Cas9 induces DNA double-strand break (DSB) which are repaired either by imperfect nonhomologous end-joining (NHEJ) to generate insertion or deletion (indels) or if a repair is provided, by homology-directed repair (HDR) (Adopted from Islam et al., 2020)

2.2 Advantages of gene editing over traditional breeding methods

Gene editing technologies offer several advantages over traditional breeding methods. Traditional breeding relies on selective mating and phenotypic selection, which can be time-consuming and less precise. In contrast, gene editing allows for the direct modification of specific genes, leading to faster and more accurate results. For instance, CRISPR/Cas9 has been used to introduce desirable traits in poultry, such as disease resistance and improved production characteristics, which would be challenging to achieve through conventional breeding (Vilela et al., 2020). Additionally, gene editing can overcome the limitations of genetic diversity in breeding populations by introducing new genetic variations that are not present in the existing gene pool. This capability is particularly beneficial in enhancing traits that are difficult to improve through traditional methods, such as resistance to avian diseases and improved feed efficiency.

2.3 Challenges in applying gene editing in poultry

Despite its potential, the application of gene editing in poultry breeding faces several challenges. One significant hurdle is the difficulty in accessing and manipulating poultry zygotes, which complicates the gene editing process (Oishi et al., 2016). Additionally, there are technical challenges related to the efficiency and specificity of gene editing tools. For example, off-target effects, where unintended genetic modifications occur, remain a concern with CRISPR/Cas9 technology (Zhang et al., 2019). Furthermore, ethical and regulatory issues surrounding the use of gene editing in animals pose additional challenges. There is ongoing debate about the acceptability and safety of genetically modified organisms (GMOs), which can impact the adoption of these technologies in the poultry industry. Addressing these challenges requires continued research and development to improve the precision and efficiency of gene editing tools, as well as efforts to engage with regulatory bodies and the public to ensure the responsible use of these technologies in poultry breeding.

3 CRISPR/Cas9 Technology in Poultry Breeding

3.1 Mechanism of CRISPR/Cas9 in gene editing

The CRISPR/Cas9 system is a revolutionary genome editing tool that allows for precise, cost-effective, and user-friendly modifications of genomes across various organisms, including poultry (Liu and Zhang, 2024; Wu and Li, 2024). The mechanism involves the use of a guide RNA (gRNA) to direct the Cas9 nuclease to a specific DNA sequence, where it introduces a double-strand break. This break can then be repaired by the cell's natural repair mechanisms, either through non-homologous end joining (NHEJ) or homology-directed repair (HDR) (Oishi et al., 2016). The simplicity and efficiency of this system have made it a powerful tool for targeted mutagenesis and gene knockout in chickens, as demonstrated by successful gene editing in chicken primordial germ cells (PGCs) and somatic tissues (Khwatenge and Nahashon, 2021).

3.2 Applications of CRISPR/Cas9 in poultry genetics

CRISPR/Cas9 technology has been applied in various ways to advance poultry genetics. One significant application is the development of disease-resistant poultry. For instance, CRISPR/Cas9 has been used to edit viral genomes, paving the way for novel and multiplex viral vectored poultry vaccines (Vilela et al., 2020). Additionally, the technology has enabled the creation of genetically modified chickens with specific traits, such as low-allergenicity eggs and enhanced disease resistance (Chojnacka-Puchta and Sawicka, 2020). Researchers have also utilized CRISPR/Cas9 to produce transgenic progeny by targeting specific loci in chicken PGCs, demonstrating the potential for efficient genetic modification in birds (Dimitrov et al., 2016). Furthermore, the technology has been employed to study gene functions and regulatory mechanisms in chicken embryos, providing insights into developmental biology and gene expression (Gandhi et al., 2017).

3.3 Limitations and risks of CRISPR/Cas9 technology

Despite its numerous advantages, CRISPR/Cas9 technology has several limitations and risks that need to be addressed. One major concern is the potential for off-target effects, where the Cas9 nuclease may introduce unintended mutations at sites other than the target locus. Although some studies have reported no detectable off-target mutations, the risk remains a significant challenge (Bai et al., 2016). Another limitation is the efficiency of HDR, which is often lower than NHEJ, making precise gene editing more difficult (Antonova et al., 2018). Additionally, the delivery of CRISPR/Cas9 components into chicken cells, particularly zygotes, poses technical challenges that need to be overcome for broader application in poultry breeding. Finally, ethical and regulatory considerations surrounding the use of gene editing in animals must be carefully navigated to ensure responsible and acceptable use of this technology (Véron et al., 2015).

4 Applications of Gene Editing in Poultry Traits Improvement

4.1 Enhancing growth and meat quality in poultry

Gene editing technologies, particularly CRISPR/Cas9, have shown significant potential in enhancing growth and meat quality in poultry. By targeting specific genes associated with muscle development and growth, researchers have been able to achieve substantial improvements. For instance, the combination of CRISPR with yeast Rad52 (yRad52) has been used to enhance targeted genomic DNA editing in chicken cells, leading to increased efficiency in gene modifications. This approach has resulted in a 36.7% editing efficiency in the myostatin gene, which is known to regulate muscle growth, thereby potentially improving meat yield and quality in poultry (Wang et al., 2017). Additionally, CRISPR/Cas9 technology allows for precise modifications that can lead to the development of poultry with desirable traits, such as increased muscle mass and reduced fat content, which are critical for meat production (Khwatenge and Nahashon, 2021).

4.2 Improving disease resistance using gene editing

One of the most promising applications of gene editing in poultry is the enhancement of disease resistance. CRISPR/Cas9 has been effectively used to confer resistance to viral pathogens that pose significant threats to poultry health. For example, precise editing of the *NHE1* gene in chickens has rendered them resistant to the J subgroup of avian leukosis virus (ALV-J). This was achieved by introducing a single amino acid deletion in the gene encoding the receptor required for ALV-J infection, resulting in chickens that are resistant to this virus without any visible side effects (Koslová et al., 2020). This demonstrates the potential of CRISPR/Cas9 to create poultry breeds that are less susceptible to diseases, thereby reducing the need for antibiotics and improving overall flock health (Wang et al., 2022).

4.3 Modifying reproductive traits for better efficiency

Gene editing also holds promise for improving reproductive traits in poultry, which can lead to better breeding efficiency and productivity. By targeting genes involved in reproductive processes, researchers can enhance traits such as egg production, fertility, and hatchability. CRISPR/Cas9 technology has been used to modify the genomes of poultry to better understand and manipulate reproductive traits. For instance, the development of innovative genome-edited avian models, including specific chicken bioreactors and knock-in/out chickens, has facilitated the study and improvement of reproductive efficiency (Chojnacka-Puchta and Sawicka, 2020). These advancements can lead to more efficient breeding programs and higher productivity in the poultry industry.

5 Ethical, Regulatory, and Safety Considerations

5.1 Ethical concerns surrounding gene editing in poultry

The application of CRISPR/Cas9 in poultry breeding raises significant ethical concerns. One of the primary issues is the potential for unforeseen and undesirable effects, which could impact animal welfare and biodiversity. Ethical debates often focus on the morality of altering the genetic makeup of living organisms, particularly when it involves germline modifications that can be passed on to future generations (Zhang et al., 2020). Additionally, there is concern about the potential exploitation of this technology for eugenics or other morally contentious purposes (Shinwari et al., 2018). The ease and precision of CRISPR/Cas9 make it a powerful tool, but this also means that its misuse could have far-reaching consequences, necessitating stringent ethical guidelines and public discourse to ensure responsible use (Véron et al., 2015).

5.2 Regulatory landscape for gene editing technologies

The regulatory landscape for gene editing technologies like CRISPR/Cas9 is complex and varies significantly across different regions. Many countries are still in the process of developing comprehensive regulatory frameworks to address the unique challenges posed by these technologies. For instance, the non-traceability of modifications and the blurring of boundaries between natural and genetically modified organisms call for a rethinking of existing regulatory approaches (Bartkowski et al., 2018). International standards and guidelines are crucial to harmonize regulations and ensure the safe and ethical application of gene editing. Organizations such as the National Academies and the Biological Toxins and Weapons Convention (BTWC) are working towards establishing these standards, but more engaged international dialogue is needed to address the rapid advancements in this field (DiEuliis and Giordano, 2017).

5.3 Safety and risk assessment of gene-edited poultry products

Safety and risk assessment are critical components of the regulatory process for gene-edited poultry products. The primary concern is the potential for off-target effects, which could lead to unintended genetic changes with unknown consequences (Dimitrov et al., 2016; Memi et al., 2018). Additionally, the long-term impacts on animal health and the environment need to be thoroughly evaluated. Studies have shown that precise CRISPR/Cas9 editing can confer resistance to diseases such as avian leukosis virus without visible side effects, indicating the potential for safe application (Koslová et al., 2020). However, continuous monitoring and periodic assessment are essential to ensure that any risks are identified and mitigated promptly. The involvement of biosafety and biosecurity communities in these assessments is also crucial to address the dual-use potential of gene editing technologies.

6 Case Study: Application of CRISPR/Cas9 in Poultry Breeding

6.1 Overview of the case study and selected poultry species

This case study focuses on the application of CRISPR/Cas9 technology in chickens, specifically targeting the avian leukosis virus subgroup J (ALV-J). ALV-J is a significant pathogen in poultry, causing economic losses due to its impact on chicken health and productivity. The study selected chickens as the model species due to their importance in the poultry industry and the feasibility of genetic modifications in this species (Oishi et al., 2016).

6.2 Implementation of CRISPR/Cas9 for disease resistance

The implementation of CRISPR/Cas9 in this case involved precise editing of the chicken Na^+/H^+ exchanger type 1 (*chNHE1*) gene, which encodes a receptor critical for ALV-J entry into chicken cells. Researchers introduced a single amino acid deletion at tryptophan residue number 38 (W38) in the *chNHE1* gene. This specific mutation was chosen because W38 is essential for the virus's ability to infect chicken cells (Figure 2) (Koslová et al., 2020).

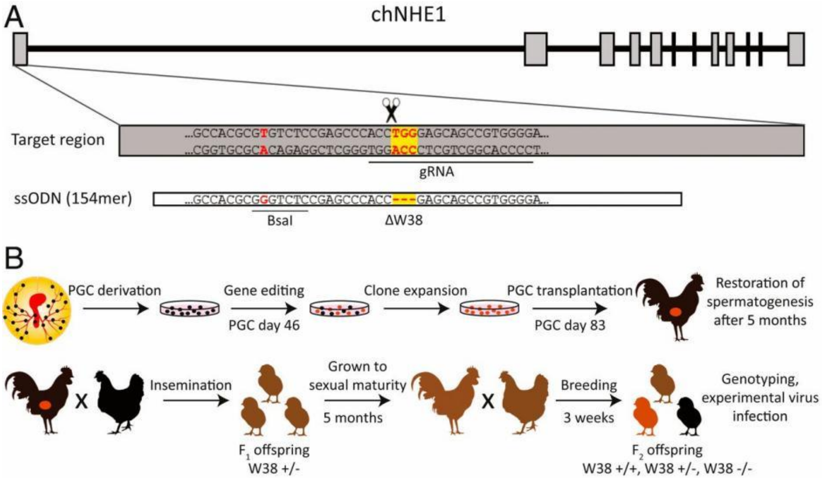

Figure 2 Design of guide RNA and homologous recombination ssODN for CRISPR/Cas9 gene editing of chicken ALV-J receptor *chNHE1* in primordial germ cells (Adopted from Koslová et al., 2020)

Image caption: (A) The structure of coding exons and introns of *chNHE1* (Top), the CRISPR/Cas9 target sequence of exon 1 with the guide RNA (gRNA) complementary sequence (underlined) and the TGG triplet encoding W38 (red on the yellow background; Middle), and the central part of the ssODN template for homologous recombination with deleted TGG triplet and a single nucleotide substitution (in red) creating the BsaI restriction site (Bottom). (B) Preparation of ΔW38 chickens: schematic representation of the workflow and timeline (Adopted from Koslová et al., 2020)

The CRISPR/Cas9 system was used to delete the W38 residue in chicken primordial germ cells (PGCs). These edited PGCs were then used to produce gene-edited chickens. The resulting ΔW38 homozygous chickens were tested for resistance to ALV-J both in vitro and in vivo. The results showed that ΔW38 homozygous chickens were resistant to ALV-J, whereas ΔW38 heterozygotes and wild-type birds remained susceptible (Table 1) (Koslová et al., 2020).

Table 1 ALV-J viremia in chickens inoculated with RCASBP(J)GFP (Adopted from Koslová et al., 2020)

chNHE1 genotype	Chicken no.	Age of chicken, d	Virus titer, IU/mL	
			6 d p.i.	13 d p.i.
$W38^{-/-}$	604	28	0	0
$W38^{-/-}$	606	28	0	0
$W38^{-/-}$	611	20	0	0
$W38^{-/-}$	612	20	0	0
$W38^{-/-}$	615	14	0	0
$W38^{+/-}$	601	28	0	101
$W38^{+/-}$	603	28	0	102
$W38^{+/-}$	605	28	0	101
$W38^{+/-}$	610	20	102	103
$W38^{+/-}$	616	20	0	103
$W38^{+/-}$	618	14	0	102
$W38^{+/-}$	619	14	101	103
$W38^{+/+}$	617	14	102	103

6.3 Analysis of results and implications for future breeding

The results of this study demonstrated that precise CRISPR/Cas9 gene editing could confer resistance to ALV-J in chickens without any visible side effects. This finding is significant as it highlights the potential of CRISPR/Cas9 technology to enhance disease resistance in poultry, thereby reducing economic losses and improving animal welfare (Islam et al., 2020; Gul et al., 2022).

The successful implementation of CRISPR/Cas9 in this case study opens up new possibilities for breeding disease-resistant poultry. By targeting specific genes associated with disease susceptibility, it is possible to develop chicken strains that are resilient to various pathogens. This approach could significantly decrease the reliance on antibiotics and vaccines, promoting more sustainable and health-conscious poultry farming practices (Vilela et al., 2020).

7 Future Directions in Gene Editing for Poultry Breeding

7.1 Innovations in gene editing tools and techniques

The CRISPR/Cas9 system has revolutionized gene editing in poultry, allowing for precise modifications in the avian genome. Recent advancements have focused on improving the efficiency and specificity of these tools. For instance, CRISPR technology has been successfully applied to modify genes in chickens and quails, enhancing genetic variations that are beneficial for poultry production. However, there is still room for improvement in the techniques used to achieve heritable edited traits in birds, which are currently quite involved and specific to avian reproductive biology (Tizard et al., 2019). Future innovations may include more refined methods for germline editing and the development of new gene-editing tools that can overcome the current limitations of CRISPR/Cas9 (Preethi et al., 2020).

7.2 Potential for integrating multi-omics approaches

The integration of multi-omics approaches, including genomics, transcriptomics, proteomics, and metabolomics, holds significant potential for advancing poultry breeding. These technologies can provide comprehensive insights into the genetic and molecular basis of important traits such as disease resistance, productivity, and welfare (Dehau et al., 2022). The decreasing cost of omics technologies makes their implementation in routine poultry monitoring systems more feasible, potentially leading to the development of diagnostic tests based on disease-specific biomarkers (Goossens et al., 2022). By combining multi-omics data with gene editing, researchers can achieve a more precise and holistic understanding of complex traits, thereby enhancing the effectiveness of breeding programs (Langridge and Fleury, 2011; Mahmood et al., 2022).

7.3 Prospects for commercialization and industry adoption

The commercialization and industry adoption of gene editing technologies in poultry breeding face several challenges, including regulatory hurdles and public perception. The regulatory landscape for genome-edited animals varies significantly across countries, which could lead to disparities in the adoption of these technologies and potential trade disruptions (Bishop and Eenennaam, 2020). Despite these challenges, the potential benefits of gene editing, such as improved disease resistance and enhanced productivity, make it an attractive option for the poultry industry. The successful commercialization of gene-edited poultry will likely depend on achieving regulatory harmony and addressing public concerns about the safety and ethics of these technologies. As the technology matures and becomes more widely accepted, it is expected that gene editing will play a crucial role in the future of poultry breeding (Khwatenge and Nahashon, 2021).

8 Concluding Remarks

The application of CRISPR/Cas9 technology in poultry breeding has shown significant promise and progress. CRISPR/Cas9 has been successfully utilized to modify gene functions for various purposes, including transcriptional regulation, gene targeting, and epigenetic modification in poultry species, particularly chickens and quails. This technology has enabled the precise editing of viral genomes, aiding in the development of novel poultry vaccines and enhancing resistance to avian diseases. Additionally, CRISPR/Cas9 has been employed to achieve targeted mutagenesis and homologous recombination in chicken cell lines, demonstrating high efficiency and specificity. The optimization of CRISPR/Cas9 for early chick embryos has further improved the efficiency and specificity of gene knockouts, facilitating advanced genetic studies in poultry. Moreover, innovative delivery systems, such as using Marek's disease virus, have been explored to enhance the practical application of CRISPR/Cas9 in poultry.

Future research should focus on addressing the limitations of CRISPR/Cas9 technology in poultry breeding. One critical area is improving the efficiency and precision of gene editing to minimize off-target effects and ensure stable genetic modifications. Developing more robust delivery systems for CRISPR/Cas9 components, such as gesicle technology, could enhance the practical application of this technology in poultry. Additionally, exploring the use of tissue-specific promoters and spatiotemporal control strategies could further refine gene editing outcomes and reduce unintended consequences. Research should also investigate the long-term effects of CRISPR/Cas9-mediated genetic modifications on poultry health and productivity to ensure the safety and sustainability of this technology in the poultry industry.

The integration of CRISPR/Cas9 technology into poultry breeding holds transformative potential for the industry. By enabling precise genetic modifications, this technology can enhance disease resistance, improve production traits, and contribute to the development of innovative poultry vaccines. The advancements in CRISPR/Cas9 applications in poultry not only promise to improve the efficiency and sustainability of poultry production but also open new avenues for scientific research and biotechnological innovations. As research continues to address the current limitations and optimize the use of CRISPR/Cas9, the full potential of gene editing in poultry breeding will likely be realized, leading to significant advancements in the field.

Acknowledgments
We are grateful to Mrs. Yuan for critically reading the manuscript and providing valuable feedback that improved the clarity of the text.

Conflict of Interest Disclosure
The authors affirm that this research was conducted without any commercial or financial relationships that could be construed as a potential conflict of interest.

References
Antonova E., Glazova O., Gaponova A., Eremyan A., Zvereva S., Grebenkina N., Volkova N., and Volchkov P., 2018, Successful CRISPR/Cas9 mediated homologous recombination in a chicken cell line, F1000Research, 7: 238.

Bai Y., He L., Li P., Xu K., Shao S., Ren C., Liu Z., Wei Z., and Zhang Z., 2016, Efficient genome editing in chicken DF-1 cells using the CRISPR/Cas9 system, G3: Genes|Genomes|Genetics, 6: 917-923.
https://doi.org/10.1534/g3.116.027706

Bartkowski B., Theesfeld I., Pirscher F., and Timaeus J., 2018, Snipping around for food: Economic, ethical and policy implications of CRISPR/Cas genome editing, Geoforum, 96: 172-180.
https://doi.org/10.1016/J.GEOFORUM.2018.07.017

Bishop T., and Eenennaam A., 2020, Genome editing approaches to augment livestock breeding programs, Journal of Experimental Biology, 223(Suppl_1): jeb207159.
https://doi.org/10.1242/jeb.207159

Chojnacka-Puchta L., and Sawicka D., 2020, CRISPR/Cas9 gene editing in a chicken model: current approaches and applications, Journal of Applied Genetics, 61: 221-229.
https://doi.org/10.1007/s13353-020-00537-9

Dehau T., Ducatelle R., Immerseel F., and Goossens E., 2022, Omics technologies in poultry health and productivity-part 1: current use in poultry research, Avian Pathology, 51: 407-417.
https://doi.org/10.1080/03079457.2022.2086447

DiEuliis D., and Giordano J., 2017, Gene editing using CRISPR/Cas9: implications for dual-use and biosecurity, Protein and Cell, 9: 239-240.
https://doi.org/10.1007/s13238-017-0493-4

Dimitrov L., Pedersen D., Ching K., Yi H., Collarini E., Izquierdo S., Lavoir M., and Leighton P., 2016, Germline gene editing in chickens by efficient CRISPR-Mediated homologous recombination in primordial germ cells, PLoS One, 11(4): e0154303.
https://doi.org/10.1371/journal.pone.0154303

Gandhi S., Piacentino M., Vieceli F., and Bronner M., 2017, Optimization of CRISPR/Cas9 genome editing for loss-of-function in the early chick embryo, Developmental Biology, 432(1): 86-97.
https://doi.org/10.1016/j.ydbio.2017.08.036

Goossens E., Dehau T., Ducatelle R., and Immerseel F., 2022, Omics technologies in poultry health and productivity-part 2: future applications in the poultry industry, Avian Pathology, 51: 418-423.
https://doi.org/10.1080/03079457.2022.2085545

Gul H., Habib G., Khan I., Rahman S., Khan N., Wang H., Khan N., and Liu Y., 2022, Genetic resilience in chickens against bacterial, viral and protozoal pathogens, Frontiers in Veterinary Science, 9: 1032983.
https://doi.org/10.3389/fvets.2022.1032983

Gupta D., Bhattacharjee O., Mandal D., Sen M., Dey D., Dasgupta A., Kazi T., Gupta R., Sinharoy S., Acharya K., Chattopadhyay D., Ravichandiran V., Roy S., and Ghosh D., 2019, CRISPR-Cas9 system: a new-fangled dawn in gene editing, Life Sciences, 232: 116636.
https://doi.org/10.1016/j.lfs.2019.116636

Islam M., Rony S., Rahman M., Çınar M., Villena J., Uddin M., and Kitazawa H., 2020, Improvement of disease resistance in livestock: application of immunogenomics and CRISPR/Cas9 technology, Animals, 10(12): 2236.
https://doi.org/10.3390/ani10122236

Khwatenge C., and Nahashon S., 2021, Recent advances in the application of CRISPR/Cas9 gene editing system in poultry species, Frontiers in Genetics, 12: 627714.
https://doi.org/10.3389/fgene.2021.627714

Koslová A., Trefil P., Mucksová J., Reinišová M., Plachy J., Kalina J., Kučerová D., Geryk J., Krchlíková V., Lejčková B., and Hejnar J., 2020, Precise CRISPR/Cas9 editing of the NHE1 gene renders chickens resistant to the J subgroup of avian leukosis virus, Proceedings of the National Academy of Sciences, 117: 2108-2112.
https://doi.org/10.1073/pnas.1913827117

Langridge P., and Fleury D., 2011, Making the most of 'omics' for crop breeding, Trends in Biotechnology, 29(1): 33-40.
https://doi.org/10.1016/j.tibtech.2010.09.006

Liu X.H., and Zhang J., 2024, CRISPR-Cas9 technology in Bt genome editing and functional studies, Bt Research, 15(2): 53-64.
http://dx.doi.org/10.5376/bt.2024.15.0006

Mahmood U., Li X., Fan Y., Chang W., Niu Y., Li J., Qu C., and Lu K., 2022, Multi-omics revolution to promote plant breeding efficiency, Frontiers in Plant Science, 13: 1062952.
https://doi.org/10.3389/fpls.2022.1062952

Memi F., Ntokou A., and Papangeli I., 2018, CRISPR/Cas9 gene-editing: research technologies, clinical applications and ethical considerations, Seminars in Perinatology, 42(8): 487-500.
https://doi.org/10.1053/j.semperi.2018.09.003

Oishi I., Yoshii K., Miyahara D., Kagami H., and Tagami T., 2016, Targeted mutagenesis in chicken using CRISPR/Cas9 system, Scientific Reports, 6(1): 23980.
https://doi.org/10.1038/srep23980

Preethi A., Saravanan K., Kumar H., Virbhai D., Tyagi S., Rajawat D., Kaisa K., Bhushan B., and Panigrahi M., 2020, Advances in genome editing technology and its applications in poultry breeding, Journal of Entomology and Zoology Studies, 8: 1416-1423.

Shinwari Z., Tanveer F., and Khalil A., 2018, Ethical issues regarding CRISPR mediated genome editing, Current Issues in Molecular Biology, 26: 103-110.
https://doi.org/10.21775/cimb.026.103

Tizard M., Jenkins K., Cooper C., Woodcock M., Challagulla A., and Doran T., 2019, Potential benefits of gene editing for the future of poultry farming, Transgenic Research, 28: 87-92.
https://doi.org/10.1007/s11248-019-00139-0

Véron N., Qu Z., Kipen P., Hirst C., and Marcelle C., 2015, CRISPR mediated somatic cell genome engineering in the chicken, Developmental Biology, 407(1): 68-74.
https://doi.org/10.1016/j.ydbio.2015.08.007

Vilela J., Rohaim M., and Munir M., 2020, Application of CRISPR/Cas9 in understanding avian viruses and developing poultry vaccines, Frontiers in Cellular and Infection Microbiology, 10: 581504.
https://doi.org/10.3389/fcimb.2020.581504

Wang L., Yang L., Guo Y., Du W., Yin Y., Tao Z., and Lu H., 2017, Enhancing targeted genomic DNA editing in chicken cells using the CRISPR/Cas9 system, PLoS One, 12(1): e0169768.
https://doi.org/10.1371/journal.pone.0169768

Wang S., Qu Z., Huang Q., Zhang J., Lin S., Yang Y., Meng F., Li J., and Zhang K., 2022, Application of gene editing technology in resistance breeding of livestock, Life, 12(7): 1070.
https://doi.org/10.3390/life12071070

Wu J.N., and Li T.H., 2024, The impact of releasing *Aedes aegypti* mosquitoes edited by CRISPR in the wild on local ecosystems, Journal of Mosquito Research, 14(1): 10-17.
http://dx.doi.org/10.5376/jmr.2024.14.0002

Zhang D., Hussain A., Manghwar H., Xie K., Xie S., Zhao S., Larkin R., Qing P., Jin S., and Ding F., 2020, Genome editing with the CRISPR-Cas system: an art, ethics and global regulatory perspective, Plant Biotechnology Journal, 18: 1651-1669.
https://doi.org/10.1111/pbi.13383

Zhang J., Liu J., Yang W., Cui M., Dai B., Dong Y., Yang J., Zhang X., Liu D., Liang H., and Cang M., 2019, Comparison of gene editing efficiencies of CRISPR/Cas9 and TALEN for generation of MSTN knock-out cashmere goats, Theriogenology, 132: 1-11.
https://doi.org/10.1016/j.theriogenology.2019.03.029