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# Opinion of Academicians Regarding the Use of the CRISPR-Cas System in Türkiye

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**Abstract**
The field of gene control is huge and still much unexplored, and one way to modify and alter genes is called CRISPR. The purpose of this study is to discuss the application of the CRISPR-Cas system in Turkey and find out what genetics professors think about it, as it has been a hot topic lately. The study's findings came from a survey that was given to 26 academicians who agreed to engage in the research and are recognized authorities in their professions. According to 74.1 % of participants, CRISPR-Cas 9 was not well known in the Turkish genetics community and was only known by those in the field. The participants stated that the CRISPR method can be used to treat cancer and many genetic diseases 100 % of the time, to breed animals for high-yield breeds in animal husbandry 100 % of the time, to provide resistance and protection against many infectious diseases 84.6 % of the time, and to improve animal welfare 88.8 % of the time. Although 76.9 % of experts said that there is room for ethical debate over the use of CRISPR, 92.3 % of experts predicted that gene-edited animal farms will be built in the near future. A sizable majority of participants 92.3 % said that Turkey must develop and apply the CRISPR method to stay ahead of developed nations in genetic science, as well as for application in animal breeding and the treatment of genetic illnesses. Therefore, even if CRISPR technology has benefits and drawbacks, it's critical to create legislative guidelines that consider the environment, public health, food safety, global trade, and the moral implications of the CRISPR technique.

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## 1. Introduction

The system known as CRISPR/CRISPR-associated protein 9 (Cas9) is an adaptive immune system that is made up of numerous bacteria and archaea that carry Cas genes, related proteins, and CRISPR loci (Jiang et al., 2024). With the help of the Cas9 protein's nuclease activity, this system can detect exogenous DNA, split the DNA double helix, and allow base addition or deletion with subsequent DNA repair (Zhang et al., 2019). Gene regulation can be classified into transcriptional regulation and post-transcriptional regulation based on the gene expression process (Sorek et al., 2008). DNA regulation at the genetic level and chromatin regulation at the epigenetic level are two categories of transcriptional regulation (Silas et al., 2016). According to Zakrzewska and Burmistrz (2023), the Cas system is regarded as one of the genome editing methods that is helpful in functional genetic research, biotechnological applications, and medical research.

According to Jinek et al. (2012), genome editing technology allows for the precise targeted change of an organism's endogenous genes. To allow for the insertion, deletion, and replacement of certain target DNA sequences, an endonuclease specific to the task is employed to cleave DNA strands (Mali et al., 2013; Cui et al., 2020). Because of its speed and ease of use, CRISPR is changing the process of gene knockout (Nishimashu et al., 2014). The temporal and geographical modulation of CRISPR-Cas activity is still difficult to achieve, despite numerous advancements (Jinek et al., 2012).

Crop development and animal breeding can both benefit from the adaptable CRISPR-Cas9 approach (Li et al., 2024). This emphasizes how far technology has come in incorporating this into practical uses. Additionally, current uses of CRISPR-Cas-based nucleic acid detection in the livestock sector are highlighted. These uses include the identification of early embryo sex, specificity and composition of meat and dairy products, and developing infectious illnesses (Zhang, 2022).

Currently, there are numerous issues contributing to the decline in animal and agricultural output. Reduced yields may result in a shortage of nutrient-dense food compared to the world's demand (Shin et al., 2017). According to reports in this context, the use of CRISPR-Cas9 technology for genomic editing offers numerous benefits for product development (Jiang et al., 2013). It has been suggested that worldwide starvation can be avoided, particularly by utilizing transgenic-free CRISPR-Cas9 technology and animal breeding-based methods to increase food productivity (Rao et al., 2022).

Up until now, vaccination and antibiotic use have been the primary methods for controlling disease in cattle. On the other hand, chronic antibiotic usage exacerbates diseases, promotes resistance to dangerous microbes, and pollutes the environment (Tian et al., 2021). Furthermore, it seems that some viral diseases cannot be prevented by effective vaccinations (Aslam et al., 2018).

It has been established that the emergence of diseases is influenced by genetic variables, animal care and nutrition, and the production environment (Wang et al., 2022). Consequently, in an effort to make animals more resistant to illness, several scientists have recently focused on enhancing animal genomes (Van, 2019). The efficiency of livestock production will rise with the application of contemporary genetic and molecular biology techniques to enhance animals' immune systems and resistance to disease (Gonen et al., 2017). In fact, research have shown that CRISPR-Cas9 may be a more effective method for improving livestock's genetic makeup (Makarove and Koonin, 2015).

Prior studies utilizing CRISPR-Cas9 have concentrated on bacterial cells. CRISPR technology started to be applied in fields such as agriculture and medicine after (Zhang et al., 2020) publication. In every area of biology, the usage of genome editing technologies has grown quickly (Pickar and Gersbach, 2019). As a result, the CRISPPR-Cas9 system has rapidly evolved and grown over the course of the 12 years between 2012 and 2024 (Liao et al., 2021).

Scientists have been able to quickly alter the genomes of mice, rats, fruit flies, and plants like rice and wheat since CRISPR-Cas9 was demonstrated to function in mammalian cells (Chen et al., 2019). Innovation in applied and technological domains, such as health, agriculture, and aquaculture, has also been significantly influenced by technology (Katti et al., 2022).

This study aims to determine the advantages and disadvantages of the CRISPR-Cas9 based method, especially in animal husbandry, and to determine the opinions of academicians working in the field of genetics regarding its applicability in Turkey.

## 2. Materials and Methods

Experts in a field provide their thoughts and recommendations on a topic using the Delphi Method (Dalkey and Helmer, 1963). In order to achieve this, a series of inquiries about the CRISPR technique and its relevance in Turkey were developed and distributed via Google Forms to specialists who expressed interest in taking part in the research, after the identification of academicians with relevant expertise in the field of genetics. The study's Cronbach's alpha reliability coefficients were computed. According to Cortina (1993), an acceptable result for the dependability coefficient of the initial survey was 0.94, provided the alpha value was greater than 0.70. The study's data came from a survey that was completed with 26 academicians who operate in the field of genetics throughout Turkey and gave their consent to take part. The data from the surveys were analyzed using the SPSS 16 program. The mean, standard deviation, and percentage values of the data were computed and displayed in tables during statistical analysis.

**Table 1.** Participant individual characteristics

| Parameter | | n | % |
|---|---|---|---|
| Gender | Woman | 9 | 25.9 |
| | Man | 17 | 74.1 |
| Degree | Prof. Dr. | 1 | 3.8 |
| | Assoc. Dr | 7 | 26.9 |
| | Dr. Lecturer | 8 | 30.8 |
| | Research Assistant | 2 | 7.7 |
| | Expert Dr. | 2 | 7.7 |
| | Expert | 6 | 23.1 |

Examining Table 1, we see that the experts who took part in the survey were titled as follows: research assistant (n = 2), expert (n = 6), associate Dr. (n = 7), professor (n = 1), and research assistant (n = 2). According to 74.1 % of participants, the CRISPR technique is not well known in Turkey and is solely used by those in the genetics field (Figure 1).

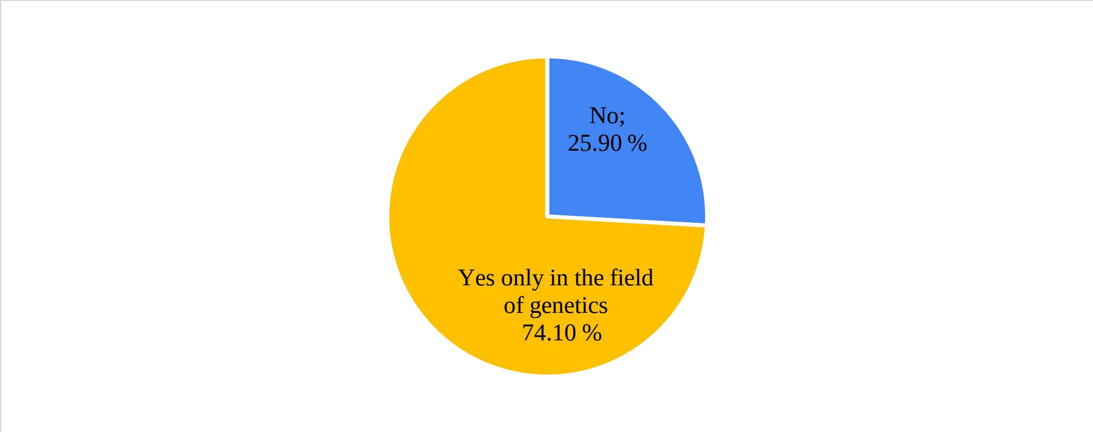


**Figure 1.** How well-known is CRISPR in Turkey's agricultural and animal husbandry fields?

**Table 2.** Questions and Answers about Applications of CRISPR

| Questions for Surveys | Yes | | No | |
|---|---|---|---|---|
| | n | % | n | % |
| Is there any promise for treating disorders impacting genetic health and cancer with Crispr cas9? | 26 | 100 | 0 | 0 |
| Can improvements in gene editing techniques benefit animal husbandry methods? | 26 | 100 | 0 | 0 |
| Can effective gene transfer and long-term stable transgenic expressions made possible by Crispr technology boost animal productivity? | 26 | 100 | 0 | 0 |
| Is it possible to prevent diseases in animals using the Crispr method? | 22 | 84.6 | 4 | 15.4 |
| Can animal welfare on farms be enhanced by the use of genome editors? | 21 | 80.8 | 5 | 19.2 |

Table 2 provides the responses to the study's inquiries regarding the domains in which CRISPR is used. Table 2 shows that a sizable percentage of experts concur that the CRISPR technique is used in animal breeding for high-yield breeds in livestock (100 %), in the treatment of cancer and many genetic diseases (100 %), in providing resistance and protection against many infectious diseases (84.6 %), and in animal breeding (100 %). Eighty-eight percent of them said it might be applied to improve animal welfare. Figure 2 displays the participants' perspectives on the usefulness of CRISPR technology in the field of animal husbandry.

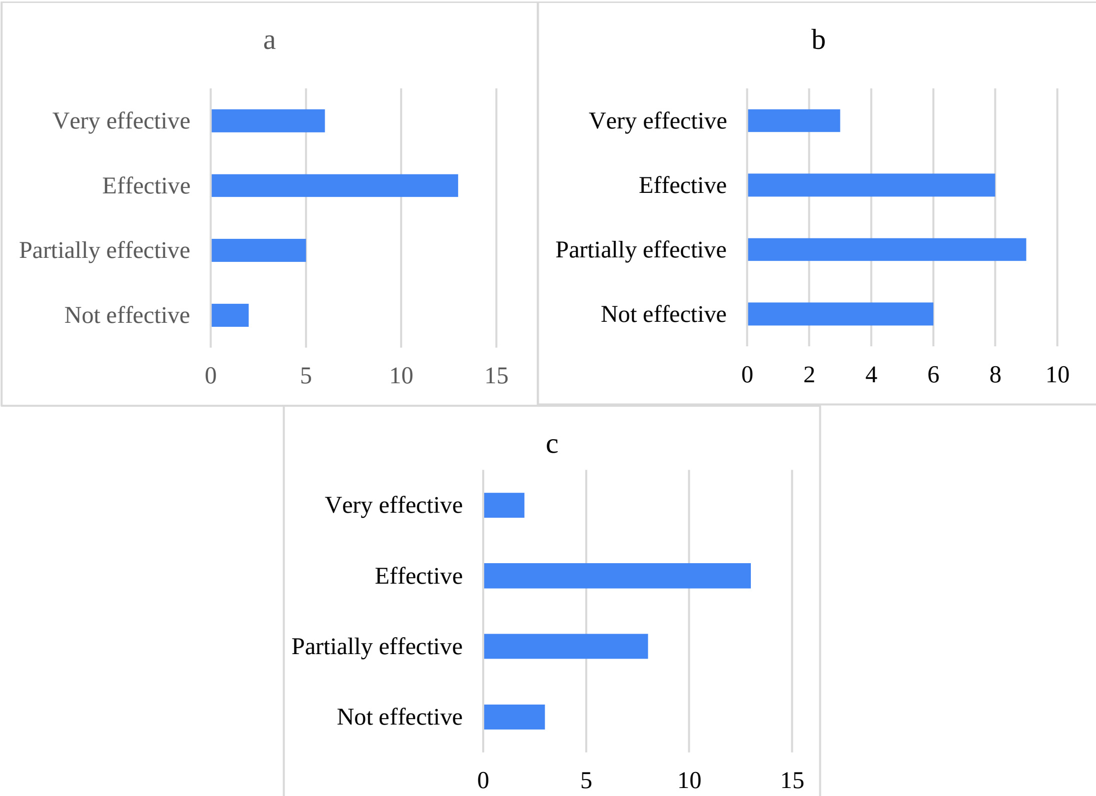


**Figure 2.** The degree of efficacy of CRISPR technology in animal husbandry applications

Examining Figure 2, one can see that professionals employ the CRISPR technology to produce high-yielding breeds. a- The effect of CRISPR on reaching high-yield breeds, b- The impact of CRISPR in disease control and prevention, c- The impact of CRISPR in improving animal welfare. 7.7 % of respondents say it is ineffective, 19.2 % say it is moderately effective, and 73.1 % say it is effective. Although 42.3 % of experts believe that the CRISPR approach is efficient in controlling and avoiding diseases, 57.7 % believe that it is an excellent method for enhancing animal welfare. Table 3 presents the views of the professionals who answered the survey with reference to the CRISPR technique.

**Table 3.** A few viewpoints regarding the CRISPR technique

| Questions | yes | | no | |
|---|---|---|---|---|
| | n | % | n | % |
| Do you believe that farm animals with altered genes will be able to be produced soon? | 24 | 92.3 | 2 | 7.7 |
| Could there be problems with food safety with goods derived from genetically engineered animals? | 20 | 76.9 | 6 | 23.1 |
| Could there be moral dilemmas with CRISPR/Cas 9? | 20 | 76.9 | 6 | 23.1 |

According to Table 3, 76.9 % of experts stated that goods derived from gene-edited animals are safe for consumption and could potentially raise ethical concerns in the future, despite 92.3 % of experts believing that gene-edited animal farms will be built soon. Table 4 presents the participants' perspectives regarding the suitability of the CRISPR technique in Turkey.

**Table 4.** Professional views regarding the suitability of the CRISPR technique in Turkey

| Questions | yes | | no | |
|---|---|---|---|---|
| | n | % | n | % |
| Does Turkey have the necessary infrastructure to use CRISPR technology? | 10 | 38.5 | 16 | 61.5 |
| Is it feasible to create and use the CRISPR system in Turkey as a tool for genome editing? | 24 | 92.3 | 2 | 7.7 |
| Do you believe Turkey should use the CRISPR method? | 24 | 92.3 | 2 | 7.7 |

Upon examining Table 4, it becomes evident that a mere 38.5 % of the experts believe that Turkey possesses the necessary infrastructure to effectively implement CRISPR technology. However, a substantial majority of responders (92.3 %) stated that Turkey should develop and apply the CRISPR system. Table 5 lists the benefits and drawbacks of using CRISPR technology in Turkey based on the opinions of the experts that took part in the survey.

**Table 5.** Lists the benefits and drawbacks of using CRISPR technology

| Advantages | n | % | Disadvantages | n | % |
|---|---|---|---|---|---|
| Creating breeds with high yields | 10 | 38.5 | Using technology in an unethical manner | 18 | 69.2 |
| In the defense against a range of illnesses | 7 | 26.9 | genetic illness | 4 | 15.4 |
| prevention of hereditary illnesses for which there is no known cure | 6 | 23.1 | Absence of expertise and experience | 2 | 7.7 |
| Increasing the animals' ability to fend off illnesses | 3 | 11.5 | Absence of infrastructure | 2 | 7.7 |
| Other | 26 | 100 | Other | 26 | 100 |

The benefits of CRISPR technology, as reported by the participants, when Table 5 is looked at, include producing high-yield breeds through animal breeding, preventing genetic diseases and cancer, making animals more resistant to disease, speeding up and improving the efficiency of production, and boosting the national economy by lowering production costs. The emergence of genetic abnormalities as a result of poor design, inexperience, and lack of infrastructure are among the drawbacks of CRISPR technology.

## 3. Discussion

To create high-yielding, stress-resistant types, scientists and breeding researchers are putting a lot of effort into their work. When paired with nanotechnology, CRISPR-Cas9 has become a powerful tool for raising the yields and quality of agricultural and animal products (Tsveta et al., 2021). Gene therapy can be used to repair or correct defective or undesirable genes in a cell (Akram et al., 2020). Gene therapy is the best course of treatment, particularly for hereditary illnesses that are not amenable to medication (Brokowski and Adli, 2019). It is now possible to execute gene therapy on both plants and animals (Akram et al., 2023; Ayanoğlu et al., 2020). The agriculture industry has begun to be the main user of the CRISPPR-Cas9 system (Naik et al., 2022). Cas9 technology has been employed in the growth of grains, vegetables, fruits, and nuts with high nutritional value because the production of nutritious goods mostly depends on the fertility of the seed and the condition of the soil (Panahi et al., 2022). In this regard, the CRISPR-Cas9 system, which is presently employed in tobacco, rice and Arabidopsis, offers enormous development potential (Kan et al., 2021). Research on heat-resistant grain production and drought has been moving quickly lately. The study concluded that the livestock industry may employ the CRISPR technology to produce 100 % high-yield breeds of animals through animal breeding. Actually, a lot of study and application of this method has been done recently to enhance animal heredity, reproduction and nutritional levels (Perisse et al., 2020). Research indicates that improving animal husbandry practices can boost the output of animal products, including meat and milk yield, fertility and egg and wool yield (Zhou et al., 2019). Growth, hairiness and milk-producing qualities have all enhanced as a result of research into changing sheep genomes using gene editing (Zhou et al., 2020). According to a recent article, it is anticipated that lambs resistant to disease would be produced by modifying the HYAL2 and PrP genes using the CRISPR-Cas9 system (Kalds et al., 2019). Expert academicians interviewed claimed that the CRISPR-Cas9 system can be used to increase resistance and protection against a variety of infectious diseases, particularly genetic diseases. In a work carried out concurrently with the results of this investigation, the NRAMP1 gene was incorporated into the FA locus and the bovine homology of the mouse Rosa26 locus using the CRISPR-Cas9 technology. As a result, it was shown that transgenic cattle had higher resistance to tuberculosis and M. bovis infection (Yuan et al., 2021). CRISPR-Cas9 technology has been claimed to have considerable potential, particularly recently, for the development of animal models and cell lines, illness treatment, pharmacological target screening, and targeted therapy (Cox et al., 2015). Recently, the CRISPR gene editing technique has been used to develop sustainable products, eradicate genetic diseases, boost productivity in agriculture and animal husbandry, make living things resistant to environmental stresses, and create individuals who may pass on desired traits to future generations (Zhang et al., 2022). Liu et al. (2013) effectively incorporated the lysostaphin encoding vector using ZFNs in bovine fetal fibroblasts into the native β-casein locus. According to Liu et al. (2013), these genetically altered cows were able to express milk that contained lysostaphin, which relieves mastitis. These gene-edited animals are only commercially available in a few countries due to various laws and restrictions (Gim et al., 2021). In the survey, all participants (100 %) said that high-yield breeds could be produced using the CRISPR approach; 19.2 % said the method was moderately effective, and 73.1 % said it was highly effective. Concurrent with the findings of this investigation, CRISPR-Cas9 has been effectively employed to enhance the meat production characteristic of diverse cattle through the elimination of the MSTN gene (Wang et al., 2015). The stearoyl-CoA desaturase gene was knocked down using CRISPR-Cas9, increasing the nutritious value of milk (Tian et al, 2018; Tian et al., 2022). Goats and sheep using CRISPR-Cas9-mediated fibroblast growth factor 5 gene deletion have better growth and hair qualities (Wang et al., 2015; Zhang et al., 2022). These research demonstrate the enormous potential of CRISPR-Cas9 to enhance contemporary livestock systems. Apart from its high efficiency and focused genome-wide editing methods, the CRISPR approach has been applied recently to control the genetic regulation and function of disease resistance genes in cattle. 42.3 % of participants in the survey said that the CRISPR method was useful in controlling and preventing infections, and 84.6 % of people said that it might be utilized to offer resistance and protection against numerous infectious diseases. In fact, research in the literature has shown that cattle with NRAMP1 knockouts caused by CRISPR/Cas9 have greater disease resistance (Xu et al., 2020; Burkard et al., 2017; Yuan et al., 2021). A Chinese team created a CRISPR-Cas9 knockout library in 2020. It contained over 85,000 single-stranded RNAs (sgRNAs) that targeted 17,743 protein-coding genes, 11,053 long non-coding RNAs, and 551 microRNAs. The work has led to significant progress in the breeding of disease-resistant animals, and it has been suggested that this breeding technology can help the livestock industry achieve sustainable development (Zhao et al., 2020). In the poultry business, CRISPR-Cas-mediated techniques are employed to get genetic traits that are unachievable through other means, resulting in the development of several genetic variations required in the chicken population (Hellmich et al., 2020). According to reports, CRISPR-Cas technology can be applied to poultry as a different approach to disease prevention (Ishii, 2016). Items derived from animals that have undergone gene editing could potentially jeopardize food safety, according to 76.9 % of survey participants. Government restrictions and public acceptance pose a greater challenge to the promotion of products derived from genetically modified animals than the technology itself (Lander et al., 2019). The introduction of genetically modified items to the consumer market in the past ten years has been met with skepticism by many practitioners. Instead of 92.3 % of the experts in this study predicted that gene-edited animal farms will be built soon, based on the study's findings. The Chinese government has started a number of initiatives to bring genetically modified crops to the public market in accordance with the study's findings (Gao et al., 2017). Predictably, given the rapid advancement of science and the ongoing growth in public awareness, it appears possible that this genetically engineered food derived from animal husbandry may soon be served on regular dinner tables. Indeed, research indicates that as gene editing technology advances, food safety and superiority will rise even higher and people will have access to better, healthier products (Ray et al., 2020; Zhou et al., 2021). The editing of a living organism's genome is one of the most talked-about CRISPR subjects (Schleidgen et al., 2020). Specifically, there is a great deal of controversy in the fields of ethics and legislation around the potential for modifying human DNA (Furtado, 2019). In fact, according to 76.9 % of the researchers surveyed, there may be ethical issues down the road as a result. Many national regulators have restricted or banned human applications due to the assumed risk associated with editing the human genome. For instance, in Germany, it is illegal to modify germline cells artificially; likewise, the National Institutes of Health in the United States has decided not to fund the use of gene editing technologies in human embryos (Locke, 2020). 38.5 % of study participants said that Turkey had the necessary infrastructure in place to use CRISPR technology. According to the literature review, there are eight research centers in Turkey focused on agricultural biotechnology, and both the R&D teams and the faculty members at universities with relevant departments have access to adequate infrastructure (Bolukbas and Gucukoglu, 2022). It has been asserted, therefore, that additional resources, investments, and unique incentives are required because the current legislative regulations are insufficient in this area (Mali, 2022). The benefits of CRISPR technology, according to this academic study, include producing high-yield breeds through animal breeding, preventing genetic diseases and cancer, boosting animal resistance to illness, speeding up and improving production efficiency, and boosting the national economy by lowering production costs. In fact, Cao (2021) stated the primary benefits of the CRISPPR-Cas9 system in parallel with the findings of this investigation, including its excellent specificity, high efficiency, straightforward design, affordable price, and capacity to edit several genes at once. Breeders now have to put in a lot of time and effort to produce plants and/or animals with the right traits. Supplying a growing population with enough food and goods is particularly difficult. It is believed that CRISPR-Cas9 genome editing (GE) using methods based on nanotechnology may be able to overcome these obstacles (Oz et al., 2021). In this case, it is believed that by expanding the CRISPR-Cas9 technology's application space, efficiency may be raised and a greater market demand can be satisfied (Naik et al., 2022). The survey study identified the following as the primary drawbacks of CRISPR technology: inadequate infrastructure, improper design, and the emergence of genetic abnormalities as a result of inexperience. These were classified as ethical issues. Parallel to the results of this study, Tavakoli et al. (2021) noted in their investigation that while there are many positive aspects of this gene technology, there are also drawbacks, such as moral dilemmas involving the disturbance of ecological balance. According to Mao et al. (2019), this approach has certain drawbacks, particularly when it comes to generating mutations with a lot of nucleotides. Although there are challenges with CRISPR-Cas9 technology, such as the insertion of foreign genes and off-target effects, these challenges are starting to be gradually addressed as a result of advancements achieved by researchers (Gasiunas et al., 2012). In light of the introduction of foreign genes via CRISPR-Cas9 technology, the study discovered that foreign genes can be eliminated by combining sgRNA and Cas9 into a ribonucleoprotein complex, which is the source, or by separating generations to produce offspring who are not transgenic (Rao et al., 2022). It is anticipated that in order to improve it, ever-more-effective optimization techniques will be used (Yang and Wu, 2018). Actually, 92.3 % of respondents to the study stated that Turkey should be the country where the CRISPR system is developed and put into use. Agriculture and animal husbandry may greatly benefit from targeted genome engineering. It is possible to either attain the desired phenotype or inactivate the undesirable trait if one knows the function of a specific gene. Therefore, CRISPR-Cas9 and nanotechnology can be used to easily design goods that can endure biotic and abiotic challenges (Jinek et al., 2012). It takes longer in traditional breeding operations to introduce new features related to quality or disease tolerance. Thus, it can be concluded that genome engineering is moving quickly in the direction of becoming an amazing technology for the production of agricultural and animal goods in the future.

## 4. Conclusion

CRISPR-Cas is no longer restricted to rupturing DNA strands, since it has led to the creation of a broad range of single-base gene editing, transcriptional regulation, and RNA strand cutting techniques after decades of research and development. Consequently, the majority of human diseases, including cancer, chronic illnesses, and hereditary disorders brought on by a single gene, can be treated using these methods. Indeed, by directly deleting disease-prone genes and pathogen receptor genes or adding disease-resistant genes, breeding using the CRISPR-Cas9 gene editing capability in cells can quickly breed new animal species with disease-resistant features, according to both the research findings and the literature review. To deliver gene editing tools to sick cells in vivo and employ them in therapeutic applications, a safe, stable, and successful technique must be developed. Additionally, off-target CRISPR-Cas9 impacts have been found to have catastrophic repercussions. In order to better ensure the safety of animal husbandry and breeding, these potential scenarios should be identified and refined prior to clinical applications. Because it is anticipated that gene editing technology will soon produce much more amazing outcomes in animal husbandry and agriculture, providing better, healthier products for all of humanity. As a result, although CRISPR technology has disadvantages as well as advantages, it is of great importance to establish legal regulations that take into account the environment, human health, food safety, international trade and ethical aspects of the CRISPR method for the development and implementation of the CRISPR system in Turkey.

## Author Contributions

The authors declare that they have contributed equally to the article. All authors declare that they have seen/read and approved the final version of the article ready for publication.

## Conflict of Interest

All authors declare that there is no conflict of interest related to this article.

## Ethical Statement

Ethical approval and permission for this study was obtained from Uşak University non-interventional clinical research ethics committee. (Date: 10.07.2024, Decision No: 410-410-08).

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