Overview of Marker-Assisted Selection in Animal Breeding

Abstract

Marker-assisted selection (MAS) in animal breeding has undergone transformative advancements, reshaping genetic improvement and reproductive traits in livestock and expanding into new domains. From its inception in the late 20th century to current developments, MAS has evolved dynamically through the integration of genomic technologies, sophisticated statistical models, and innovative gene editing techniques. These advancements have significantly enhanced the precision and impact of MAS, revolutionizing genetic improvement and reproductive performance. By synergizing MAS with cutting-edge technologies, the collective aim is to optimize reproductive performance and genetic potential, fostering a sustainable and productive future for livestock breeding and beyond. The genetic markers, typically DNA sequences or genes linked to specific traits, revolutionized animal breeding by enabling the identification of animals carrying favorable genetic variants without the need for resource-intensive phenotypic evaluations. Marker-assisted selection in reproduction also laid the foundation for genomic selection, an advanced approach utilizing genome-wide markers to estimate breeding values and predict genetic potential. This comprehensive review encapsulates the journey of MAS, emphasizing its historical significance, current advancements, and promising future trajectories, showcasing its pivotal role in shaping the landscape of animal breeding and reproductive science.

Keywords: Marker-assisted selection, Livestock Breeding, DNA markers, QTL

1. Introduction

Marker-assisted selection (MAS) has revolutionized animal breeding by significantly improving genetic traits and reproductive outcomes in livestock and beyond. This approach, which originated in the late 20th century, has evolved with the integration of genomic technologies, advanced statistical models, and innovative gene editing techniques [1]. The collective progress in MAS precision and impact has led to unprecedented advancements in genetic improvement and reproductive performance [2]. Looking forward, MAS is expected to expand into new areas such as aquaculture species and emerging animal breeding sectors [3].

The collaborative efforts of researchers, breeders, and technological innovators are propelling MAS toward a future marked by unparalleled precision and efficacy in genetic improvement. The shared objective of combining MAS with cutting-edge technologies is to optimize reproductive performance and genetic potential in animal populations, contributing to a sustainable and productive future for livestock breeding and beyond [4].

The roots of marker-assisted selection in reproduction can be traced back to the late 20th century when researchers recognized the potential of using genetic markers to expedite the breeding process and enhance trait selection in livestock populations [5]. These markers, typically DNA sequences or genes linked to specific traits, allowed for identifying animals carrying favorable genetic variants without the need for costly and time-consuming phenotypic evaluations. This innovation transformed animal breeding, enabling informed decisions by breeders and accelerating the rate of genetic gain in herds or flocks [6].

Marker-assisted selection in reproduction enables the early selection of animals with desired traits, leading to more efficient breeding programs and a reduction in the generation

interval. Moreover, it paved the way for genomic selection, an advanced approach that utilizes genome-wide markers to estimate breeding values and predict the genetic potential of animals [7, 8].

Marker-assisted selection (MAS) techniques revolutionize plant and animal breeding by leveraging quantitative trait locus (QTL) maps, which pinpoint the proximity of markers to relevant gene regions or quantitative features on chromosomes. These maps guide the application of molecular techniques, facilitating the rapid attainment of desired traits and yield in breeding programs. MAS, a cornerstone of modern breeding strategies, accelerates the development of superior varieties with precise characteristics. Single nucleotide polymorphism (SNP), the third-generation molecular marker following restriction fragment length polymorphism (RFLP) and microsatellite polymorphism (MPP), plays a pivotal role in MAS. SNPs represent DNA sequence polymorphisms resulting from single nucleotide variations in the genome, including insertions, deletions, transversions, and transitions, with allele variation frequencies exceeding 1%. These mutations can alter genetic codons, leading to diverse proteins and biological phenotypes. By integrating SNP markers into MAS, breeders enhance their ability to select for desired traits efficiently, thereby fostering the production of high-quality crops and livestock [9, 10, 11]

2. History of MAS in Reproduction

MAS in reproduction has evolved to enhance the efficiency and precision of selective breeding in animals and plants. Initially, breeders relied on phenotypic selection, visually evaluating individuals based on physical characteristics, which proved to be time-consuming and subjective [12]. However, in the past decade, the use of molecular markers has revolutionized reproductive selection, allowing scientists to identify candidate genes influencing reproductive traits and map quantitative trait loci related to traits like litter size in pigs, egg yield in chickens, and reproductive traits in minks [13]. With MAS, breeders can now select individuals based on their genetic profile, enabling more accurate and efficient breeding programs. Markers associated with desired traits inform decisions about which individuals to breed, moving beyond reliance solely on physical appearance [14].

The history of marker-assisted selection in reproduction as a crucial tool for genetic improvement dates back to conventional breeding methods, which were time-consuming and subjective [15]. The introduction of molecular markers, such as microsatellites, single nucleotide polymorphisms, and more recently, genotyping by sequencing, has allowed breeders to incorporate genetic information into selection programs, resulting in more precise and efficient breeding [16]. This advancement facilitates the selection of individuals with desired reproductive traits.

In livestock species, where improvements in reproductive traits can significantly impact profit, marker-assisted selection has proven valuable. It enables breeders to select animals with desired genetic traits related to reproduction, such as increased fertility or larger litter sizes [17]. The application of MAS in reproduction has emerged as a powerful tool for genetic improvement, facilitating informed decisions about breeding and enhancing reproductive traits and efficiency in selective breeding programs [18]. This approach also contributes to more accurate and efficient breeding programs, particularly in livestock species where improvements in reproductive traits translate to substantial gains in profit [8, 19].

The origins of marker-assisted selection in reproduction trace back to the late 20th century with the development of genetic markers and mapping techniques [20]. These markers facilitated the identification of specific genes associated with reproductive traits, allowing for a more targeted selection of individuals for breeding. MAS continues to evolve

and improve, incorporating high-throughput genotyping technologies and advanced statistical models for predicting breeding values [12].

3. DNA-based Genetic Markers

The development of DNA-based genetic markers in the late 1970s made MAS a reality because researchers could now find numerous markers distributed throughout the genetic makeup of any species of interest and utilize the markers to find relationships with desired characteristics [8]. Gene mapping and animal and plant genetics have been profoundly changed by the advent of DNA-based markers. It has become theoretically feasible to make use of all of the variation in DNA sequence found in any cross using DNA-based markers [3].

Molecular markers are not considered normal genes because they usually serve no biological purpose. Rather, they can be considered perpetual markers inside the genome [8]. They are recognizable DNA sequences inherited according to the normal rules of inheritance and can be discovered at particular sites across the genome. There are several types of molecular markers, including microsatellites, single nucleotide polymorphisms (SNPs), amplified fragment length polymorphisms (AFLPs), random amplified polymorphic DNA (RAPDs) markers, and restriction fragment length polymorphisms (RFLPs).

4. Various kinds of DNA-based markers

DNA markers in molecular genetics can be divided into three primary groups based on the kind of information they offer at a particular locus: Bi-allelic dominant: Random Amplification of Polymorphic DNA (RAPDs), and AFLPs (Amplified Fragment Length Polymorphism), Bi-allelic co-dominant: Examples include RFLPs (Restriction Fragment Length Polymorphism), SSCPs (Single Stranded Conformation Polymorphism), and Multi-allelic co-dominant: For instance, microsatellites [21, 22].

4.1.Bi-allelic dominant markers

4.1.1. RAPD

Randomly Amplified Polymorphic DNA markers, commonly known as RAPDs, make use of PCR to amplify DNA segments of any sequence between a single primer [23, 24]. The success of PCR amplification hinges on the proximity of complementary sequences to the primers. The average length of the oligonucleotide used in RAPDs is typically ten bases. RAPDs have the unique capability to amplify multiple loci simultaneously, facilitating the analysis of various markers in a single lane on an agarose gel and a single PCR reaction [8].

The RAPD marker application process involves the use of random oligonucleotide short primers, usually 8 to 15 nucleotides in length, to indiscriminately amplify DNA segments of large genomic DNA through PCR [25]. The choice of primer sequence for RAPD should ideally contain as much GC (guanine and cysteine) as possible, with a preference for 50%–80% GC content [26]. This GC content allows the primer to effectively work at the annealing temperature, facilitating DNA polymerase operation and DNA elongation [27]. Additionally, the primer should not possess a palindromic sequence [26]. Notably, the arbitrary nature of the primers used in RAPDs allows this technique to be applied directly to any species, eliminating the need for prior sequence knowledge [28].

While RAPDs are valuable for assaying loci across the entire genome, it is important to note that the presence or absence of a band with a specific molecular weight is the sole means of detecting polymorphisms. However, distinguishing between homozygous and heterozygous markers can be challenging. Various factors, including the quality of the DNA template, PCR conditions, reagents, and equipment, can contribute to the amplification or

failure of any band using RAPDs [8]. Despite their somewhat lower reliability, RAPDs find widespread use in measuring population inbreeding, assessing genetic diversity and similarity, and generating genomic maps for agricultural animals [22, 29, 30, 31, 32, 33, 34].

4.1.2. AFLP

Amplified Fragment Length Polymorphisms (AFLPs) are generated through the selective amplification of restriction segments [35]. AFLP DNA fragments, ranging in size from 80 to 500 base pairs (bps), are formed by breaking down Genomic DNA with a pair of restriction enzymes and attaching oligonucleotide adaptors to the ends of each resulting fragment [8]. Following this, oligonucleotide adapters are added to the fragments, and a subset of them is selectively amplified using PCR [28]. A small percentage of restriction fragments (between 50 and 350 bp) fall within the size range suitable for PCR amplification and visualization on polyacrylamide gels. To reduce the number of co-amplified bands in larger genomes, more selective nucleotides can be added to the primers [8].

While AFLPs and RAPDs share several advantages, AFLPs demonstrate greater reproducibility. However, compared to RAPDs, AFLPs require more technical expertise and larger equipment investment, as they involve the use of polyacrylamide gels instead of agarose. The identification of AFLP bands in manual gels can be achieved through silver stain or radioactive isotope tagging of the primers. Alternatively, AFLPs can be located using an automated DNA sequencer with fluorescently tagged primers for higher throughput [8]. The Diversity array technology (DArT) further enhances AFLP throughput on a microarray platform, where DNA fragments from a single specimen are arrayed using differential hybridization to identify polymorphisms in other samples [36, 37].

AFLP is a crucial technique for identifying genetic diversity and allows the detection of changes originating from Single Nucleotide Polymorphisms (SNPs) and insertions/deletions (indels) [22]. This versatility has made AFLP popular in various applications, including linkage mapping, Quantitative Trait Loci (QTL) analysis, genetic relationship research, and investigating genetic diversity in cDNA for gene expression profiling [38, 39, 40, 41, 42, 43, 22].

4.2.Bi-allelic co-dominant markers

4.2.1. RFLP

The Restriction Fragment Length Polymorphisms (RFLP) technique, established in 1974 following the discovery of restriction endonucleases in the 1960s [22,44], is a practical method for detecting mutations. RFLP analysis involves the use of enzymes with recognition sequences generated by mutations. This method relies on variations in DNA sequence length caused by the cleavage of genomic DNA by restriction enzymes at specific and random recognition sites, combined with Southern blotting DNA probe hybridization [28].

In RFLP, the presence or absence of fragments with different base pair lengths, created when DNA samples are cut using specific endonucleases, reveals variation in DNA sequences [45]. Nucleotide alterations in eukaryotic genomes can lead to the gain or loss of the ability to be cleaved by a particular restriction endonuclease if the alteration affects the formation or elimination of a recognition site [28]. Digestion with the appropriate restriction enzyme will result in a long fragment if a recognition site is missing, while the presence of a recognition site will yield two shorter fragments. If the recognition site is present in only one of the two parental alleles, digestion will produce two distinct electrophoretic patterns—a long fragment and two shorter fragments. This is the foundational principle of RFLP, one of the earliest methods for typing DNA polymorphisms. While RFLP mapping is a widely used and robust technology, its gel-based approach limits its feasibility for high-throughput screening.

Compared to other marker systems like RAPD and SSCP, the PCR-RFLP approach is more frequently used, less expensive, and describes only one polymorphism per probe [22]. This technique is commonly applied in defining, identifying, and diagnosing nucleic acid hybridization, describing gene polymorphisms, creating genetic linkage maps, and employing recombinant DNA technology in farm animals [21, 46, 47, 48, 49, 50].

4.2.2. SSCP

Single-strand conformation polymorphism (SSCP) serves as a method for detecting and screening mutations, offering the ability to identify mutant variations across various species. It is a theoretically straightforward, high-throughput, and uncomplicated technique that relies on the electrophoretic mobility differences of single-stranded DNAs. SSCP is proficient in accurately analyzing allelic and mutational sequence variants, as well as discovering unknown mutations [28]. This method is particularly effective in detecting single nucleotide changes, small deletions and insertions, and micro-inversions.

In PCR-based SSCP analysis, DNA fragments of the same size can be separated, making it a mutation scanning tool capable of identifying DNA polymorphisms and mutations at multiple locations within a single locus [51, 52]. The technique exploits the fact that different nucleotide sequences in denatured single strands of DNA fold into various secondary structures or conformations. Non-denaturing SSCP analysis on DNA fragments with even a single nucleotide alteration reveals detectable variations in gel electrophoretic mobility. A mutation is indicated by a slight change in the mobility of the experimental sample compared to a wild-type fragment or a normal control [53].

During electrophoresis, variations in strand fold conformation result in different rates of gel migration speed. Since DNA strand folding is highly nucleotide-specific, even a single base variation can alter the shape of folding, producing distinct conformations. The initial nucleotide sequence determines the distinct conformations of the resultant secondary structures of DNA. When using non-denaturing acrylamide gel electrophoresis, the fragment's mobility is influenced by the type of secondary structure formed [28]. To separate heteroduplex and homoduplex strands, denaturing high-performance liquid chromatography (DHPLC), an upgraded version of the SSCP technique, is utilized [54]. SSCP is estimated to exhibit approximately 80 to 90% detection of potential point mutations [22].

SSCPs are widely used to screen for mutations in agricultural animals and identify sequence variants. Under certain circumstances, mutations that do not exhibit a shift in mobility may become visible [55, 56, 57].

4.3.Multi-allelic codominant markers

One of the widely used molecular genetic techniques involves the utilization of microsatellite markers—short, tandemly repeated DNA sequences ranging from 1 to 6 base pairs, with varying copy counts at different locations and throughout the genome [5, 58, 59, 60, 61, 62, 63, 64, 65, 66]. Microsatellites offer several advantages, including co-dominance, multiallelicity, high polymorphism, and the ability for PCR assays. The development of PCR revealed that a brief sequence from any point in the single-copy section of the genome could be used to create primers for selectively amplifying that region for various applications [3], leading to the concept of the sequence-tagged site (STS) [67]. A subset of STS with short tandem repeats is referred to as a microsatellite, and another subset of STS with mRNA expression is termed an expressed sequence tag (EST) [3]. STS, in conjunction with extensive insert clone libraries, acts as a powerful tool for genome study and physical markers for genome mapping [68]. Moreover, the polymorphism of the STS sequence within a specific family serves as a genetic marker, enabling the measurement of the segregation of STS alleles [3].

Various methods exist for assaying sequence polymorphism, typically involving a single base pair change. Although electrophoretic methods like cleavase fragment length polymorphisms, denaturing gradient gel electrophoresis [69], and single-strand conformational polymorphism [51] can be employed, it is often simpler to directly sequence amplified products due to the widespread availability of DNA sequencing machines. Once the polymorphic sequence difference is known, rapid assays for progeny testing can be created, often involving variations of PCR amplification of specific alleles (PASA) [70] or PCR amplification of multiple specific alleles (PAMSA) [71]. The limited polymorphic nature of any single base change polymorphism often reduces the utility of STS (except for microsatellites) as genetic markers. However, as demonstrated by Nickerson *et al*. (1992) [72], linkage disequilibrium between closely spaced markers can result in a small number of such polymorphisms within a sequenced region producing haplotypes with significantly higher polymorphic information content. STS polymorphisms have proven particularly effective in efforts to add Type I anchor sites to genetic maps [73, 74, 75, 76, 77].

Microsatellites, comprised of small repetitions with a highly variable number, are DNA sequences with the ability to span an entire genome. These markers bring several benefits, including high polymorphism, co-dominant inheritance, and simplicity in genotyping and scoring [22]. Consequently, microsatellite markers find frequent application in research related to genetic diversity and paternity analysis [5, 46, 49, 60, 62, 78, 79, 80, 81, 82, 83, 84, 85, 86, 87, 88].

5. Modern Types of Molecular Marker

Rapid advancements in genetic technology have brought several applications to animal husbandry. The annual fraction of genetic gain is expected to increase with the identification of gene regions influencing complex quantitative features of economic significance. Over the past two decades, paternity analyses, genetic diversity assessments, and other molecular genetic research heavily relied on traditional molecular markers. Single-nucleotide polymorphisms (SNPs) have emerged as a focal point for scientific research aimed at detecting genetic changes, providing substantial resources to animal breeding programs. SNP-based genomic selection, a novel method for choosing superior breeding animals, offers significant advantages [22]. High-density maps utilizing SNPs also provide valuable genetic tools for investigating variants in quantitative traits [89, 90].

Genetic polymorphisms known as SNPs arise from single-base pair positional changes in DNA sequences across individuals of the same or different species [28]. As proposed by Lander (1996) [91], SNPs represent a sequence polymorphism caused by a single nucleotide mutation at a specific locus in the DNA sequence [92, 93]. SNPs are the most prevalent type of DNA sequence polymorphisms in organisms, occurring frequently in intergenic, non-coding, and coding sections of genomes. Their frequency varies, with one SNP per 100–300 base pairs of DNA [94].

The presence or absence of SNPs is determined by the presence of a restriction enzyme recognition site on one allele and the absence of the same recognition sequence on the other allele. Mutation events, such as transitions or transversions, and insertions or deletions caused by nucleotide substitutions, contribute to SNP variation. During enzyme digestion and electrophoretic separation in gels, variations in DNA fragments result from the presence or absence of restriction enzyme recognition sites in the DNA region. SNPs, characterized by low mutation rates, are easily amplifiable for testing [5, 21, 95, 96]. Accounting for 90% of genetic variation, SNPs are a modern genotyping method known for higher sensitivity and ease of automation [97]. Various SNP genotyping methods employ detection platforms and allelic differentiation approaches [28], with molecular methods like PCR primer extension,

intrusive cleavage involving restriction digestion, ligation-based oligonucleotide joining, and allele-specific hybridization commonly used [98].

Until recently, Sanger dideoxy-sequencing was the primary method for detecting SNPs, but it has drawbacks such as large data volumes, inefficiency in detecting SNPs in heterozygous DNA templates, time consumption, and high cost. In response to these challenges, simpler gel-based SNP variant detection techniques have emerged. However, high throughput with gel-based methods presents challenges. Current high-throughput SNP genotyping platforms, utilizing mini-sequencing, heteroduplex analysis, and allele-specific hybridization, provide solutions to overcome these challenges. Recent developments include allele-specific PCR, chip-based next-generation sequencing (NGS), and genotyping-by-sequencing (GBS), offering highly informative SNPs [28]. Consequently, these markers are now the most desirable among the various molecular markers accessible for genotyping [99]. SNP discovery involves generating sequence alignments and analyzing sequence data stored in databases.

Several SNP approaches aim for large-scale analysis simultaneously, requiring sequencing, marker-specific oligonucleotide primers, or the use of probes like Taqman or Molecular Beacon. SNPs find utility in genetic mapping of different cattle species, estimating genomic breeding values (GEBVs), traceability, paternity testing, and genetic disease research. Genomic selection, with the ability to identify millions of SNPs in different animal species with a single assay, has become feasible [100, 101, 102, 103, 104, 105, 106, 107].

6. Marker Assisted Selection

The effectiveness of DNA-based markers in guiding selection processes, known as MAS, has been extensively discussed in theory. MAS offers the advantage of making selections without the cost, risk, or stress associated with rearing and evaluating offspring for an animal [3]. Furthermore, MAS allows for the selection among related individuals that do not exhibit the relevant trait, such as males laying eggs. It can also be applied in introgression techniques to select against unwanted background features and for the trait to be introgressed [108].

However, it is expected that MAS will be most beneficial for highly heritable traits with significant influence, traits that are already considered fixed in commercial lines with close-to-ideal alleles. Additionally, challenges arise from recombination between the marker and the actual Quantitative Trait Loci (QTL), as well as mutations occurring elsewhere in the genome [109], which can complicate the efficiency of MAS. Classical selection takes these effects into account, but MAS tends to disregard them. The recombination issue is expected to be resolved with the identification of genes and even the precise polymorphic alleles responsible for QTL, along with ongoing advancements in genome mapping and QTL research.

6.1.Determining QTL and key genes affecting intricate quantitative traits

Molecular biology plays a significant role in identifying factors influencing the expression of quantitative traits. The level of detail in mapping these factors determines the various applications of the obtained information. Markers are employed to trace the inheritance of chromosomal regions in families or inbred line crosses, mapping loci with substantial impacts on quantitative traits (QTL) based on induced linkage disequilibrium in the population [8].

Fine mapping is crucial in narrowing the confidence interval of the QTL's position, serving as the initial step towards identifying polymorphisms in the genes influencing performance. This process involves additional markers and individuals sampled across the outbred population. Genetic mapping (linkage and fine mapping) is used to localize the QTL to a small region on the chromosome, followed by candidate gene or positional cloning

approaches to identify the genes within the QTL region, thereby identifying genes influencing specific traits [8].

Physiological or biochemical data may provide insights into the relationship between the quantity of marker polymorphisms inside a gene and its quantitative expression. However, this approach requires extensive data to select the gene explaining the largest effect and establishing a robust relationship. The effectiveness of MAS in increasing the rate of genetic gain depends on the continuous identification of new QTL. MAS can result in genetic improvements ranging from 10% to 20%, depending on the QTL size. The frequency of the advantageous QTL allele increases rapidly over initial generations when MAS is applied to a population [110]. Exceptional individuals are often chosen as parents for the subsequent generation in MAS. In an inbred line where individuals are fixed for alternative marker and QTL alleles, several crucial features emerge [111]. All individuals in the Fl exhibit heterozygosity for both the marker and the QTL, ensuring genetic diversity within the population. Furthermore, complete linkage disequilibrium prevails between the marker and the QTL in the Fl, indicating a strong association between the two genetic loci. This intricate interplay of genetic factors underscores the importance of understanding population dynamics and genetic relationships in shaping trait inheritance and breeding outcomes.

MAS is particularly beneficial for traits regulated by large-effect QTL where phenotypic selection is costly. Linkage disequilibrium is essential for MAS application, making it suitable for dairy cattle within the family. Information from within families serves as a crucial basis for dairy cow marker information selection techniques [112]. After fine-mapping, utilizing QTL for breeding value prediction improves the accuracy of identifying superior animals in a population with linkage disequilibrium. In crossbreeding programs, MAS can be especially helpful when incorporating desired genotypes into productive local breeds with generally higher breeding values [113].

7. The Evolution of Marker-Assisted Selection in Reproduction- Recent Trends

MAS in reproduction has brought about a revolutionary transformation in breeding programs, fundamentally altering the way breeders make decisions and expediting genetic improvement in animal populations. Throughout its history, MAS has been a crucial tool in genetic enhancement, empowering breeders to make more informed decisions about breeding individuals based on their genetic profiles [114]. This approach has ushered in more targeted and efficient breeding programs, resulting in enhanced genetic traits and overall productivity in livestock species. The evolution of MAS continues, showcasing ongoing advancements and refinements in genetic technologies. With the continuous progress of high-throughput genotyping technologies, breeders now have access to more comprehensive genomic information, enabling even more accurate predictions and selections in breeding programs [115].

Recent strides in MAS have focused on integrating genomic data with advanced statistical models, providing breeders with improved insights into and predictions of the heritability of reproductive traits. This integrated approach has significantly boosted the effectiveness of breeding programs, leading to a swifter and more targeted genetic improvement in livestock species [116]. Furthermore, MAS has expanded its application beyond traditional livestock species to include aquaculture and other animal breeding sectors, contributing to advancements in reproductive traits and productivity [115]. The incorporation of gene editing technologies and CRISPR-based methods has broadened the scope of MAS, offering the potential for precise modification and enhancement of reproductive traits in animal populations [117]. These cutting-edge developments position MAS as a pivotal tool in shaping the future of animal breeding, driving unprecedented advancements in genetic improvement and reproductive performance [118].

In summary, the current landscape of marker-assisted selection reflects a dynamic and progressive era marked by the integration of genomic technologies, advanced statistical models, and innovative gene editing techniques. These collective advancements have elevated the precision and impact of MAS in shaping the genetic potential and reproductive traits of livestock species and beyond [119].

8. Advancements in Technology and Techniques

The application of MAS in reproduction has witnessed notable enhancements through technological and methodological advancements. High-throughput genotyping platforms, such as SNP arrays and next-generation sequencing, have emerged as crucial tools, enabling the rapid and cost-effective genotyping of animals on a large scale [7]. These developments have substantially increased the number of markers that can be analyzed, providing breeders with a more comprehensive understanding of an animal's genetic makeup. Additionally, the integration of bioinformatics and statistical modeling has heightened the accuracy and predictive power of MAS in reproduction. In the current landscape, MAS continues to evolve and plays a pivotal role in livestock breeding and genetic improvement, extending its application to other domains such as plant breeding and human fertility [120].

The recognition of MAS in reproduction as a valuable tool for increasing genetic gain and expediting breeding programs in livestock populations dates back to the late 20th century [121]. Since then, it has undergone significant technological and methodological advancements, enabling more efficient identification of animals with desired traits and the development of genomic selection. This advancement has revolutionized animal breeding by reducing the reliance on cost- and time-intensive phenotypic evaluations. Genetic markers facilitate informed decisions for breeders in selecting optimal parent animals to enhance desired traits in their offspring [4]. MAS in reproduction has significantly improved the accuracy of selecting animals with specific traits, thereby accelerating genetic improvement in livestock populations [110].

9. The Future of Marker-Assisted Selection in Reproduction

The future trajectory of Marker-Assisted Selection in animal breeding holds tremendous promise, with anticipated trends and developments. MAS is poised to extend its reach beyond traditional livestock species, benefiting emerging areas like aquaculture species and unconventional animal breeding sectors [3]. This broadening scope underscores MAS's pervasive influence in driving progress across diverse domains of animal husbandry [115, 122]. The collaborative endeavors of researchers, breeders, and technological innovators are driving MAS toward a future marked by unparalleled precision and efficacy in genetic improvement. Through the synergistic integration of MAS with cutting-edge technologies, the collective aim is to enhance reproductive performance and unlock genetic potential in animal populations, ultimately fostering a sustainable and productive future for livestock breeding and beyond [123].

Continuous integration with advanced technologies, including genomic sequencing, gene editing, and data analytics, will elevate the precision and efficacy of MAS. This integration is pivotal for realizing the full potential of genetic improvement and reproductive performance in animal populations [1]. As genomics knowledge advances, MAS is expected to become more refined and customizable, enabling breeders to target specific genetic traits with greater precision for efficient and tailored breeding programs. The widespread adoption of high-throughput sequencing technologies will facilitate the analysis of a larger number of genetic markers, providing a comprehensive view of the genome and enhancing MAS accuracy [89]. Advancements in statistical modeling and computational techniques will lead to sophisticated predictive models, aiding in estimating breeding values, predicting genetic potential, and optimizing reproductive performance in animal populations [2]. The future

emphasis of MAS is likely to revolve around sustainability in animal breeding practices, with a focus on selecting traits that contribute to environmental resilience, disease resistance, and adaptability to changing agricultural landscapes.

In addressing global animal breeding challenges, increased international collaboration among researchers, breeders, and policymakers is anticipated. This collaboration will involve sharing data, best practices, and standardized methodologies to establish a unified and effective approach to MAS on a global scale. However, as MAS continues to evolve, ethical considerations regarding gene editing, cloning, and other technologies may take center stage. Addressing these concerns and maintaining transparent communication with the public will be crucial for the acceptance and adoption of MAS in animal breeding. Consequently, there is likely to be a growing emphasis on education and training in the field of MAS, building expertise among breeders, researchers, and agricultural professionals to ensure the successful implementation of MAS practices.

In essence, the trajectory of MAS in animal breeding promises to be vibrant, fueled by technological advancements, interdisciplinary partnerships, and an unwavering dedication to sustainable and effective genetic enhancement across various animal populations. The ongoing amalgamation of MAS with cutting-edge technologies and an emphasis on precision breeding is anticipated to play a substantial role in bolstering the resilience and productivity of livestock and aquaculture industries on a global scale.

10. Conclusion

In conclusion, the evolution of marker-assisted selection (MAS) in animal breeding represents a remarkable journey marked by significant advancement and innovation. Since its inception in the late 20th century, MAS has fundamentally transformed selective breeding practices, providing breeders with unprecedented insights into genetic traits and reproductive outcomes. By integrating DNA-based genetic markers, sophisticated statistical models, and cutting-edge genomic technologies, MAS has empowered breeders to make informed decisions, thereby accelerating genetic improvement in livestock populations.

Moreover, MAS has not only enriched traditional breeding programs but has also extended its reach into new frontiers such as aquaculture and emerging animal breeding sectors. This expansion underscores MAS's adaptability and its capacity to drive genetic enhancement across diverse species and industries.

Looking forward, MAS is poised to continue its trajectory of growth and innovation, buoyed by ongoing advancements in gene editing techniques like CRISPR and high-throughput genotyping technologies. These breakthroughs hold the promise of further enhancing MAS's precision and efficacy, ensuring its pivotal role in shaping the future of animal breeding and contributing to sustainable and productive agricultural practices.

In essence, MAS serves as a testament to the power of scientific progress and collaborative efforts among researchers, breeders, and technological innovators. As we embrace the potential offered by MAS, we move towards a future where genetic potential and reproductive performance in animal populations are optimized, fostering a more resilient and efficient agricultural landscape. Additionally, the application of MAS, guided by quantitative trait locus maps pinpointing marker proximity to relevant gene regions, facilitates the use of molecular techniques in plant and animal breeding, leading to the expedited development of high-quality products with desired characteristics and yield. Today, thanks to advancing

technological methods and applications, breeding studies can leverage genome-level mapping and genome-wide association studies [124, 125, 126].

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