International Journal of Advanced Research in Biological Sciences
ISSN: 2348-8069
www.ijarbs.com
DOI: 10.22192/ijarbs Coden: IJARQG(USA) Volume 4, Issue 8 - 2017

Review Article

DOI: http://dx.doi.org/10.22192/ijarbs.2017.04.08.020

Review on current application of genomic biotechnologies to improve livestock and livestock products

YohanisAbera, Tarekegn Tintagu*, Bethlehem Alemu and Nesanet Berhe
School of Veterinary Medicine, Wollo University, Dessie, Ethiopia. P.O Box 1145
*Corresponding author: *drtarekegn@yahoo.com*

Abstract

An organism's genome is the 'blueprint of life,' the complete set of instructions for making the molecules, cells and tissues that control how an animal can perform throughout its life. Every cell in the body contains the complete genome, encoded in the form of DNA. This review was conducted to give emphasis about the role or the current application of some advance genomic biotechnologies in animal breeding, disease diagnosis and prevention and control strategies. There are opportunities for using molecular genetics to identify genes that are involved in variety of traits. Revolutionary opportunities for the modification of animal performance are being created by the development of new methods (reproductive technology) for embryo manipulation and the application of molecular biology (cloning and transgenesis). Consequently, new approaches are needed to develop improved tools and strategies for prevention and control of infectious diseases in animal agriculture. Among the most effective and successful of these tools are animal vaccines using recombinant deoxyribonucleic acid (rDNA) technologies. In the recent years a profound change has occurred with the introduction of new genomic biotechnological assays for the diagnosis of infectious diseases of livestock and zoonotic pathogens. These new assays include various forms of PCR, genomic sequencing, DNA probes and DNA microarray technology, Nanotechnology, Restriction fragment length polymorphisms and Pulsed field gel electrophoresis. Generally animal genomics is of interest because of its importance to produce high quality food products economically and efficiently to furnish for the increasing supply demand gap all over the world.

Keywords: Animal breeding, Diagnosis, Genomic biotechnology, livestock and vaccine

Introduction

An organism's genome is the 'blueprint of life,' the complete set of instructions for making the molecules, cells and tissues that control how an animal can perform throughout its life. Every cell in the body contains the complete genome, encoded in the form of DNA. DNA is a long chain molecule made by combining four possible types of building blocks: organic molecules named adenine (A), cytosine (C), guanine (G) and thymine (T) and sugar-phosphate backbone (Lemieux et al., 1998).

Livestock have provided a high quality and passionately accessible protein source for human consumption since their domestication. In addition man has constantly modified the genomes of these species through a variety of selective breeding practices for trait ranging from growth, color, composition and disposition. These all selective breeding practice of animals was result from phenotypic information that have tremendous potential for unblocking the secrets and furtive hidden in the billions of bases that make up an organism's genome. Animal genomics studies are very concern with challenges, in relation to the increase worldwide

demand for high quality and healthy food, the prerequisite of sustainable economics and environments of breeding system and want to adapt to global changes (Charles et al., 2010).

Animal genomics is of interest because of its importance to produce high quality food products economically and efficiently to furnish for the increasing supply demand gap all over the world. There is evidence for a rapidly increasing demand for livestock products in developing countries as a result of high population and income growth and life style changes. Genomic biotechnologies in farm animals offer a major opportunity to address shortages in agriculture production to feed the global society at large (Koopaei and Koshkoiyeh, 2011). Animal form a distinctive genomics resource as a result of their significant phenotypic diversity and of their population structure which make them particularly furnish for positional cloning. These are the assembly of techniques used in genetic screening to identify the precise area of interest in genome. The purpose of genomic technologies is the characterization and mapping of the locus that affected these traits of interest (Fan et al., 2010).

Benefiting from the PCR techniques, the Molecular markers have now become a fashionable ways for the identification and characterization of animal species and pathogens. In the last decades a number of marker techniques were consequently developed, in particular RFLP (restriction fragment length polymorphism), AFLP (amplified fragment length polymorphism), RAPD (randomly amplified polymorphic DNA), microsatellite (simple sequence repeat), DNA probe and microarray technology and **Recombinant DNA (rDNA) Technology for Vaccine Development.** The presence or absence of markers allows the genotyping of individuals and populations. A very exhilarating and fast developing application of genetic markers is in the mapping of the various animal genomes (Fan et al., 2010).

The theoretical studies of linkage mapping, finding quantitative trait loci (QTLs) and the marker assisted selection or genotype selection have been developed in the previous decade. DNA markers can be employed to make out the specific region of chromosome where genes affecting quantitative traits are located. One approach is known as Marker assisted selection (MAS) uses information about these sections of chromosomes in livestock selection programs to recognize individuals with favorable combination of quantitative trait loci (QTL). Recently there have Been numerous progresses in whole genome sequencing, in the development of next generation sequencing technologies and high throughput genotyping platform (Archibald et al., 2010).

There is a rapid development and progression of farm animals' genomics has introduced novel technologies skilled presenting global description of biological system at the level of gene and protein expression (Bendixen et al., 2005). In the past few years, an innovative technology known as microarray has attracted fabulous interest among biomedical and biological researcher. This technique can be used for gene expression analysis ,polymorphism detection, DNA sequencing and genotyping on a genomic scale (Lemieux et al.,1998).These all methods help us to increase our knowledge about the genetic architecture of complex quantitative traits in farm animals and to estimate the distribution of the genetic variation across and within breeds and population.

The objective of this paper is to review some advance genomic biotechnology applications in animal breeding, disease diagnosis and prevention and control strategies.

Literature Review

History and Overview of Vaccine Development

The history of vaccination dates back to the 1798 studies by Edward Jenner, an English physician who used cowpox virus to immunize people against smallpox (Jenner 1798). Almost 200 years later, the comprehensive smallpox vaccination program established by the World Health Organization eventually led to the worldwide eradication of that disease. That success story is proof of the tremendous potential of vaccination and has led to the development of vaccines against almost all infectious agents affecting people and animals. The ultimate objective of vaccination is to induce an immune response that subsequently recognizes the infectious agent and fights off the disease (Kim et al., 2000).

Vaccination usually is accomplished with either weakened or attenuated live agents; with inactivated agents that no longer can cause disease; or with selected, *immunogenic* parts of the disease agent called subunit vaccines. Traditional methods of creating vaccines include using a similar agent that does not cause disease, such as Jenner's cowpox virus, or passing a pathogenic disease agent through a laboratory host system to weaken or attenuate the agent. Inactivating the disease agent with one or more chemicals also can be used to create vaccines.

In addition, extracting, purifying, and using one or more parts of the disease agent can be used to induce a protective immune response (Liao et al., 2006).

An immune response is stimulated when a foreign substance called an *antigen* is encountered by the immune system. The animal's immune system has the ability to distinguish between a foreign substance, such as the proteins in a virus or bacterium, and its own proteins. It does not matter whether the foreign proteins are from a disease agent or a vaccine against the disease agent, the immune response is similar: when the animal encounters the virus or bacteria again, the immune system recognizes it and, ideally, responds to protect the animal from the disease. Certain vaccines specifically, live vaccines can revert back to pathogenic organisms and produce disease or, in some instances, even death. The development of rDNA technologies has provided new ways of attenuating disease agents by modifying their genetic makeup, or genomes, to create safer, more efficacious vaccines (Vassilev and Donis, 2001).

The genetic material consists of nucleic acids (DNA and ribonucleic acid [RNA]) that carry and convey genetic information through their bases (adenine, cytosine, guanine, and thymine); in RNA, thymine is replaced by uracil). The bases are uniquely ordered to make up the sequence of the particular gene. Modifying or deleting the genes responsible for causing disease in an organism can be accomplished in the laboratory using rDNA technologies. Typically, rDNA technology refers to laboratory methods used to break and recombine DNA molecules from different organisms (Wesley and Lager, 2006).

Using rDNA technologies, scientists can isolate a disease agent, reduce it to its basic components, examine its genetic makeup, and modify it so that it no longer causes disease but still induces a strong immune response. Methods of extracting and purifying genes from extremely small amounts of even the tiniest organisms have become routine for many laboratories. Once extracted, the nucleic acids can be modified and the genes reinserted into the organism to produce a vaccine that is attenuated and/or capable of inducing better immunological protection (Rydell et al.,2005).

Vaccine development using rDNA technologies requires a thorough understanding of the disease agent, particularly the antigens critical for inducing protection. In addition, it is important to understand the *pathogenicity* of the disease agent and the immune response of the host, to ensure that the vaccine induces the appropriate immunological reaction. Increasingly, genetic information from both microbial genomics studies and *proteomic studies* is being used to gain a better understanding of the interactions between the disease agent and the host (Uzzau et al., 2005).

Vaccine Development Using Recombinant DNA Technology

Infectious animal diseases continue to rank foremost among the significant factors limiting efficient production in animal agriculture. In addition, infectious agents that are transmitted from animals to humans by way of food and water present an increasing threat to the safety and security of the world food supply and continue to affect human health significantly. Awareness is increasing that animal agriculture could lose the use of several important antimicrobial agents and drug classes for two reasons: (1) increased resistance among pathogens and (2) public health threats posed by the potential spread of antimicrobial-resistant *zoonotic*microbes (Belli et al., 2004).

Consequently, new approaches are needed to develop improved tools and strategies for prevention and control of infectious diseases in animal agriculture. Among the most effective and successful of these tools are animal vaccines. Using *recombinant deoxyribonucleic acid (rDNA) technologies*, these vaccines no longer cause disease, but still induce a strong immune response. Paralleling the development of new, more efficacious, stable, and safe recombinant vaccines has been the study of vaccine delivery methods and *immunostimulating adjuvant* compounds that enhance the immune response (Bergman et al.,2006).

Advances in gene discovery of animal pathogens can be expected to identify new proteins and metabolic pathways, thereby providing a foundation for improved understanding of pathogen biology and, ultimately, aiding in the design of new and effective therapies. New treatments, whether vaccines or new drugs, must rely on more than empirical methods of discovery and must be based on a fundamental knowledge of pathogen biology and genetics (Butter et al., 2003).

Categories of Vaccines

Recombinant vaccines fall into three basic categories: live genetically modified organisms, recombinant inactivated vaccines, and genetic vaccines (Ellis, 1999).

Live genetically modified vaccines

The first category live genetically modified vaccines can be viruses or bacteria with one or more genes deleted or inactivated, or they can be vaccines carrying a foreign gene from another disease agent, which are referred to as vaccine *vectors*. Deletion or gene-inactivated vaccines are developed to attenuate the disease agent. Generally two (double-knockout) or more genes are deleted or inactivated so the vaccine remains stable and cannot revert to a pathogenic agent (Uzzau et al., 2005). Developing a vaccine of this type requires knowledge of the gene(s) responsible for pathogenicity and assumes that those genes are not the same genes governing viability and the ability of the modified organism to induce an immune response. Examples of gene-deleted vaccines include a *Salmonella* vaccine for sheep and poultry and a pseudo rabies virus vaccine for pigs (Rodriguez and Whitton 2000).

Another relatively recent method of creating a live genetically modified vaccine is to use an infectious clone of the disease agent. An infectious clone is created by isolating the entire genome of the disease agent (usually viruses) in the laboratory. This isolated or cloned genome can be specifically and purposefully modified in the laboratory and then used to re-create the live genetically modified organism. Vector-based vaccines are bacteria, viruses, or plants carrying a gene from another disease agent that is expressed and then induces an immune response when the host is vaccinated. For viral and bacterial vectors, the vaccine induces a protective response against itself (the vector) as well as the other disease agent. Foreign genes must be inserted into the genome of the vaccine vector in such a way that the vaccine remains viable (Uzzau et al., 2005).

The first commercial vaccine vector was Vector Vax FP-N (Zeon Corporation, Japan), a vaccine primarily used in turkeys; it consists of a fowl pox vaccine virus that carries genes from Newcastle disease virus. Other agents used as vectors of foreign genes are *Salmonella*, herpesviruses, adenoviruses, and adeno-associated viruses. Edible plant-derived vaccines take advantage of the ability of some antigens to induce an immune response when delivered orally. Foreign genes from disease agents have been inserted into potatoes, soybeans, and corn plants and fed to animals; the expressed proteins from those foreign genes immunized the animals against the disease agent (Streatfield, 2005).

Recombinant inactivated vaccines

The second category recombinant inactivated vaccines are subunit vaccines containing only part of the whole organism. Subunit vaccines can be *synthetic peptides* that are synthesized in the laboratory and represent the most basic portion of a protein that induces an immune response. Subunit vaccines also can consist of whole proteins extracted from the disease agent or expressed from cloned genes in the laboratory. Several systems can be used to express recombinant proteins, including expression systems that are cell free or that use whole cells. Whole-cell expression systems include prokaryotic (bacteria-based) systems such as *Escherichia coli*, and eukaryotic (mammalian, avian, insect, or yeast-based) systems (Piller et al., 2005).

The baculovirus expression system is a widely used eukaryotic system because it is applicable to many different proteins and because relatively large amounts of protein can be produced. Baculovirus expression systems are engineered specifically for expression of proteins in insect cells. It is important to express proteins of disease agents with the greatest possible similarity to the authentic molecule so that the proper immune response is induced when the proteins are used as a vaccine. Baculovirus expression systems are effective for properly modifying recombinant proteins so they are antigenically, immunogenically, and functionally similar to the native protein (Kim et al., 2000).

Another type of recombinant subunit vaccines, called virus-like particles (VLPs), can be created when one or more cloned genes that represent the structural proteins of a virus are expressed simultaneously and self-assemble into VLPs. These VLPs are immunogenic (i.e., they look like the virus on the outside, but cannot replicate because they do not contain any genetic material on the inside). Because subunit vaccines do not replicate in the host, they usually are administered (injected) with an adjuvant, a substance that stimulates the immune system of the animal to respond to the vaccine (Rydell et al., 2005).

The mechanisms of action of many adjuvants are poorly understood; consequently, they usually are selected on a trial-and-error basis. Adjuvants can enhance the response to a vaccine by protecting the vaccine from rapid degradation in the animal; these are usually oil-based adjuvants. Or, they can attract so-called antigen-presenting cells, which process and deliver antigens to the immune system. Adjuvants also can be molecules that enhance the immune response by stimulating immune cells directly, or by stimulating immune-modifying and immune-strengthening or *immunopotentiating* substances called *cytokines*. In addition, adjuvants can be a combination of these types, response by stimulating immune cells directly, or by stimulating immune-modifying and immune-strengthening or *immunopotentiating* substances called *cytokines* (Nobiron et al., 2003).

Genetic vaccines

The third category of recombinant vaccines is referred to as genetic vaccines because they are actually DNA alone. Genetic or DNA vaccines usually are circular pieces of DNA, called plasmids, which contain a foreign gene from a disease agent and a promoter that is used to initiate the expression of the protein from that gene in the target animal (Rodriguez and Whitton, 2000). Plasmids can be maintained in bacteria (usually *E. coli*) and have been designed to accept foreign genes for expression in animals. The recombinant plasmids containing a foreign gene are purified from the bacteria, and the "naked" DNA is injected directly into the animal, usually intramuscularly or intradermally (into the skin). The animal's cells take up the DNA, and an immune response is induced to the protein expressed from the foreign gene (*Piller*et al., 2005).

In addition to genes coding for immunogenic proteins, genetic vaccines also have been designed to include different immune-stimulatory genes that trigger different compartments of the immune system, depending on the type of immunity desired. Unique features of DNA vaccines are intrinsic sequences embedded in the DNA, so-called *CpG motifs*. These unmethylated motifs were shown to act as an adjuvant, stimulating the innate immune responses and enhancing the effectiveness of the vaccine. The following sections describe commercially available recombinant vaccines for ruminants, swine, poultry, fish, and companion animals. In addition, each section addresses recent advances in recombinant vaccines for control of infectious agents in those animals, as well as future vaccine technologies being explored for animal health and protection (Fingerutet al.,2003).

Molecular Diagnostic and Animal Health

Infectious animal diseases are the major constraints to the efficient production of livestock and poultry, and are serious threat to future food security. Timely and efficient diagnosis is a benchmark for the control and prevention of these diseases. Efficient management of infectious diseases, in particular those of a transboundary nature, require rapid, sensitive, specific and confirmatory identification of the pathogen. Conventional diagnostic techniques such as the isolation of the pathogen (the gold standard for disease diagnosis), are not only laborious for fastidious pathogens that are difficult to cultivate in vitro, but could also pose a risk to diagnostic technicians if they are zoonotic in nature. As these limitations often require high level security laboratories (e.g. BSL3 level), the amplification of nucleic acids has been shown to have advantages (Beláket al.,2009).

Nucleic acid (DNA or RNA based) diagnostics

The use of nucleic acid-based diagnostics in veterinary medicine has increased exponentially in recent years. These techniques have redefined the level of information available for animal disease control programs. In addition, modifications of nucleic acid detection techniques such as polymerase chain reaction (PCR) have led to the development of rapid, specific assays (Johnson et al., 2003).

Polymerase chain reaction (PCR):The PCR exploits natural DNA replication mechanisms and results in the in-vitro production of large quantities of a desired sequence of DNA from a complex mixture of heterogeneous sequences (Saiki et al., 1988). Polymerase chain reaction (PCR) is a highly sensitive and reliable molecular technique to amplify a single or few copies of a piece of DNA under *in- vitro* condition. PCR can amplify a selected region of from fifty to several thousand base pairs into billions of copies. The specificity of the amplified region can be targeted by specific primers (short synthetic molecules of DNA complementary to both strands and flanking the target sequences), which are annealed to the single-stranded template and extended with the DNA polymerase (Sambrook and Russell, 2001).

The amplification of DNA in PCR protocols is accomplished using a succession of cyclic incubation steps at different temperatures. The target DNA is first heat-denatured (94-95ºC) to separate the two complementary strands to provide a single-stranded template. Specific primers are then annealed to the single-stranded template at a low temperature (50-65ºC) and extended with DNA polymerase at an intermediate temperature (72ºC). Once the polymerase has synthesized a new strand of DNA, the product is separated from the template by heating to a higher temperature. These steps, referred to as cycles, are repeated 20–40 times, resulting in amplification of target DNA sequences (Collins et al., 2003).

The key to the geometric amplification of target DNA sequences by the PCR is the selection of paired primers that, when extended, will create additional reciprocal primer-annealing sites for primer extension in subsequent cycles. To detect RNA (e.g. RNA viruses), a cDNA copy of the RNA must first be made using reverse transcriptase (RT). The cDNA then acts as the template for amplification by the PCR. This technique is referred to as reverse transcription PCR (RT-PCR) (Spackman *et al.*, 2003). The identity of the PCR product is defined by its characteristic size, and/or confirmed using DNA probes, or restriction digests, which can be used to provide restriction fragment length polymorphisms (RFLP). More commonly, since the advent of automated cycle sequencing techniques, identification can be made unequivocally via direct sequencing of the PCR product. For example, sequencing is used in the virulence typing of avian influenza A virus, in which virulence-associated structural motifs at the haemagglutinin gene cleavage site are reliable indicators of high pathogenicity in chickens (Collins et al., 2003).

The sensitivity of a PCR may be enhanced by the use of a second set of primers to amplify a sub-fragment from the PCR product of the first reaction. This technique is commonly referred to as 'nested PCR' and has been used to detect low levels of pathogens in the sample. Using this approach increases the sensitivity of the PCR and generates two amplified products for confirmation purposes. This technique has been used to detect a number of agents of veterinary interest including West Nile virus (Saiki ret al., 1988). A disadvantage of the nested PCR is the increased risk of cross-contamination due to the opening of amplification tubes to add an additional set of primers and consequently increase the risk of false-positive results (Johnson et al., 2003).

PCR is a highly sensitive procedure for detecting infectious agents in host tissues and vectors, even when only a small number of host cells are infected. PCR can target and amplify a gene sequence that has become integrated into the DNA of infected host cells. It can also target and amplify unintegrated viral gene sequences. It is clear that PCR has a role in the testing of vaccines to detect contamination. However, it does not differentiate between viable and nonviable organisms or incomplete pieces of genomic DNA, and this may complicate interpretation of results and affect the applicability of PCR in this role (Stender, 2003).

PCR may prove to be very useful in the diagnosis of chronic, persistent infections, such as bovine viral diarrhea (BVD), enzootic bovine leukosis or caprine arthritis/encephalitis virus. These diseases present serious problems in terms of diagnosis and prevention as infected animals are a constant potential source of transmission. To expand its utility in veterinary diagnostics and pathogen identification, PCR has been extensively modified over the years (Ridpat and Bolin, 1998; Weidmannet al., 2003). PCR using broadly conserved primers is designed for identification of classes of pathogens. Using PCR primers that are complementary to these conserved sequence regions, the presence in the sample of any bacteria of a desired class can be determined. It must be noted that a positive PCR result needs to be further characterized by hybridization with species-specific probes, analysis by RFLP, or by sequencing (Stender, 2003). Similarly, consensus PCR can be designed to use degenerate primers targeted at conserved sequence regions or motifs of a group of related pathogens. The targeting of degenerate primers (i.e. a mixture of similar primers with different bases in some positions) has led to the identification of many previously unrecognized viruses in various animal species (Heim et al., 2003).

On the other hand, multiplex PCR has been designed to use two or more primer pairs directed at pathogen-specific unique sequences within a single reaction for simultaneous detection of multiple pathogens. Multiplex PCR has the advantage of a high degree of sensitivity and specificity. However, there have been reports that multiplexing can reduce sensitivity compared with single reactions, because of competition. If it is important to have a very sensitive assay, this should be considered during the validation procedure (assay development and optimization) Johnson et al., 2003).

Classical PCR methods for diagnosis of pathogens, both bacterial and viral, are now widely replaced with real time PCR assays. In real-time PCR assays, intercalating dyes or a target-specific probe or primer

(labelled with fluorescent dye) are used. The measured fluorescent signal is proportional to the number of specific DNA fragments produced. Thus, during the real-time PCR, the accumulation of PCR products can be monitored in each consecutive cycle as a change in the degree of fluorescence. In other words, the assay can be used for absolute or relative quantification of the DNA or RNA content in a given sample. In contrast to conventional PCR, real-time PCR requires less manipulation, is more rapid, has a closed-tube format (decreased risk of cross contamination), is highly sensitive and specific thus retaining qualitative efficiency, and provides quantitative information. In many cases, real-time PCR assays have proved to be more sensitive than existing reference methods. The development of portable real-time PCR machines and assays raises the prospect of these techniques being used for rapid (less than 2 hours) diagnosis of disease outbreaks in the field (Collins et al., 2003).

When PCR is used for diagnosis, a great deal of care is required to avoid contamination of the samples because the exquisite sensitivity of the technique can easily lead to false-positive results. Multicenter studies have shown that positive samples are detected consistently between laboratories, but that false positives are frequently obtained with known negative samples, indicating the continuing presence of contamination problems. A new generation of robotic workstations is now available where PCR reactions may be set up with only a single tube open at any one time. This greatly reduces the risk of contamination. It is also important to control for potential 'negative' results caused by the presence of PCR inhibitors in the reaction mixture (Moore et al., 2004).

A template, independent of the target DNA, known to produce a PCR product (mimics) with specific primers can be used as a control for the PCR inhibitors, thus indicating false-negative results (Belák et al., 2009). Use of these precautions allows the PCR to become a realistic option for the diagnostician. The generation of the signal in a real-time PCR assay has been limited, until recently, to certain chemistries, such as intercalating dyes (SYBR Green and EvaGreen), hydrolysis probes, molecular beacons, primer probe energy transfer (PriProET), scorpion primers, dual hybridization probes and dye-labelled oligonucleotides ligation (Belák et al., 2009).

Alternative labelling has been developed using tags that enable high multiplexing of the assay. The mass tag PCR assay is an improvement of the real-time PCR platform, in which the primers are tagged with tags of known, but different, molecular weights. After amplification of the targeted DNA fragments, the tags are released using UV light and subsequently measured using mass spectrometry (Lipkin, 2010). This approach enables multiplexing of much larger panels of target DNA fragments (and hence multiple diseases), as the assay is not limited to the number of dyes available. Application of the mass tag PCR assay has been already proven in differential diagnosis of syndromic diseases (respiratory, hemorrhagic, enteric pathogens, meningitis/encephalitis syndrome) and detection of new clades of pathogens. A modification of this method uses matrix-assisted laser desorption-Ionization (MALDI), which directly measures the molecular weights of the PCR products and compares them, with known databases (Lipkin, 2010).

Additional improvement in the sequencing technology has been achieved by shifting from photometric to chemical detection of the PCR reaction in real time (Collins *et al.*, 2003). The technology is called ion torrent sequencing (pH-mediated sequencing, silicon sequencing or semiconductor sequencing) and is based on detection of the release of hydrogen ions, when a nucleotide is incorporated into a strand of DNA by the polymerase. Hydrogen ions will change the pH of the solution, which can be detected by an ion sensor (micro pH meter). The whole setup uses high-density array of micro-machined wells to perform the biochemical process in a massively parallel way. Fourth generation sequencing platforms, such as nanopore sequencing technologies, long read extension methods and methods based on direct video recording of nucleic bases, have already been developed as a proof of principle (Kunin et al., 2008).

Genome sequencing**:** The techniques by which DNA from a pathogen may be detected and characterized continue to improve and evolve. Presently, the ultimate discriminatory procedure is that of genome sequencing. Since 1977, the Sanger method (Sanger et al., 1977) has been the dominant approach and gold standard for DNA sequencing. Conventional DNA sequencing is based on cycle sequencing of targeted DNA fragments with labelled di-deoxy nucleotides, which have a property to stop the elongation at their place of binding. Each di-deoxy nucleotide is labelled with different dye, enabling distinction between individual di-deoxy nucleotides. As each di-deoxy nucleotide competes with the 'normal' nucleotides for their complementary binding sites, the result of such PCR amplification will be a mixture of DNA fragments of different length, each ending with a

defined di-deoxy nucleotide (identifiable by its color) ((Belák et al., 2009).

The PCR mixture is then analyzed using capillary electrophoresis, which separates the fragments by length and reads the color of each fragment. Analytical software is then used to convert the colour signals to a layout of nucleotides. Development of microarray technologies, as well as the improvements in DNA manipulation, has contributed significantly to the development of direct sequencing protocols capable of detecting unknown pathogens. This technology enables sequencing of large DNA fragments, allowing sequence comparison with sequence databases available locally or in the public domain. Sequencing of a well characterized portion of the genome is playing an important role in pathogen characterization and epidemiological studies. Sequencing the products amplified by PCR using degenerate primers targeting a gene common to the viruses in the same family has become an important diagnostic tool, especially for identification of previously unrecognized members of the family (Lipkin, 2010).

Sequencing the products in a defined region of the genome is used in epidemiological studies to evaluate the genetic similarity to other pathogens of the same species (subtype), to determine the phylogenetic properties or to determine the origin of an outbreak/infection. Additionally, by analyzing and comparison of different sequence motifs, this technology offers the possibility of predicting the tendency of pathogens to mutate into more pathogenic strains, allowing, to a certain level, tracing forward the spread of an outbreak or infection (Belák et al., 2009). Moreover, high-throughput sequencing applied in multiplex platforms is capable of generating random whole genome sequencing, giving the opportunity for simultaneous pathogen detection and comparison in different regions of the genome. The commercial launch of the first parallel pyrosequencing, 454 DNA sequence platform in 2005, introduced the new era of high-throughput genomic analysis now referred to as next-generation sequencing (NGS). This allows the sequencing of a large genome in a short time, facilitating the study of genetic material recovered directly from environmental samples, or metagenomics. These new technologies have made it possible quickly to identify an unknown pathogen (emerging pathogens) or one difficult to cultivate in vitro, or to identify a variant that is present in small quantities within a mixture (Kunin et al., 2008).

High-throughput sequencing is a significant challenge for the bioinformatics solutions needed to analyze the vast quantities of data generated, in order to answer specific biological questions including possible amplifications of high numbers of unexpected pathogens and their interactions with the host cell genome (Kunin et al., 2008). The process of differentiation of these pathogens, when performed using conventional blasting, is still time-consuming and inappropriate for routine use. Several approaches are currently under development to solve this problem, such as sample preparation (removal of the host cell eukaryotic DNA during the extraction phase), reducing the entry datasets for evaluation (submitting parts of the obtained amplicons instead of the whole genome), targeting towards a limited (reduced) panel of pathogens (e.g. animal pathogens, only viruses/bacteria, only a group of viruses/bacteria, etc.) and optimization of bioinformatics (production of specialized software platforms capable of analyzing large amounts of data using built in algorithms) (Belák et al., 2009).

Diagnosis by DNA probes and DNA microarray technology: Conventional DNA probing and microarray analysis are different but closely related processes. Fundamental to both processes is the binding (hybridization) of DNA, derived from a sample suspected of containing a pathogen (the ‘unknown’), with highly characterized DNA derived in advance from a pathogen of interest (the ‘known’ DNA).In conventional DNA probing, the unknown DNA (or RNA) the target is immobilized on a solid surface e.g. a membrane. The known DNA, made into a probe by labelling or tagging it in some way, is in the liquid phase and is applied to the target. Additionally, in conventional DNA probing, the target can be nucleic acids extracted from clinical material or cultured cells and either (a) added to membranes (a dot or slot blot) or (b), less conveniently in a diagnostic context, transferred to a membrane after gel electrophoresis (Petricoin et al., 2002).

The amount of pathogen in a clinical sample mightbe too low for detection. Consequently the nucleic acid might be amplified by PCR or RT-PCR, the PCR product being applied to a membrane. To visualize a probe bound to its target, the probe can be labelled with a radioactive nuclide or, more commonly and safely, ‘tagged’ non-radioactively. For example, biotin or psoralen - biotin may be incorporated into the probe, the bound probe then being detected by addition of streptavidin linked to an enzyme for subsequent generation of color or light

(chemiluminescence). In microarray diagnosis, it is the known DNA (large oligonucleotides or complementary DNA) that is the target, immobilized on a glass slide, and the unknown DNA, in the liquid phase that is labelled to make a probe (Moody, 2001).
In microarray probing the probe is made from the nucleic acid of the test sample. The nucleic acid is extracted from a sample and a PCR or RT-PCR performed using random oligonucleotides primers. In this way, part of all the nucleic acids in the sample both of host and pathogen origin is amplified. These PCR products, representative of every nucleic acid in the sample, are labelled with a fluorescent dye and applied to the microarray. Under optimized conditions only the DNA derived from the pathogen will bind to the DNA on the glass slide. If detection of a particular pathogen or group of related pathogens is the object then pathogen-specific oligonucleotides can be used to amplify these within the sample for probe production (Hoheisel, 2006).

A microarray is so-called because it can comprise several thousand different known DNAs, each DNA being spotted onto glass slides to form the array. Each spot is only around 10 μm in diameter. DNAs complementary to parts of selected genes of pathogens can be used to make the arrays (Beláket al., 2009). However, if large numbers of pathogens are to be investigated then it would be easier logistically to use large oligonucleotides. Microarrays for detecting pathogens can be designed for several levels of differentiation. In the case of oligonucleotides target DNAs oligonucleotides may initially be designed to be able to detect and differentiate pathogens at the genus level(Moody, 2001).

A number may be chosen, perhaps 10 or so, of oligonucleotides with a high degree of sequence conservation (consensus oligonucleotides) within a given genus, such that a probe made from a field sample containing a member of that genus would be likely to hybridize to at least some of theoligonucleotides, while not hybridizing (or hybridizing to a lesser degree) to those corresponding to related genera, e.g. to differentiate Apthovirus (foot and mouth disease virus (FMDV) isolates from Enterovirus strains in the Picornaviridae family. Other sets of oligonucleotides, placed on the same array slide, able to characterize a pathogen more specifically, e.g. to differentiate the seven types of FMDV, and potentially for even further refinement at the subtype level, could then be selected (Stender, 2003).

In conventional DNA probing the detection of a pathogen is limited by the number of probes used, whereas microarray analysis is limited only by the number of target DNAs on the array. If a microarray has 1000 different oligonucleotides, then to achieve the same resolving power by conventional probing would require 1000 probes and 1000 separate probing reactions. The great advantage of microarray analysis in searching for pathogens is that hundreds of pathogens can be looked for simultaneously when probing a single microarray slide (Walsh and Henderson, 2004).

Clearly, microarray analysis has great potential when investigating diseases of unknown etiology or diseases where more than one pathogen might be present and when subtyping is required. To enhance sensitivity in pathogen detection, microarrays can be coupled with PCR amplifications. These PCRs are usually designed to amplify one or more conserved genes, or multiple sequences, such as PCR using broadly conserved primers, consensus PCR and multiplex PCR. When a particular pathogen needs to be identified, then the use of a microarray would be less justifiable, as the production and hybridization of slides is relatively expensive. Instead, for these more simple cases, it would be more appropriate to use pathogen/subtype specific PCRs, followed by sequencing or RFLP for confirmation (Petricoin et al.,2002).

Nucleic acid sequence-based amplification (NASBA): NASBA is a promising gene amplification method. This isothermal technique is comprised of a two-step process whereby there is an initial enzymatic amplification of the nucleic acid targets followed by detection of the generated amplicons. The entire NASBA process is conducted at a single temperature, thereby eliminating the need for a thermocycler. The use of this technique has been shown to detect avian and human influenza viruses (Collins et al., 2003; Moore et al., 2004).

Nanotechnology: Nanotechnology is broadly defined as systems or devices related to the features of nanometer scale (one billionth of a meter). The small dimensions of this technology have led to the use of nanoarrays and nanochips as test platforms (Jain, 2003). One advantage of this technology is the potential to analyze a sample for an array of infectious agents on a single chip. Applications include the identification of specific strains or serotypes of disease agents or the differentiation of diseases caused by different viruses but with similar clinical signs. Another facet of nanotechnology is the use of

nanoparticles to label antibodies. The labeled antibodies can then be used in various assays to identify specific pathogens, molecules or structures. Example of nanoparticle technology includes the use of gold nanoparticles, nanobarcodes, quantum dots and nanoparticle probes (Santra et al., 2004; Zhao et al. 2004). Additional nanotechnologies include nanopores, cantilever arrays, nanosensors and resonance light scattering. Nanopores can be used to sequence strands of DNA as they pass through an electrically- charged membrane. Nanotechnology is still in the research stage but it is anticipated that nanotechnologies will eventually be applied to the diagnosis of endemic veterinary diseases in the future (Emerich et al., 2003).

Restriction fragment length polymorphisms (RFLP): This DNA- based method is used to distinguish between isolates of closely related pathogens, whether they are viruses, bacteria, fungi or parasites. The RFLP approach is based on the fact that the genomes of even closely related pathogens are defined by variation in sequence. The RFLP procedure consists of isolating the target pathogen, extracting DNA or RNA (with subsequent reverse transcription to DNA) and then digesting the nucleic acid with one of a panel of restriction enzymes (Loza-Rubio et al., 1999). The individual fragments within the digested DNA are then separated within a gel by electrophoresis and visualized by staining with ethidium bromide. Ideally each strain will reveal a unique pattern, or fingerprint. The results can be further analyzed with the help of computer software. PCR-RFLP is a modification of the basic RFLP technique whereby the polymerase chain reaction (PCR) is incorporated as a preliminary step. The PCR method is used to amplify a specific region of the genome (known variable sequence between pathogens), which then serves as the template DNA for the RFLP technique. This new combination (PCR-RFLP) offers a much greater sensitivity for the identification of pathogens and is especially useful when the pathogen occurs in small numbers or is difficult to culture (Lewin et al., 2002).

Pulsed field gel electrophoresis (PFGE): The limitation to separate very large DNA molecules by standard gel electrophoresis techniques can be overcome with this new technique, called pulsed field gel electrophoresis (PFGE). In PFGE an alternating voltage gradient is applied which facilitates the differential migration of large DNA fragments through agarose gels by constantly changing the direction of the electrical field during electrophoresis (Stender, 2003). The development of PFGE expanded the range of resolution for DNA fragments by as much as 2 orders of magnitude. PFGE has been successfully applied in subtyping of many pathogenic bacteria among other applications such as cloning of large plant DNA, construction of physical maps, genetic fingerprinting, etc. This technique is time consuming and require high-level of skill (Moore et al., 2004).

Reproductive Technology in Animal Breeding

Biotechnology has been directed primarily towards reproductive technology have been employed for genetic improvement of farm animals which is foremost concern over the years for scientist and researchers. Advances in assisted reproductive technologies (ART) like Artificial insemination, In vitro Production, Superovolution, Embryo transfer, transgenesis and cloning have become significant in livestock breeding, have been introduced to overcome reproductive problems (Vikrama and Balaji, 2002). All these technologies able to speed up genetic changes due to shorter generation interval and improving accuracy in selection program (Anonymous, 1992). The aim of reproductive technologies in animal breeding is overcome the ambiguity about the true genetic merit of breeding animals. ART is a general term which is used to achieve pregnancy by artificial means. It's aimed and application in routinely used today of reproductive technology in the treatment of infertility. Artificial Insemination (AI) and embryo transfer (ET) are probably the most well-known methods that have been adopted in developed and developing livestock production (Kahi1 and Rewe 2008).

The recent advances in biotechnology technologies in reproduction included production of transgenic animals and cloning (Smidt and Niemann, 1999). RT has prolonged effects on animal breeding in the future, as the increases the rate of reproduction and decrease the generation time (Abu etal., 2008). The most successful reproductive technologies like AI and ET necessitated applying on large extent, some emerging biotechnologies such as Multiple Ovulation and Embryo Transfer (MOET), In Vitro Fertilization (IVF) and cloning provides prevailing tool for rapidly changing the animal populations genetically. These technologies will absolutely play an imperative role in the future perspective and visions for efficient reproductive performance in livestock (Vikrama and Balaji, 2002).

Transgenic technology

Transgenic animal technology is in the practice of revolutionizing the manner we domesticate the livestock. The transgenesis means transfer of foreign gene (gene of interest) into the genome of other species in a way that it stably passed from generation to generation. It is been a potential way in accelerating and facilitating genetic improvement in livestock. The process to produced transgenic animals initiated with the purpose of producing better breed lines, which are strong, more caracass, high growth rate and increase milk production (Venkatesh, 2008).

In breeding, transgenic animals are created to improve qualitative and quantitative treats in livestock and to reduce susceptibility to diseases (Duszewska1 et al., 2010). This technology uses the transgene encoding a particular trait is clone into a vector which may be synthetic, virus or plasmid DNA, and hybrid vector is inserted into the genome of the host organism. A variety of methods have been developed to produce transgenic animals, some have had much success and others are being further researched. Gene transfer has been achieved in all the major livestock species and since the first success in 1985, more than 50 different transgenes have been inserted into farm animals. Because so many separate steps are involved, the success rates are often low usually one or two percent. This imposes an enormous cost in the case of cattle; so most work has been done in mice, pigs and sheep (Vajta and Gjerris, 2006). There are several methods have been introduced to create transgenic animals, in these the most common method is the microinjection of a transgene into pronucleus of a newly fertilized egg, the introduction of desired gene into embryonic stem cells and the transgenic somatic cell nuclear transfer (TSCNT) which is the variant of SCNT.

Pronuclear microinjection: The microinjection into pronucleus is the most common method known in microinjection of exogenous DNA into the pronucleus of a newly fertilized egg (zygote). This technique is used to produce transgenic sheep and pigs (Hammer *et al.*, 1985) and also transgenic cattle (Krimpenfort et al. 1991). Using this technique earliest successful creation of transgenic mice was reported in 1980s (Gordon et al., 1980). In this technique of transgenesis, animal sperm and egg are united by *in vitro* fertilization (IVF) (Bailey, 2010). The newly fertilized zygote before dividing has one male and one female pronucleus. Both male as well female nuclei is use for microinjection, but male nuclei which is larger is often preferred. A glass micropipette pulled have a very small diameter is used, with micropipette cell membrane is penetrated without causing any damage. In this technique many copies of genes are inserted into the donor nucleus, there is no control of the incorporation of transgenes into the genome of the host (Vajta and Gjerris, 2006). The successful microinjected zygote is transfer into the uterus of a psuedopregnant foster mother. Because all cells of any organism is derive from the zygote, if this technique gets a positive result then the transgene will be present in all cells, thus it will create a transgenic lines, so these are identified as germ-line transgenic animals (Harper, 1999).

Sperm mediated gene transfer (SMGT): Sperm mediated gene transfer (SMGT) is an alternative technique using natural ability of spermatozoa as a vector to transfer exogenous DNA into the egg at fertilization (Bacci, 2007; Lavitranoet al., 2002 and Zani, et al., 1995). This technique was first introduced in mice by Dr. Lavitrano in 1989. In this technique the sperm cells are stripped with DNA of interest, which binds to the surface of the sperm through specific protein-protein interaction. The DNA correlated with the sperm is then incorporated via protein dealings into the sperm nuclei (Zani, 1995). The sperm then act as a vector carries the genetic materials into the oocyte to incorporate the foreign DNA. There are several methods have been attempts at delivery of foreign DNA to the head of the sperm, including electroporation, liposome and plasmid delivery (Celebi, 2003). The positively charged basic linker protein monoclonal antibody is bind to negatively charged DNA through ionic interaction, which specifically binds the foreign DNA to sperm. The linker based sperm mediated gene transfer method (LBSMGT) if improve is one of the best way to create the transgenic animals (Chang et al., 2002).

DNA recombination in embryonic stem cells: Embryonic stem cells (ESC) have achieved major consideration in recent years in the field of medicine, agriculture and biomedical research due to their unique property of pluripotency. ES cells are derived from inner cell masses (ICM) of embryo at blastocysts stage. This type of embryo manipulation is used when inserting a transgene into a specific location in the genome (Bradely and Brosius, 2006). Two complementary strategies have been considered for the insertion of transgene in ES cell: homologous recombination and integrase mechanisms (Norman and MacInnes, 2002). With the introduction of homologous recombination the scientists and

researchers are able to restore gene function (knock-in animals), take out gene function (knock-out animals), inactivated, or introduce any alteration in gene of interest. Es cells are injected into blastocyst of developing embryo, which is implanted into the uterus of surrogate host At this stage it is possible to injecting cells into blastocyst to obtain chimeras (Mullins, 1996).

Animal cloning

Animal cloning has helped us rapidly incorporate improvements into livestock herds for more than two decades and has been an important tool for scientific researchers since 1950s. The first mammal cloned from an adult cell was Dolly the sheep in 1996 (Wilmut et al, 1997). The process that created Dolly is called somatic cell nuclear transfer (SCNT). Although the 1997 debut of dolly, the cloned sheep, brought animal cloning into the public consciousness. Today, there are estimated to be around 6000 farm animal clones worldwide (Plume, 2009). The cloning technology has been applied in the breeding of elite cattle (Kato et al.,1998), goat (Baguisi et al.,1999), pig (Polejaeva *et al.*, 2000), horse (Galli et al.,2003), buffalo (Shi et al.,2007), camel (Wani et al.,2010), Rabbet (Chesne *et al.*, 2002) and other pet species like dog, cat, rat, ferret, mouse (Wakayama et al.,1999; Roslin Institute online, 2003; Lee et al.,2005and Shin et al.,2002). Cloning is an asexual reproduction of genetically identical organism can be achieved by nuclear transfer (NT) or by embryo splitting (Abu et al., 2008).

While the process of cloning is through somatic cell nuclear transfer, Cells are collected from the donor animal (the animal to be cloned) (a) and cultured in vitro (b). An oocyte (egg cell) is collected and matured either in vitro (collection from dead animal and maturation in the laboratory) or in vivo (collection from live animal following super ovulation) (c). The oocyte is enucleated (removal of the nucleus containing the primary DNA sequence) (d). A donor cell is transferred into the enucleated oocyte (e). The donor cell and the oocyte are fused by application of an electrical pulse and the reconstructed embryo is activated by electrical or chemical stimulation (f). The reconstructed embryo is cultured in vitro or in vivo (g) and then transferred to a surrogate animal for gestation (h). The offspring is a clone of the donor animal (figure: 1) (Campbell et al., 2007 and Wani et al., 2010).

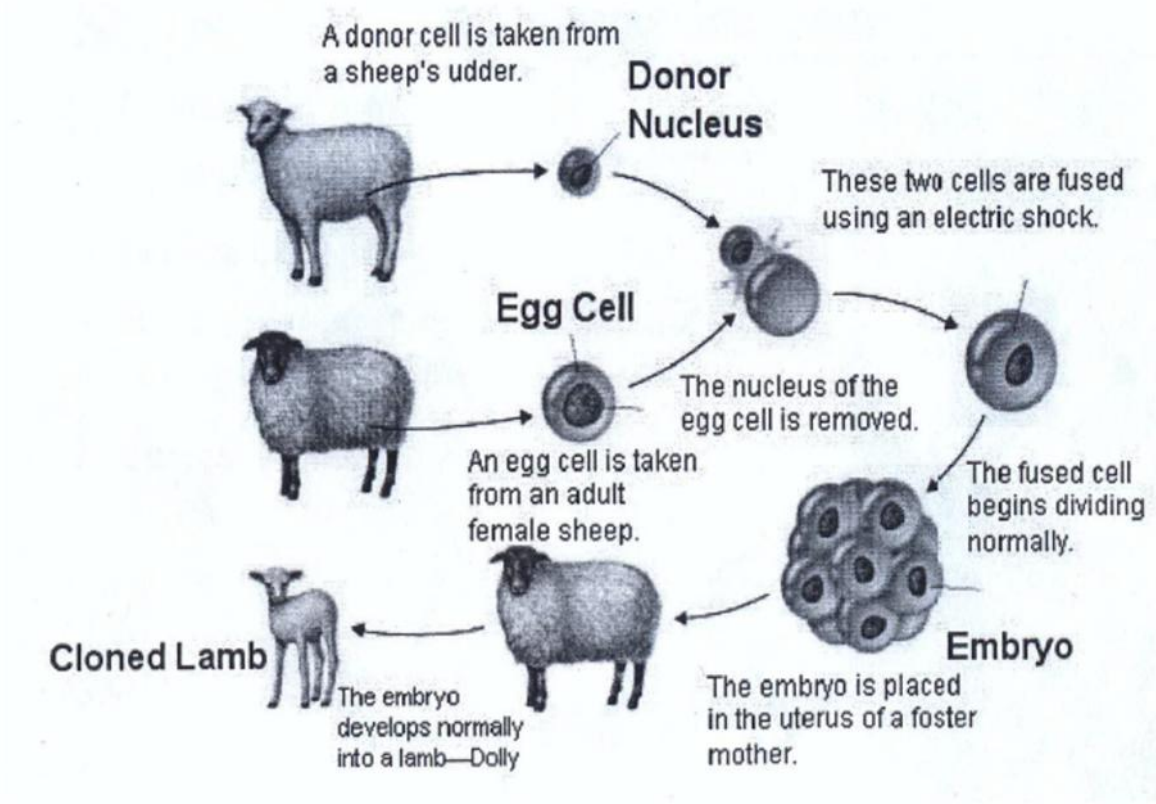

Figure 1: Animal cloning by somatic cell nuclear transfer (source: Campbell et al., 2007).

Cloning technology is already being used commercially in some parts of the world for the replication of elite breeding animals, mostly cattle, which are used to produce animals farmed for food production. Cloning can also be used in the production of genetically modified animals for biomedical, research and food production purposes (Shi et al., 2007 and Plume, 2009).The use of cloning technology is therefore facilitating the development and commercialization of genetically modified animals for food production purposes. Potential applications include: The production of animal products with altered characteristics, for example, milk with higher levels of proteins called caseins (to increase the yield of cheese that can be obtained) or lower levels of lactose or lactoglobulin (substances in milk which can cause allergic reactions in some people) (Hugo, 2006).

Genetic engineering and animal welfare

While genetically engineering farm animals to increase bone strength or reduce reception to pain, for example, can improve animal well-being, the broad use of such technology generally does not result in a reduction of suffering. Gene insertion techniques have limited success, as inserted genes may fail to properly reach target cells and may finish in cells of unintended organs. Many embryos develop abnormally and die in utero, while others may be infertile or born with developmental defects, some of which are attributable to so-called insertional problems (Li et al., 2006). Still other health issues may not become apparent until later in life. Transgenic animals often exhibit variable or uncontrolled expression of the inserted gene, resulting in illness and even death. In one study, ten transgenic piglets were followed from birth through puberty, and half of the animals died or had to be euthanized due to severe health problems during the investigation, indicating a high mortality rate among cloned piglets. In addition, three of the surviving piglets showed decreased cardiac output values (Plume, 2009).

The genetic modification of sheep containing an extra copy of a growth hormone gene resulted in animals that grew faster, leaner, and larger than those conventionally bred or produced more wool or milk for prolonged periods. Developing more economically profitable sheep resulted in negative welfare side effects from the excess growth hormone, including increased incidences of diabetes and susceptibility to parasites. While that was partially achieved, the genetically modified animals suffered from numerous problems that severely compromised their welfare, including diarrhea, mammary development in males, lethargy, arthritis, lameness, skin and eye problems, loss of libido, disruption of estrous cycles, pneumonia, pericarditis and peptic ulcers (Kakar, 2004).

Conclusion and Recommendations

Genomics is setting new paradigms in research approaches within biological sciences, and will be a major force in enhancing the rate of progress in understanding biological systems and exploiting them for development of products. Nevertheless, by breeding livestock that is more resistant to infectious disease, eliminating genetic disorders and reducing the need for interventions, genomics can not only keep animals healthy but save farmers money while ensuring that the best products are available to consumers. Diagnosis of infectious diseases of livestock and zoonotic pathogens primarily comprise of traditional diagnostic techniques. However, in the recent years a profound change has occurred with the introduction of new genomic biotechnological assays. These new assays include various forms of PCR, genomic sequencing, DNA probes and DNA microarray technology, Nanotechnology, Restriction fragment length polymorphisms and Pulsed field gel electrophoresis have become routine diagnostic tools in veterinary laboratories for specific typing as well as rapid screening of large numbers of samples during disease outbreaks and the development of rDNA technologies has provided new ways of attenuating disease agents by modifying their genetic makeup, or genomes, to create safer, more efficacious vaccines. Genetic improvement programs for livestock species can be enhanced by the use of molecular genetic information in introgression, genotype building and recurrent selection programs.

Recommendations:

- ✓ Genomic biotechnology and its applications hold great promise for improving the speed and accuracy of diagnostics for veterinary pathogens and much developmental work will be required to provide improved diagnostic capabilities to safeguard animal health.
- ✓ Awareness on the technologies to the implementing sites (Example; training for farmers) should be created.
- ✓ As all technologies are implemented to upgrade the productivity of animals, there must be a detailed selection of animals which are fit for the technology.

✓ Before application of any technologies they must be evaluated for their applicability and importance in research centers, which have a great importance for further succession of the technology.

✓ Vaccine development using rDNA technologies requires a thorough understanding of the disease agent, particularly the antigens critical for inducing protection

Acknowledgement

First and for most, I would like to express my special gratitude and thanks to almighty God for his endless mercy and existence of my life in this dynamic world. Secondly, I would like to express my deepest gratitude to my advisor, Dr. Tarekegn Tintagu, for his intellectual guidance and encouragement during manuscript writing.

References

Abu, N. M., Rahman, A., Ramli, B. and Wkhadijah, B. 2008. A review of reproductive biotechnologies and their application in Goat. Biotech.**7** (2): 371-384.

Anonymous 1992. Biotechnology. Commition, Milton Keynes.MLC Beef Year Book.Pp, 118-127.

Archibald, A. L., Bolund, C.,Churcher, M.,Fredholm, M. A., M. and Groenen, B. 2010. Pig genome sequence analysis and publication strategy. Gen.**11**:438.

Bacci, M.L. 2007. A Brief Overview of Transgenic Farm Animals. Vet. Res., Commun.**31**(1): 9–14.

Baguisi, A., Behboodi, E., Melican, D., Pollock, J., Destrempes, M., Cammuso, C., Williams, J. and Nims, S 1999. Production of goats by somatic cell nuclear transfer. Natur Biotech.**17**: 456-461.

Bailey, E. 2010. Transgenic animals. An Interactive Qualifying Project Report Submitted to the Faculty of worcester polytechnic institute. IQP- 43-DSA-8051.

Belák.S.,Thorén. p., leblanc, N. andviljoen, G. 2009. Advances in viral disease diagnostic and molecular epidemiological techniques. Exp. Rev. Mol. Diagn.**9** (4): 367–381.

Belli, S. I., Mai, M. T. Gleeson, D. Witcombe, M.,Katrib, A., Finger, M. G. and Smith, N. 2004. Characterisation of the antigenic and immunogenic properties of bacterially expressed, sexual stage antigens of the coccidian parasite, Eimeria maxima. Vac-.**22**:4316–4325.

Bergman, P. J., Camps-Palau, J. A., McKnight, N. F., Leibman, D. M., Leung, J. and Liao, I. 2006. Development of a xenogeneic DNA vaccine program for canine malignant melanoma at the Animal Medical Center. Vac.**24**:4582–4585.

Bradely, M. and Brosius, J. 2006. Transgenic animals. An interactive qualifying project, Worcester polytechnic institute. 2nded, London, Pp, 22-24.

Butter, C., T. D. Sturman, B. J. Baaten, and T. F. Davison. 2003. Protection from infectious bursal disease virus (IBDV)-induced immuno-suppression by immunization with a fowlpox recombinant containing IBDV-VP2.Av.Pathol.**32:597**–604.

Campbell, K. S., Fisher, P., Chen, W. C., Choi, I., Kelly, R. D. W., Lee, J. H., Xhu, J. and Dieleman, S. J. 2007. Somatic cell nuclear transfer: past, present and future perspectives. Therio.**68**: 214-231.

Celebri, C., Guillaudeux, T., Auvray, P.,Vallet-Erdtmann, V. and Jegou, B. 2002. The Making of Transgenic Spermatozoa. Biol.Repr.**68** (5): 1477-1483.

Chang, K. 2002. Effective generation of transgenic pigs and mice by linker based sperm-mediated gene transfer. BMC Biotech.pp, 2 (5).

Charles, H., Godfray, J.R.,Beddington, I.R.,Crute, L. and Lawrence, D. 2010. Food Security: Challen.Fed. Peop. Sci.**327**: 812.

Choe L.H., Green A., Knight R.S., Thompson E.J. and Lee K.H. 2002. Apolipoprotein E and other cerebrospinal fluid proteins differentiate ante mortem variant Creutzfeldt-Jakob disease from ante mortem sporadic Creutzfeldt- Jakob disease. Electroph.**23**, 2242–2246.

Collins R., Ko L., So, K., Ellis, T., Lau, L. and Yu, A. (2003): A NASBA method to detect high and low pathogencity H5 avian influenza viruses.Av. Dis.**47** (3): 1069- 1074.

Duszewska, A. M., Trzeciak, P., Compa, A. and Rąpała, L. 2010. Selected issues concerning biotechnology of farm animals breeding. Ani. Sci. Pap. Repo.**28**(4): 295-306.

Ellis, R. W. 1999. New technologies for making vac-cines.Vacc.**17**:1596–1604.

Emerich, D. and Thanos, C. 2003. Nanotechnology and medicine.*Exper*.*Opin.* Biol. Ther.**3** (4): 655-663.

Fingerut, E., Gutter, G. Gallili, A. and Pitcovski, J. 2003. A subunit vaccine against the adenovirus egg-drop syndrome using part of its fiber protein. Vacc.**21**:2761–2766.

Galli, C., Lagutina, I., Crotti, G., Colleoni, S., Turini, P., Ponderato, N., Duchi, R. and Lazzari, G. 2003. Pregnancy: a cloned horse born to its dam. Natur **424**: 635.

Gordon, J. W. 1980. Genetic transformation of mouse embryos by microinjection of purified DNA. Proc. Natl. Acad. Sci. USA.**77**: 7380-7384.

Hammer, R. E., Pursel, V. G., Rexroad, C., Wall R. J., Bolt, D. J., Ebert, K. M., Palmiter, R. D.andBrinster, R. L. 1985. Production of transgenic rabbits, sheep and pigs by microinjection. Natur.**315**: 680-683.

Harper, S. B. 1999. How Transgenic Are Produced. FDA Veterinarian Newsletter.Vol**14.** FDA U.S. Department of Health and Human Services.

Heim A., Ebnet C., Harste G. and Pring-Akerblom, P. 2003. Rapid and quantitative detection of human adenovirus DNA by real-time PCR. J. Med. Virol.**70**: 228-239.

Hoheisel, J. 2006. Microarray technology beyond transcript profiling and genotype analysis. Natur Rev. Gen. 7: 200-210.

Hugo, H. M. 2006. Genetic engineering applications in animal breeding. J. Biotech.**9**: 3450-3458

Jain, K. (2003): Nanodiagnostics: application of nanotechnology in molecular diagnostics.Rev.mol. Diagn.**3** (2): 153-161.

Jenner, E. 1798. An Inquiry into the Causes and Effects of the VariolaeVaccines, a Disease Discovered in Some of the Western Counties of England, Particularly Gloucestershire, and Known by the Name of the Cow-Pox. Printed for the author by Low, S. London.pp, 46.

Johnson, D.,Ostlund, E. and Schmitt, B. 2003. Nested multiplex RT-PCR for detection and differentiation of WestNile virus and eastern equine encephalomyelitis virus in braintissues. J. vet.diagn.Invest.**15** (5): 488-493.

Kahi1, A. K. and Rewe, T, O. 2008. Biotechnology in livestock production: Overview of possibilities for Africa. Afri. J. Biotech.**7** (25): 4984- 4991.

Kakar, A., Babar,M. and Bajwa, M. 2004. Reproductive Biotechnologies and Their Future in Pakistan.Int. J. Agri. Biol. **6** (2): 723–731.

Kato, Y., Tani, T., Sotomaru, K., Kurokawa, J. and Kato, H. 1998. Eight calves cloned from somatic cells of single adult. Sci. 282: 2095-2098

Kim, C. H., Johnson, M., Drennan, J., Simon, B.,Thomann, E.andLeong.J. A. 2000. DNA vaccines encoding viral glycoproteins induce nonspecific immunity and Mx protein synthesis in fish. J.Virol.**74**:7048–7054.

Koopaei, H. and Koshkoiyeh, E. 2011. Application of genomic technologies to the improvement of meat quality in farm animals. Biotech and Mole.Bio. Rev. **6** (6): 126-132.

Krimpenfort, P., Rademakers, A., Eyestone, W.,Van der schans, A., Van den broek, S.andKoiman, P. 1991. Generation of transgenic cattle using in vitro embryo production. Biotech.**9**: 844-847.

Kunin, V., Copeland A., Lapidus, A., Mavromatis, K. andHugenholtz, P. 2008. A bioinformatician's guide to metagenomics. Microbiol. Mol. Biol. Rev.**72** (4): 557–578.

Kurstak, E. 1999. Towards new vaccines and modern vaccinology: introductory remarks. Vacc. **17**, 1583-6.

Lavitrano, M., Bacci,M ., Forni, B.and Rossi G. 2002. Efficient production by sperm-mediated gene transfer of human decay accelerating factor (hDAF) transgenic pigs for xenotransplantation. J. PNAS.**99**: 14 230.

Lee, B. C., Kim, G., Jang, H. J. and Yuda, H. J. 2005. Dogs cloned from adult somatic cells. Natur.,Pp, 436-641.

Lemieux, B., Aharoni, A. and Schena, M. 1998. Overview of DNA chip technology. Mole.Bred.**4**: 277–289

Lewin, S., Schonian G., El, N., Oskam L., Bastien, P. and Presber, W. 2002. Strain typing in Leishmaniadonovani by using sequence-confirmed amplified region analysis. Int. J.Parasitol.**32**: 1267–1276.

Liao, C. M., Huang, S. L.,Hsuan, Z. W., Chen, W. C., Lee, C. I., Liu, J. R., Winton, J. and M. S. Chien, M. 2006. Immunogenicity and efficacy of three recombinant subunit *Pasteurellamultocida*toxin vaccines against progressive atrophic rhinitis in pigs. Vacc. **24**:27–35.

Lipkin, W.I. 2010. Microbe hunting. Microbiol.Molec. Biol. Rev.**74** (3): 363–377.

Loza-Rubio E., Aguilar-Setién, A.andBahloul, Ch. 1999. Discrimination Between epidemiological cycles of rabies in Mexico. Arch. Med. Res. **30**: 144–149.

McCouch, S.R., Chen, X., Panaud, S. and Huang, T. 1997. Microsatellite marker development, mapping and application in rice genetics and breeding. Plan. Mol. Biol. **35**: 89-99.

McKusick, V. A. (1997): Genomics: structural and functional studies of genomes.Genom. **45**: 244-249.

Moore C., Hibbitts, S., Owen, N., Corden, S., Harrison, G., Fox, J., Gelder, C. and Westmoreland D. (2004): Development and evaluation of a real-time nucleic acid sequence based amplification assay for rapid detection of influenza A. J. med. Virol. (**4**): 619- 628.

Mowat, N., and Reyemamu, M. (eds.) 1997 Vaccine manual: the production and quality control of veterinary vaccines for use in developing countries. FAO Animal Production and Health Series vol**35**.

Mujer V.C., Wagner M.A., Eschenbrenner, M., Horn, T., Kraycer, J.A., Redkar, R., Hagious S., Elzer, P. andDelvecchio, V.G. 2002. Global analysis of *Brucellamelitensis*proteomes. Ann. NY Acad. Sci.**969**: 97–101.

Nobiron, I., Thompson, I., Brownlie, J. and Collins,M. 2003. DNA vaccination against bovine viral diarrhoea virus induces humoral and cellular responses in cattle with evidence for protection against viral challenge. Vacc.**21**:2082–2092.

Norman, A. and MacInnes, M. 2002. Genetic engineering of embryonic stem cellsviasitedirected DNA recombination. Rev. Undergra. Resear.**1**: 29-37.

Petricoin, E. F. 2002. Medical applications of microarray technologies: a regulatory science. Natur.Genet. Supp., **32**: 474-479.

Piller, K. J., Clemente, T., Jun, S., Petty, C., Sato, S., Pascual, D.andBost, K. 2005. Expression and immunogenicity of an *Escherichia coli* K99 fimbriae subunit antigen in soybean. Plan.**222**:6–18.

Plume, K. 2009. Special Report: Welcome to the clone farm. Reuters, 13 November 2009. http://www.reuters.com/article/idUSN1278871.

Polejaeva, I. A., S. H. Chen, T. D. Vaught, R. L. Page, J. Mullins, S. Ball, Y. Dai, J. Boone, S.Walker, D. L. Ayares, A. Colman and K. H. Campbell 2000. Cloned pigs produced by nuclear transfer from adult somatic cells. Natur. **407**: 86-90.

Ridpath, J. and Bolin, S. 1998. Differentiation of types 1a, 1band 2 bovine viral diarrhea virus (BVDV) by PCR. Molec.Cell. Prob.**12**(2): 101-106

Rodriguez, F. and Whitton J. 2000. Enhancing DNA immunization. Virol. **268**:233–238.

Rout, M. P. and Field, M.C. 2001. Isolation and characterization of subnuclear compartments from *Trypanosomabrucei*. Identification of a major repetitive nuclear lamina component.J. Biol. Chem.**276**: 38261–38271.

Rydell, N., Stertman, L. and Sjoholm, I. 2005. Starch microparticles as vaccine adjuvant. Exper.Opin. Drug Deliv. **2**:807–828.

Saiki R., Gefland, D., Stoffel S., Schanf S., Higuchi, R., Horn G., Mullis, K. andErlich, H. 1988. Primer-directed enzymatic amplification of DNA with thermostable DNA polymerase. Sci.,**293**: 487–491.

Sanger, F., Nicklen S. and Coulson, A.R. 1977. DNA sequencing with chain-terminating inhibitors. Proc. Natl Acad. Sci. USA. **74** (12), 5463–5467.

Santra, S., Xu, J., Wang, K. and Tan, W. 2004. Luminescent nanoparticle probes for bioimaging. J. Nanosci. Nanotechnol. **4** (6): 590-599.

Shi, D., Lu, F., Wei, Y., Cui, K., Yang, S., Wei, J. and Liu, Q. 2007. Buffalos (Bubalusbubalis) cloned by nuclear transfer of somatic cells. Biol. Rep.**77**: 285 291.

Smidt, D. and Niemann, H. 1999. Biotechnology in genetics and reproduction. Livest. Prod. Sci.**59**: 207-221.

Smidt, D. and Niemann. H. 1999. Biotechnology in genetics and reproduction. Livest. Prod. Sci., **59**: 207-221.

Spackman, E., Senne, D., Bulaga, L., Trock, S. and Suarez, D. 2003. Development of multiplex real-time RT-PCR as adiagnostic tool for avian influenza. Avian Dis.**47** (3): 1079-1082.

Stender H. 2003. PNA FISH: An intelligent stain for rapid diagnosis of infection disease. Exp. Rev. mol. Diagn. **3** (5): 649-655.

Streatfield, S. J. 2005. Plant-based vaccines for animal health. Rev. Sci. Tech. **24**:189–199.

Uzzau, S., Marogna, G., Leori, G., Curtiss, G., Schianchi, G. and S. Rubino. 2005. Virulence attenuation and live vaccine potential of aroA, crpcdtcya, and plasmid-cured mutants of *Salmonella enterica*serovarAbortusovis in mice and sheep. Infect. Immun.**73:4302**–4308.

Vassilev, V. B., Gil, L. and Donis, R. 2001. Microparticlemediated RNA immunization against bovine viral diarrhea virus. Vacc.**19**: 2012–2019.

Venkatesh, M.N. 2008. Transgenic Animals and Current Scenario. Res. J. Biotech.**3** (1): 324_236.

Vikrama, C.P. and Balaji N. S. 2002. Use of Assisted Reproductive Technologies for Livestock.

Wakayama, T. R. Ivan, C., Anthony, Y. and Peter M. 1999. Mice cloned from embryonic stem cells. Proce.Natio. Acad. Sci.USA., **26**: 14984- 14989.

Walsh, B., and Henderson, D. 2004. Microarrays and beyond current and future genomics tools. **82**: 292-299.

Wani, N. A., Wernery, U., Hassan, F. A., Wernery, R. and Skidmore, J. A. 2010. Production of the first cloned camel by somatic cell nuclear transfer. Biology of Reproduction.**82**: 373–379.

Weidmann M., Meyer-Konig, U. and Hufert, F.T. 2003. Repid detection of herpes simplex virus and varicella-zoster virus infection by real-time PCR. J. Clin. Microbiol.**41**: 1565- 1568.

Wesley, R. D. and Lager, K. 2006. Overcoming maternal antibody interference by vaccination with human adenovirus 5 recombinant viruses expressing the hemagglutinin and the nucleo-protein of swine influenza virus. Vet Microbiol. **118**:67–75.

Wilmut, I., Schnieke, A. E., McWhir, J., Kind, A. J. and Campbell, K. H. 1997. Viable offspring derived from fetal and adult mammalian cells. Natur.**385**: 810-813.

Yatsuda, A.P., Krijgsveld, J., Cornelissen, A.W., Heck, A.J. and De Vries, E. 2003. Comprehensive analysis of the secreted proteins of the parasite *Haemonchuscontortus*reveals extensive sequence variation and differential immune recognition. J. Biol. Chem.**278**, 16941–16951.

Zani, M., Lavitrano, M., French, D., Lulli, V., Maione, B.,Sperandio, S. andSpadafora, C. 1995.*E*xp. Cell Res. **217**: 57–64.

Zhao X., Hilliard, L., Mechery, S., Wang, Y., Bagwe, R., Jin, S. and Tan, W. 2004. A rapid bioassay for single bacterial cell quantitation using bioconjugated nanoparticles. Proc. nat. Acad. Sci. USA.**101** (42): 15027-1532.

Access this Article in Online

Website: www.ijarbs.com

Subject: Biotechnology

Quick Response Code

DOI:10.22192/ijarbs.2017.04.08.020

How to cite this article:

YohanisAbera, Tarekegn Tintagu, Bethlehem Alemu and Nesanet Berhe. (2017). Review on current application of genomic biotechnologies to improve livestock and livestock products. Int. J. Adv. Res. Biol. Sci. 4(8): 152-168.

DOI: http://dx.doi.org/10.22192/ijarbs.2017.04.08.020