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Part 1

1.1 Introduction

The aim of the thesis is to collect the known most common inherited disorders in the sheep species, with special regards to the endangered breeds in Norway.

Sheep was domesticated for more than 10 000 years ago. That makes sheep one of the oldest domesticated livestock. «Since then mankind has transformed the sheep by selective breeding, but the sheep has in turn, powerfully shaped the course of human history» (Ryder, 1983). The impact the sheep industry had on humans is clearly seen by the wool industry in Europe and later in the colonial histories of Australia and New Zealand.

Today the modern sheep is most likely originating from wild sheep from the area of Turkey, Iran and Iraq. It do still exist some wild sheep breeds around in the world, for example the Bighorn in North-America and the Mouflon in West Asia and Europe. All the different sheep breeds in Europe are descending originally from the Mouflon. The changes seen from the Mouflon, to the modern sheep is mostly human made changes through genetic selection.

Sheepherders traditionally did their selection on the basis of observable physical characteristics, also called phenotypes. The first European sheep had short tail, many colors, double fleece and horns. Later came a more refined sheep breed with more wool, more meat and long tail. This long tailed modern breed came to Norway in middle of 1700s and the modern breed outperformed the old breed, which then almost disappeared. Luckily a small but sustainable herd survived near «Austvoll» in Norway. Thanks to this herd genes, there are 50 000 sheep of the Old Norwegian breeds in 2012 and the number is increasing. But because it was so few sheep left and because breeders want to keep the breed as «clean» as possible, by using genes from the same breed, inbreeding has become a problem.

1.1.1 Selection

Observational selection, also called the traditional way of selection, has today mostly been replaced by measurement and selection based on performance of an individual and its relatives. By selection, humans have made new and «better» breeds for production, more production effective breeds. The modern sheep breeds are producing much more meat and wool than the old traditional breeds. Even though there are much more economical having

effective breeds, some farmers has in Norway switched back to the old breeds like «Spælsau» and «Pelssau». The reasons that some formers has switched back into old breeds is that they are very hardy in the Norwegian climate and habitat. They are mostly kept in areas where people want to prevent the trees to grow, preserve the pasture without having to work very hard. Another reason is that the Norwegian society sees the importance of preserving the genetic diversity of the specie.

Norway is committed internationally to guard the genetic resources. FAO (Food and Agricultural Organization of the World`s United Nations) has a global overview: The State of the World`s Animal Genetic Resources for food and Agriculture. The Norwegian contribution is called: «Husdyravl og Husdyrgenetiske reserver i Norge». The global action plan for animal genetic resources will contribute to ensuring food safety for future generations.

With other words, breeders will select out from great production, fertility and disease resistance in their flocks, but it is also important to preserve the genetic diversity. Inbreeding causes increased genetic diseases and disorders because of the recessive traits these disorders have. It is important to trace the genes causing the disorders to prevent economical losses.

1.1.2 Gene mapping

Gene mapping describes the methods used to identify the locus of a gene and the distances between different genes. The essence of all genome mapping is to place a collection of molecular markers onto their respective positions on the genome, and in this way identify the different genes position.

Researchers began a genetic map by collecting samples of blood or tissue from related members that carry a prominent disease or trait and from related members that do not carry the disease or trait. Scientists then isolated DNA from the samples and closely examined it. They were looking for unique patterns in the DNA of the related members who do carry the disease that the DNA of those who do not carry the disease do not have. These unique molecular patterns in the DNA are referred to as polymorphisms, or markers.

Gene mapping has a very ongoing development with many future fields of use. It is a resource for determining the link between genotypes and phenotypes, which may help in understanding the evolutionary heritage of mammals. Gene mapping can also be used to develop marker-assisted selection methods to accelerate the genetic improvement of animals. Gene mapping

of animals began in 1913 when Sturtevant publicized a map of the Drosophila X chromosome.

The genetic maps can be extensive and very detailed or simple. They can cover the entire genome of an animal or be limited to a single chromosome, a part of the chromosome or a single sequence of nucleotides.

Genetic mapping can be used in diagnosing genetic diseases. A basic recommendation in the case a genetic disease or disorder have been diagnosed in animals, is that the affected individuals should not be breed and that carrier animals with affected descendants should not be breed again.

When there is a correct diagnosis of a disorder and the genetic nature of the defect is known, other possibilities than "not to breed" are possible, but the risk of recurrence should always be taken into consideration. To avoid autosomal recessive phenotypes, an important strategy is never to mate individuals who are known to be heterozygotes with another, such as those who have already had an affected descendent. For recessive X-linked phenotypes, daughters of affected individuals are all carriers and should not be mated, even when the males are non carriers. When there is a family history of a recessive defect and the zygosity of an individual is unknown, crossbreeding of them should be avoided.

From a gene map the identification of heterozygotes can be an economically advantageous procedure. It can for example allow people to make appropriate preventive decisions concerning the mating of such individuals. For several recessive genetic diseases, commercial DNA tests are now available, which is not too expensive. For instance, there is a test for arthrogryposis multiplex in cattle, pulmonary hypoplasia with anasarca (PHA) in Dexter cattle, polycystic kidney disease in cats, Sly disease (mucopolysaccharidosis VII) in dogs and there is a test for hereditary microphthalmia in sheep.

Another powerful tool, from the use of the gene map, is the identification and isolation of genes for important traits. With other words, the map can be used for monitoring the genetic variability, designing breeding programs and the selection of animals can be done at an early age for fattening, breeding and so on.

When making a gene map it is important to use highly variable or polymorphic markers spaced at regular intervals over the entire genome. With about 200 such markers over the

sheep genome, there is about 90% chance that any of these markers will be a gene of economic interest.

"The International Sheep Genomics Consortium" is seeking new collaborations to map disease genes. In partnership with Illumina, the sheep consortium launched the SNP50 BeadChip in early 2009. The SNP50 BeadChip has been used extensively for genome wide association studies (GWAS) and the implementation of genomic selection. It is very well suited for the fine-mapping of monogenic traits, and a number have already been successfully characterized. (See Tables 1 and 2 at the end of the text).

1.1.3 The sheep genome

All mammals are diploid organisms characterized by the presence of the complete sets of paternally and maternally inherited chromosomes in each somatic cell. The sheep genome is consisting of 54 diploid chromosomes. The identification of these chromosomes is essential for the gene mapping. The identification of the chromosomes in sheep may be difficult because the chromosomes are mainly acrocentric, in which the centromere is located quite near one end of the chromosome, making one of the chromosome arm much shorter than the other.

A normal mammalian development requires that the paternal and maternal copy of each gene is expressed correctly. Each copy of the genes has the potential to be expressed equally to any cell. However, a subset of mammalian autosomal genes has been identified where expression is restricted to one of the two parentally inherited chromosomes in a "parent of origin" specific manner. Such genes are said to be imprinted because one copy of the gene was epigenetically marked or imprinted in either the egg or the sperm. Thus, the allelic expression of an imprinted gene depends upon whether it resided in a male or female of the previous generation.

Imprinted genes on autosomal chromosomes can affect both male and female offspring, and such imprinting effects do not arise as a consequence of sex chromosome inheritance. «The definition of "Classically defined autosomal imprinting" is the consequence of the parental origin of each allele such that, paternally expressed/maternally imprinted genes are transcriptionally silenced on the maternally inherited chromosome only, while maternally expressed or paternally imprinted genes are silenced solely on the paternally inherited
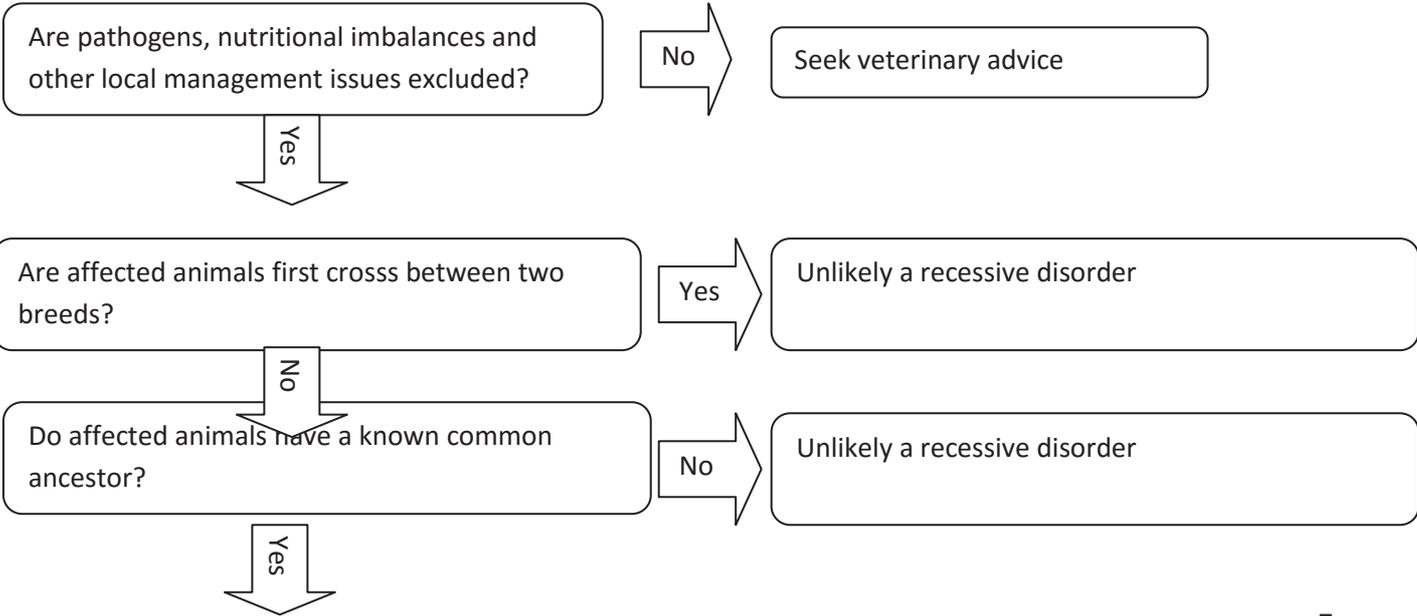
chromosome.» (Barlow and Bartolomei, 2014). Although, not all imprinted genes adhere to this classic definition. «For some genes transcriptional repression of the «imprinted» parental allele is partial, also termed «preferential» or «allele-specific» gene expression, where in one allele displays higher levels of expression relative to the other allele in a parent-of-origin manner, while other genes display tissue-and/or temporal-specific imprinting or imprinting patterns that differ between individuals of the same species.» (Giannoukakis et al., 1996; Prickett and Oakey, 2012).

Some traits, such as eye color, are fully the result of genetics, but most traits are influenced by both genetics and environment. For example when a particular breed may have a high incidence of multiple births, but if the ewes aren't fed adequately, their conception rate won't be as high despite their genetic potential. Or a breed may be known for producing really fine fleece, but if an individual animal has been sick, its fleece may be of poor quality. Since many traits are at least partially heritable, they can be taken into account when you're making breeding program decisions.

Breeding program should result in a healthy and well producing herd. Most genetic defects and diseases are recessive, which means that they can be kept away from the herd by selection..

1.1.4 Genetic recessive disorders

An indicative decision tree for deciding whether or not a disease is caused by a recessive defective allele:

```
┌─────────────────────────────────────┐
│ Are pathogens, nutritional imbalances│      No   ┌──────────────────────────┐
│ and other local management issues    │  ────▷    │ Seek veterinary advice    │
│ excluded?                             │           └──────────────────────────┘
└─────────────────────────────────────┘
              │ Yes
              ▽
┌─────────────────────────────────────┐
│ Are affected animals first crosss    │      Yes  ┌──────────────────────────┐
│ between two breeds?                   │  ────▷    │ Unlikely a recessive      │
│                                       │           │ disorder                  │
└─────────────────────────────────────┘           └──────────────────────────┘
              │ No
              ▽
┌─────────────────────────────────────┐
│ Do affected animals have a known     │      No   ┌──────────────────────────┐
│ common ancestor?                      │  ────▷    │ Unlikely a recessive      │
│                                       │           │ disorder                  │
└─────────────────────────────────────┘           └──────────────────────────┘
              │ Yes
              ▽
```

7

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┌─────────────────────────────────────┐        ┌─────────────────────────────────────┐
│ Is there a common ancestor on both  │  No    │ Unlikely a recessive disorder       │
│ sides of the pedigree?              │ ═════▷ │                                     │
└─────────────────────────────────────┘        └─────────────────────────────────────┘
              ║ Yes
              ▼
┌─────────────────────────────────────┐        ┌─────────────────────────────────────┐
│ Is it more often than chance?       │  No    │ Unlikely a recessive disorder       │
│                                     │ ═════▷ │                                     │
└─────────────────────────────────────┘        └─────────────────────────────────────┘
              ║ Yes
              ▼
┌─────────────────────────────────────┐        ┌─────────────────────────────────────┐
│ Do some sires have frequency of     │  Yes   │ Unlikely a recessive disorder       │
│ affected offspring around 50 % or   │ ═════▷ │                                     │
│ more?                               │        │                                     │
└─────────────────────────────────────┘        └─────────────────────────────────────┘
              ║ No
              ▼
┌─────────────────────────────────────┐        ┌─────────────────────────────────────┐
│ Do unaffected animals sometimes     │  Yes   │ Unlikely a recessive disorder       │
│ have both parents affected?         │ ═════▷ │                                     │
└─────────────────────────────────────┘        └─────────────────────────────────────┘
              ║ No
              ▼
┌─────────────────────────────────────┐        ┌──────────────────────────────┐
│ Has prevalence of disease recently  │  Yes   │ Have changes in prevalence   │
│ increased?                          │ ═════▷ │ and management coincided?    │
└─────────────────────────────────────┘        └──────────────────────────────┘
              ║ No                                   ║ No        ║ Yes
              ▼                                      ▼           ▼
┌─────────────────────────────────────┐        ┌──────────────────────────────┐
│ Possible Recssive                   │        │ Possible recessive with      │
│                                     │        │ impact associated with       │
│                                     │        │ environment                  │
└─────────────────────────────────────┘        └──────────────────────────────┘
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Explanation of the figure: To rule out any recessive disorder, the pathogenic or toxigenic agents being cause of disease first has to be eliminated. The nutrient balance and the local management do also have to be checked. If the disorder or disease is not caused by any of these, it is a possibility for a genetic disease.

But to be a genetic disorder it has to be common ancestors on both sides of the pedigree. The common ancestors may go way back and if this is the first cross it makes it unlikely to be a recessive genetic disease. In all the cases the same ancestors on both sides has to be common for it to be a recessive disorder. The probability for randomly chosen individuals from the same breed share a common ancestor on both sides of the pedigree has to be calculated. This

probability can also be estimated by doing an experiment. By taking repeated random samples from the breed and the see how often a common ancestor can be observed, either by the same number or fewer generations back as observed in the real cases. If the frequency of diseased lambs is over 50%, the probability of it to be a genetic disease is small.

If it is a simple recessive disease and both parents are affected by the disease, then both parents must be homozygous of the defective allele. The offspring from these parents must inherit two copies of the defective allele and will therefore be affected by disease. With other words, if it is possible for an unaffected offspring to have both parent affected by the disease, it cannot be controlled by a single recessive gene but by a dominant gene.

Based on past research, it is estimated that every time the DNA replicates in an animal, an average of around 30 copying errors occur. The vast majority of these errors will have little or no effect on the developing animals, but on the odd occasion they can have significant effects. If positive, they can help drive evolution or selection, but if negative it can result in disease. The ability to develop useful marker tests to help manage any problems that arise during replication has therefore been high on the wish list for animal breeders for a long time.

Homozygosity mapping is the mapping of homozygous genes on the chromosome. When there are homozygous genes of a defective allele it will be the cause of disease when there is a recessive disease. An animal carrying two copies of the defective allele need not imply that it will show the disease symptoms, however having the disease must imply the animal carries two copies of the defective allele. Therefore homozygosity mapping may be effective even if there are environmental factors that influence not to show any symptoms. For example, homozygotes for the defective allele controlling susceptibility to a pathogenic disease will not acquire the disease if they have not been exposed to a disease agent (For example Scrapies). Congenital abnormalities are commonly associated with recessive diseases.

Most recessive diseases result from a mutation which first arises in one founder animal which is then favored for breeding. If used extensively, and the frequency of carriers can increase rapidly, it results in the disease when two carriers are mated. If around 1% of the animals have the disease, around 11% are carriers. The carriers will maintain the disease.

Lack of openness and honesty in the occurrence of diseases is a major contributor to the spread and maintenance of the recessive disease. The breeder does not want any bad rumor

about the herd. Homozygosity mapping provides a cost effective opportunity for removing the animals having the defected genes, but it also depends on the readiness of the breeders to alert the breed-society to the possibility of a disease and the support of the breed-society in responding positively.

Preventing the diseased animals from breeding does not remove the whole problem. The key to remove a recessive disease is to also prevent the carriers from breeding. Carriers can often appear fit and healthy, therefore they are difficult to identify, which is the main problem. However, as a result of recent advances, DNA technology may offer a fairly low cost route to identify carriers of recessive diseases and therefore a way to remove them.

The risk of removing a recessive defect from a small population, by rapid selection against carriers, is that favored blood lines can be severely and unnecessarily curtailed and an overemphasis on normal sires. The normal or unaffected sires which do not have the mutated allele, may actually increase the frequency of other deleterious alleles that are likely to be present in the population. Another effective way of removing the disease is to allow only non-carrier males to breed, or to be registered in the herd book. In this way no offspring can inherit 2 defective alleles. And over time the frequency of the defective allele will diminish to a level that is sufficient to allow non-registration of females. The benefit of this method of slower approach to remove the allele is that it gives an opportunity for favored characteristics of blood lines that have high carrier frequencies to remain in the population.

Homozygosity mapping is based on the idea that for any recessive disease, the one thing that all cases have in common is that they all have two copies of the defective allele. With a large enough number of cases it would be highly significant to see complete homozygosity for the same allele in the cases, but variation in the controls. If we sequence the genome of a number of cases and a number of controls, then this homozygosity will be observed in the cases but not in the controls. Then one can go further if the disease is entirely under genetic control. In this case none of the controls would be expected to share the homozygosity seen in the cases, but they may be homozygous for a different allele. Even though there are around 3 billion nucleotide bases in the genome, with as few as 5 cases and 5 controls, it is unlikely this would occur by chance. This approach makes it possible to identify the position of the mutation with relatively little effort through the use of SNPs.

1.1.5 SNP

Whole genome SNP (single nucleotide polymorphism) marker technology is under rapid development. SNPs are the most common type of genetic variation. Each SNP represents a difference in a single DNA building block, called a nucleotide. For example, a SNP may replace the nucleotide cytosine (C) with the nucleotide thymine (T) in a certain stretch of DNA. They occur once in every 300 nucleotides on average. Most commonly, these variations are found in the DNA between the genes. They can act as biological markers, helping scientists locate genes that are associated with disease. When SNPs occur within a gene or in a regulatory region near a gene, they may play a more direct role in disease by affecting the genes` function.

Most SNPs have no effect on health or development. Some of these genetic differences, however, have proven to be very important in the study of the health of animals and humans. Researchers have found SNPs that may help predict an individuals` response to certain drugs, susceptibility to environmental factors such as toxins, and risk of developing particular diseases. SNPs can also be used to track the inheritance of disease genes within families.

The culling because of conformation faults is a routine practice in most sheep farms. An increased selection for production traits is seen, but there are still many defects present today. Faults caused by recessive genes or additive genes showing incomplete penetration, the identification and elimination of carriers becomes difficult using traditional selection methods, where the carriers do not show any symptoms. Advances in molecular genetics have enabled the development of technologies for genotyping with multiple DNA markers simultaneously. It is the said to be the future in selection of farming animals. In sheep, SNP`s are associated with economically important production traits and disease resistance. Simulation is used to predict the changes in gene frequencies of genetic merit of production traits over time. The SNPs are assumed to predict the presence of deleterious recessive genes. As selection pressure applied to individual or combination SNPs increases, the rate of increase in production trait genetic merit slowed down. Thus, a balance would be required between the emphases on SNPs actively used to select against genetic faults, relative to emphasis on genetic merit. Application of SNP technology for removal of faults to both stud breeders and commercial sheep farmers should give economic benefits.

If ovine SNP tests could be identified for defect genes, lambs carrying the undesirable SNP allele could be culled to reduce the frequency of the defect causing genes. This could improve the efficiency of sheep breeders, by increasing the number of lambs suitable for breeding or as commercial ewe flock replacements.

The same result can be achieved by using SNP chips with a high number of markers spaced evenly along the genome. It is taken advantages of the phenomenon of close linkage of some bits of DNA on the same chromosome. The structures that the DNA in our genomes is organized into tend to be inherited.

Recombination, which is the production of offspring with combinations of traits that differ from those found in either parent, will stop the process of sharing the whole chromosome. This will make the probability of inherit defective genes causing disease small. However, large segments around the defective allele are likely to be shared by carriers for numbers of generations. This will also be repeated in each generation. When an individual inherits two identical copies of the mutated allele a segment of DNA is completely homozygous. Over time, recombination events would be expected to reduce the size of the homozygous region, but in most cases this would be expected to be a relatively slow process.

The larger the segment of homozygous DNA is, the larger the target. And the more likely it can be found using a SNP chip. The length of this segment will decrease over time. But with a sufficiently dense SNP chip, the homozygosity will be observed even a number of generations after it first arises. This homozygosity will not occur in the controls. Current technology is delivering more dense SNP chips, making it possible to observe a segment of homozygosity for most recessive diseases in domestic animal species.

It is sufficient to sample only the cases at the very early stages and to gather the controls at a later date, although this minimal strategy runs the risk of particularly useful samples from dams and sires becoming unavailable and lengthening the process. It is recommended to have a biobank containing DNA samples from cases of disease as well as from healthy animals, working as controls. When an inherited disease is suspected, the biobank can help accelerate the process if homozygosity mapping is pursued. Given the recent advances in DNA technology and bioinformatics, obtaining such samples should be considered as part of the veterinary routine.

1.1.6 Homozygosity mapping

Homozygosity mapping does only work if the diagnostic test for identifying a disease has perfect specificity, in other words, no healthy animal is identified as having the disease. Homozygosity mapping will still work if the diagnostic test has less than perfect sensitivity, since it is the shared properties of the cases that are critical, and missed cases or cases treated as controls will not disrupt these properties.

One method is the early homozygosity mapping using allele frequencies at the loci within the homozygous segment. By this way the significance of a finding is assessed, making some assumptions on the independence of these sites. There are some problems with this method, the estimation of frequencies from selected samples and the incorrect assumption of mutual independence for neighboring loci.

Other problems are that chromosomes will be of different lengths and this will introduce some differences in expected shared homozygosity. The properties of the markers collected on a dense SNP chip will introduce variation. If markers are less dense on a chromosome A than on B it may be more likely for A to show a greater length of shared homozygosity as measured by cM, but on the other hand it is more likely for B to show a greater run of shared homozygosity as measured by markers.

The markers chosen in one region may have a different distribution of frequencies, or a different linkage disequilibrium (LD) structure to markers chosen in another. If a segment has markers with low minor allele frequencies it is a priori more likely to appear homozygous by chance. The issues associated with the choice of SNP markers appearing on chips will diminish as chips become denser and more refined in marker selection.

For these reasons it is advisable to compare the cases with a set of controls taken from the same gene pool, and with as much shared breeding history as possible. This eliminates, to a large degree, the impact of selection footprints and characteristics of the marker sets used to assess homozygosity lengths. The shared breeding history of the cases argues that the cases may still have longer runs of shared homozygosity than controls, and identifying the segment harboring the shared, defective allele requires the seeking of an outlier from the relationship between homozygosity runs in cases and runs in controls segment by segment.

1.1.7 Future homozygous mapping

DNA technology is predicted to get cheaper and more effective in homozygosity mapping. The technology is advancing rapidly today, and it is expected that development will continue at a very rapid pace in the future too.

The density of SNP chips will increase making the identification of homozygous segments cleaner. The information on each sequence, and hence the SNP order, will remove some of the existing pitfalls in identifying homozygous segments. The process of sequencing, whether for a segment of the genome or the whole genome will become more affordable, making the step from homozygous segment to causal mutation and a more daily routine. The annotation of sequences will become more comprehensive and bioinformatic interpretation of sequence will become more reliable, in turn making the identification of putative causal mutations more reliable.

The techniques for assembling haplotypes libraries from dense SNP chip data on a breed will improve leading to the development of libraries for each mainstream breed, at a differing rate depending on the breed. In turn, this will allow more informed assessments of the statistical significance of observing homozygous segments.

In other words, the future homozygosity mapping looks bright giving more information, the results being more reliable and the technology becoming less expensive. Today some disorders in sheep have been identified by their defective genes. According to Rigene technical manual from 2008, these disorders have been identified:

- Microphthalmia in Texel.
- Dwarfism /Achondroplasia in both Cheviot and Texel sheep.
- Junctional epidermolysis in German Black Headed sheep.
- The presence or absence of horns (poll).
- Yellow muscling found in Perendale.
- Wobbler, initial work has suggested that the inheritance pattern is more complex than that expected for a recessive inherited disease. Therefore further cases are being examined.

- Hydrocephaly, available samples provided insufficient. DNA quantity for the technique to be applied, therefore further samples are required. In the future more genetic diseases will be mapped.

Part 2

2.1 Genetic disorders

In humans, genetic factors are responsible for most dysmorphisms with a known cause (Kumar & Burton, 2008). Considering that mammals in general share most of their genes with humans and that the genes responsible for morphogenesis are highly conserved, the genetic factors must be equally responsible for most of the congenital defects of domestic mammals.

For a congenital defect to occur a mutation has happened. There are differently linked mutations:

1) **Dominant autosomal mutations**. For example the fibrillin-1 gene (FBN 1), which cause the Marfan syndrome in cattle. If the mutation of the genes is found in one of the two copies of the chromosomes it will be expressed as a disorder.

2) **Recessive autosomal mutations**, such as the one that causes anotia, cleft palate and bifid tongue in St. Bernard dogs. To get an expression of a disorder the mutation of the genes must be found on both copies of the chromosomes.

3) **Recessive X-linked mutations**. Like the one that occurs in the ectodysplasin A1 and A2 gene isoforms (EDAA1 and EDA-A2) and causes X-linked hypohidrotic ectodermal dysplasia in dogs.

4) **Deletions of Y chromosome genes**, such as the one of the sex-determining region Y gene (SRY), which determines sex reversal in horses. It leaves individuals with a male XY karyotype with a female phenotype and ovaries.

5) **Chromosomal aberrations**, such as trysomy of chromosome 30 in horses, which makes them smaller than normal, with a serious angular deviation of the thoracic limbs and polydactyly.

6) **Structural chromosomal aberrations**, such as the translocation between chromosomes.

2.2 Genetic disorders in sheep

In Norway there is no control of the genetic disorders in sheep, meaning there is no overview over how often, where and when the disorders occurs. Although, the disorders do exist, this is stated by the veterinarians, farmers and in Norwegian literature. When found in an animal, often early in the animals life, the animal are sent directly to the slaughterhouse or killed. There is no documentation how often, where and which animals may be carriers. But out from what experienced farmers and veterinarians have seen, there are some disorders being much more common than others. For example inverted eyelids, hernia and antresia ani.

I have chosen to write about some of the common disorders found in sheep (listed under). Of some of the disorders there is a lot of information about, others not so much. I have collected general information and information about the mutations or genetics of the disorders.

- Spider Lamb Syndrome (SLS): Also known as ovine hereditary chondrodysplasia, SLS is a genetic disorder causing skeletal deformities in young lambs, including abnormally long, bent limbs; twisted spines; shallow bodies; flattened rib cages; and long necks. Most lambs die shortly after birth.
- Arthrogryposis
- Cryptorchidism: One or both testicles do not drop down into the scrotum. (associated with polled trait in Merino and Rambouillets).
- Inverted eyelids, also called entropion. This trait is highly heritable and widespread among breeds of sheep. The eyelids turn in, bringing the eyelashes into direct contact with the cornea. This contact irritates the eye, making it necessary for the animal to blink constantly, which simply aggravates the problem. It can be treated if caught early.
- Brachygnathia
- Cleft palate
- umbilical/abdominal and scrotal/inguinal hernias
- Antresia ani
- Microtia
- rectal prolapse
- microphtamia

2.2.1 Hereditary chondrodysplasia (spider lambs)

2.2.1.1 General information of hereditary chondrodysplasia

Hereditary chondrodysplasia, also called Arachomelia, is the deformations in the skeletal system which is found especially in the limbs. It was first described in young lambs during early 1970s in the US.

2.2.1.2 Clinical signs of hereditary chondrodysplasia

Clinical signs seen are disproportionately long legs (spider like legs), curvature of the spine, deformed ribs and sterna, facial deformities, lack of body fat and muscular atrophy. The forelimbs are bended outward from the carpal joints. Typically seen is a crocked spine, a roman nose, extreme height, fineness of the bone, poor muscling and fail to thrive. The vertebrae are often seen with scoliosis or mild kyphosis and the sternum is found to be concave. In the face angular deformities and brachygnatia superior may be observed.

The abnormal asymmetrical growth and the abnormal tendon positions can be seen from birth or they can occur when the animal is 3-4 weeks or 3-8 weeks old. Although all the clinical signs that can be seen, the major economic impact on lamb production efficiency is less viable lambs per ewe.

2.2.1.3 Diagnosis of hereditary chondrodysplasia

Chondrodysplasia can be diagnosed by x-ray evaluation. It is especially seen in the shoulders, elbow (olecranon) and in the sternum. In these areas multiple islands of ossification which is irregular in size and symmetry can be seen. Some growth plates may be missing.

The genes causing spider leg syndrome is mapped to the distal end of the ovine chromosomes. Here the FGFR3 (fibroblast growth factor receptor 3) is found as a positional candidate for the disorder. Homozygous spider leg syndrome allele is causing bone deformities (skeleton overgrowth), while heterozygous spider syndrome allele is causing normal, though perhaps through relaxed inhibition of chondrocyte proliferation at growth plate, are physically larger than normal lambs. This is causing greater bodyweight. Carrying a single copy of a defective gene can sometimes result in a breeding advantage or a more desirable phenotype. This can result in a rapid increase in the number of carriers within a breed and therefore in the incidence of disease.

Progeny testing of potential breeding rams will decrease the frequency, but the method is very time consuming. It is much more effective to locate the gene by mapping.

2.2.1.4 Treatment of hereditary chondrodysplasia

It is mostly found in Suffolk and Hampshire sheep breeds, but it can also be found in other breeds, especially when crossbreed. A way to treat or remove the disease is to identify the affected animals and the animals carrying the mutation for the disease. When identified the animals carrying the mutation, these animals should not be breed or they can be removed from herd. The disease is semi-lethal with autosomal recessive trait.

2.2.2 Arthrogryphosis

2.2.2.1 General information of arthrogryphosis

Arthrogryposis was first described in New Zealand in 1957. It literally means joint pie, that is defined as stiffness or limited movements of multiple joints and change of posture and limb function due to permanent contracture of joints at birth (Doherty et al., 2000; Radostits et al., 2007).

Main etiologies are hereditary or genetic causes, although ingestion of toxic plants such as lupines and some viral infections during the early stages of gestation have also been suggested as the causative agents (Nawrot et al., 1980). Congenital malformations are structural and functional abnormalities present at birth. They can affect a single structure or function, parts of various systems, or an entire system (Noden and De Lahunta, 1985).

2.2.2.2 Clinical signs of arthrogryphosis

Arthrogryphosis is characterised by bilateral flexion rigidity of the metacarpophalangeal and carpal joints. The extent of this malformation is variable and may affect only one, two, or four legs and the axial skeleton (Van Vleet, 2007). Arthrogryposis usually affects the fore- and hind limbs and the distal joints (Belli, 2007).

Arthrogryposis may be associated with the other deformations such as palatoschisis or cleft palate, brachygnatia, scoliosis, lordosis, kyphosis, hydranencephaly and torticollis (Kacar et al., 2008).

2.2.3 Cryptorchidism

2.2.3.1 General information of cryptorchidism

Rams with one or both testicles retained in the abdomen, or not descended fully into the scrotum are cryptorchids. Cryptorchidism presents itself in one of two forms:

1. Unilateral cryptorchidism - normal descent of only one testicle
2. Bilateral cryptorchidism - retention of both testicles.

Unilateral cryptorchid lambs are usually capable of breeding, whereas bilateral cryptorchids often are sterile. The condition is usually inherited as a simple recessive trait. There seems to be some association between this condition and the polled characteristic found in some fine-wool rams. Purebred breeders should make every effort to eliminate this condition by not breeding affected individuals. In spite of the fact that bilateral cryptorchid lambs are sterile, both bilateral and unilateral cryptorchids should be castrated, to reduce the risk of possible future complications. Unilateral cryptorchids should never be used in a breeding program**.**

2.2.3.2 Genetics of cryptorchidism

Cryptorchidism is a common anomaly of the male sexual differentiation. There are two different forms or phases of cryptorchidism depending on the anatomic position:

1. Transabdominal
2. Inguinoscrotal

Hormonal factors involved in the process of sexual differentiation and being responsible for normal descent testicles are androgens, müllerian inhibitory hormone, which is a hormone mediating testicular descent called descending and Insulin-like 3 (INSL3) also known as relaxin-like factor (RLF) and leydig insulin-like protein (LEX 1-L) expressed in the Leydig cells. The process of testicular descent involves multiple genes, some with redundant function, to ensure proper testicular descent and fertility.

An experiment from the article "Insulin-like 3/Relaxin-Like Factor Gene Mutations Are Associated with Cryptorchidism" from 2003, describes how the INSL3 is important in normal development of the male sexual differentiation.

The phenotype of transgenic mice with targeted deletion of the INSL3 gene was bilateral cryptorchidism with morphological evidence of abnormal gubernacular development. With this implicit gives evidence that INSL3 mediates testicular descent in mice. Normal external genitalia was seen, and also a normal androgen-dependent behavior.

It was done mutation detection analysis of the coding regions of the 2 exon INSL3 genes in genomic DNA samples. Two mutations, R49X and P69L, and several polymorphisms were identified. The frequency of INSL3/RLF gene mutations as a cause of cryptorchidism was found to be low.

Morphological evaluation of the homozygous knockout males revealed developmental abnormalities of the gubernaculum. Development of the Wolffian duct structures was normal, and Müllerian duct structures were absent, indicating appropriate testosterone and MIH secretion. Thus, these findings excluded the possibility that cryptorchidism in the INSL3 knockout mice was secondary to androgen deficiency or the loss of the MIH-mediated activity.

The phenotype of heterozygous male mice differed according to the age at which the animals were examined. At birth, approximately 75% of transgenic mice heterozygous for INSL3 deletions had partial unilateral or bilateral undescended testes. But, all adult heterozygotes showed full testicular descent. With other words, spontaneous resolution of cryptorchidism can often occur with age.

2.2.3.3 Treatment of cryptorchidism

Consequences of cryptorchidism include the need for surgical intervention. It does also increase the risk to develop oligo- or azoospermia and testicular tumors. These conditions may influence the reproductive competence, and therefore is important economically.

2.2.4 Entropion

2.2.4.1 General information of entropion

Entropion is the inward turning of the eyelids. Generally, the most common location of the condition is the lower eyelid. The condition can be found in one eye, called unilateral entropion or in both eyes, called bilateral entropion.

The development of entropion can be due to different causes:

- Congenital
- Selective breeding
- Scar tissue formation
- Age related processes, due to large differences in size of tarsal plates or muscular hypertrophy. Tarsal plates are the dense fibrous connective tissue giving support and shape to the eye.

Entropion is reported frequent worldwide, everything from 1,1% to 80% in a herd. The heritability is 0,08-0,21 in Columbia, Polpay, Ramboullet, Suffolk and Targhee breeds. However entropion can be found in all breeds and in both rams and ewes.

2.2.4.2 Clinical signs of entropion

Contact between eyelashes and cornea may cause secondary inflammation and corneal abrasions. Common clinical signs can be recognized by lacrimation and soiling of the hair close to the eyes. The condition is non-lethal, but if not corrected by suitable treatment it may cause blindness. So economically, treatment is important.

2.2.4.3 Treatment of entropion

There are different possible treatments available. One way of treatment is surgical. The condition is corrected by removing an elliptical portion of the skin under the lower lid. In this way the ensuing scar formation draws the lower lid into proper placement. The surgical way of correction is mostly used when there is an advanced case. The second way of correction or treatment is seen as more esthetic. This is made by injecting 1 ml of a long-acting, slowly absorbed antibiotic under the skin of the lower lid and in this way correcting the skin position. Stainless steel surgery clips or staples can also be used to draw the skin up of the lower lid

and correct the inversion. These procedures are most effective early in the life of lambs, because the skin is softer and more pliable.

The above mentioned treatment possibilities of entropion is effective for the individual animal having the condition. But since it can be a heritable condition, the most effective way of correcting and controlling the disorder is by selection. The heritability can be reduced by culling the parents and by not breeding affected sheep. A more efficient way is by using genetic tests. It can for example be useful in replacement sheep selection. By mapping the total genome of the sheep, the genes causing entropion can most likely be found in the future.

2.2.4.4 Genetics of entropion

Today the exact genes or genes regions in mammalian species causing entropion is not known. Ovine SNP50 beadchip is used to identify new markers associated with inherited diseases, erythrocyte traits, parasite infection and other infectious diseases.

Instead of exact genes or gene regions, genes coding for different matters in the pathway of development of the eyelid is found. These genes have been identified by using multiple phenotypes. Eyelid development in embryonic mammals has 4 primary stages:

1. Specification
2. Growth
3. Epithelial fusion
4. Reopening

The developmental stages are grouped into two major pathways, called the Activin-MEKK1-JNK and the TGFalpha-EGFR-ERK. These two pathways are required for the correct eyelid development of both the epithelial and mesenchymal cell layers. The pathways are also important in coordinating the interactions between the two cell layers.

In the genome the genes encoding important matters are found. For example chromosome 6 within the SLC2A9. SLC2A9 encodes glucose, fructose and urate transport. In muscle cells it is involved in muscle tone contributions to the development of entropion. Another example is the chromosome 16 within OAR16-14874751, which encodes for Neurolysin. Neurolysin cleaves many substrates including Neurotensin, which has an important role in energy metabolism and it also increases epidermal growth factor expression. Neurotensin is

important for eyelid development and does also have a role in the macrophage migration and inflammation response under hyperglycemic conditions. Macrophages are one of the major cell types within eyelids. It is possible that SLC2A9 is functionally related to OAR16-14874751 in the development of entropion through the influence of sugar transport on neurotensin activity. Chromosome 1 within PIK3CB has an important role in the cellular pathway. Chromosome 2 within MYO3B mediates movement of cells and do also have a role in the adipose deposition. Because decreased adipose tissue during development can cause entropion. Chromosome 13 within KCNB1 has a role in apoptosis in neurons. With other words, it has a role in cell volume. Chromosome 15 within ZC3H12C has a role in inhibiting inflammation in vitro. Chromosome 9 within JPH1 has a role in skeletal muscle in the intramembrane Ca2+ movement. If the Ca2+ channel is not working, the eyelid gets weak and in that way it might be causing entropion.

2.2.5 Brachygnathia inferior

2.2.5.1 General information of brachygnathia inferior

Brachygnathia inferior is the shortness of lower jaw. It is also called underbite, overshot and parrot mouth. It is a common anomaly in sheep. Brachygnatia can be a hereditary malformation, come from a viral infection, teratogenic drugs, alkaloids of plants or a maternal deficiency of iron. (The latter one is still under discussion). The cause is most likely oligogenic inheritance including a dominant and recessive locus responsible for the major gene effect.

2.2.5.2 Clinical signs of brachygnathia inferior

The malformation can be seen in many sheep breeds and the affection can be very different from case to case. In extremely affected animals they can develop palatine ulcers and have growth retardation. Because the palatine ulcers cause malocclusion of the incisors, it can have a negative effect on grazing efficiency and, consequently, body condition and production.

2.2.5.3 Brachygnathism, cardiomegaly and renal hypoplasia syndrome

It is a lethal, autosomal recessive disorder that results in brachygnathism, cardiomegaly, and renal hypoplasia (BCRHS) in poll Merino sheep. The disorder causes dwarfism or a small body size with multiple congenital defects. The congenital defects are mandibular

abnormalities, enlarged heart and liver, small kidneys and the skull may be smaller than normal causing exophthalmia. The diseased lambs may be stillborn or they may die immediately after birth. Annual incidence in a sheep flock were the syndrome has been identified is around 2.5%.

By genotyping with SNP50 Beadchip, association and homozygosity mapping analyses the gene region causing the syndrome has been identified. The region comprises 20 consecutive SNPs spanning of the distal end of OAR2. It has been found that all affected lambs were homozygous for the associated haplotype in this region.

2.2.6 Cleft palate

2.2.6.1 General information of cleft palate

Cleft palate is also called palatoschisis. The defect is found centrally in either or both hard and soft palate. A cleft palate is when the roof of the mouth contains an opening into the nose. These disorders can result in feeding problems in the way that the lambs may have difficulties in suckling, they may also have hearing problems, frequent ear infections and nasal regurgitation. The lambs usually die within few days of life because of aspiration pneumonia.

Cleft palate is not so common in sheep. However, if it is found it is often associated with other diseases. The disorder can for example be found together with microtia. The cause of the disorder can be either genetically or by the ingestion of Veratrum Californicum.

2.2.6.2 Genetics of cleft palate

A deficiency of GTP cyclohydrolase, encoded by the GCH1 gene, results in two neurological diseases: hyperphenylalaninaemia type HPABH4B and DOPA-responsive dystonia. Genes involved in neurotransmitter metabolism and motor systems may contribute to palatogenesis.

2.2.7 Hernias

2.2.7.1 General information of hernias

A true hernia is defined as having a hernia ring, sac and content. They can be direct, through a rent in the body wall, or they might be indirect, through an already existing ring, for example through the inguinal ring or the umbilical ring. Although, congenital hernias tend to be indirect even though direct hernias might happen during dystocia or obstetrical manipulation. Hernia is a well known disorder in sheep. There are different types of hernias, depending on their anatomic position. The most common hernia type in sheep is the ones in the abdominal wall; especially the umbilical hernia, inguinal and scrotal hernia. Diaphragmatic hernia might also be seen.

Umbilical hernia is more seen in females than in males, and more common in lambs less than 6 months. The umbilical hernia may be congenital or develop shortly after birth. Often they are considered to be inherited, but they might also be acquired due to infection of the umbilical cord which leads to imperfect closure of the umbilical opening. It can also be a cause of the exertion of the abdominal muscles during playing or due to external trauma, this can cause infection and abscess formation which will weaken the tissues of the abdominal wall. Another cause can be as simple as paroxysm of coughing and dyspnea.

2.2.7.2 Persistent pulmonary hypertension

In diaphragmatic hernia inhaled nitric oxide plays a major role in the modulation of perinatal pulmonary vascular tone. Congenital diaphragmatic hernia is a major cause of severe persistent pulmonary hypertension of the newborn. Alterations in NO/cyclic guanosine 3.5 monophosphate (cGMP)-mediated pulmonary vasodilatation may contribute to Persistent pulmonary hypertension of the newborn in congenital diaphragmatic hernia.

It was found to be relaxation of fourth generation intralobar pulmonary artery rings in response to the endothelium-dependent vasodilator, acetylcholine, and to the specific inhibitor of cGMP-phosphodiesterase, zaprinast. It was also relaxation because of the impaired calcium ionophore A23187. Relaxation in response to the NO donor sodium nitroprusside was also impaired in congenital diaphragmatic hernia animals as compared with controls. Repeating the challenge increased vasorelaxation in response to sodium nitroprusside in congenital diaphragmatic hernia.

Immunohistochemistry revealed the presence of endothelial NO-synthase in the endothelium of pulmonary arteries from both control and congenital diaphragmatic hernia animals. the conclusion was that endothelium-dependent vasodilatation in response to ACh and A23187 was differently affected in the fetal surgical congenital diaphragmatic hernia-lamb model. Furthermore, activity of sGC but not that of PDE was impaired in congenital diaphragmatic hernia animals. PPHN and decreased inhaled NO responsiveness in congenital diaphragmatic hernia may involve decreased sGC activity.

2.2.8 Atresia ani

2.2.8.1 General information of atresia ani

Atresia ani is the failure of the development of the anal opening. It is a congenital abnormality, with clinical sigs are manifested by an absence of feces, dullness, anorexia with abdominal distension, discomfort and straining at an attempt to defecate. The rectal lumen can sometimes be seen bulging subcutaneously at the normal site of the anus when the abdomen is compressed. In sheep recto-vaginal fistula was also noticed with this abnormality.

2.2.8.2 Treatment of atresia ani

The site is selected for operation and prepared for surgery. The animal is operated under local infiltration anesthesia with Lignocaine HCl 2% solution injected subcutaneously. After the cruciate skin incision, the blind rectum is identified and opened, which voided feces and air on abdominal compression. The rectal walls are sutured with the skin. Such cases of atresia ani with normal rectal development are surgically treated with 100% success rates provided the anal sphincter is not damaged.

2.2.9 Microtia

2.2.9.1 General information of microtia

Microtia is a developmental disorder where the ear is small or abnormally shaped. It can occur unilaterally or bilaterally, though unilateral is more commonly seen. The ear canal may be narrow, blocked or absent. Microtia can be the only clinical sign or the disorder can be a part of a syndrome. The annually prevalence in a sheep flock is 0.8-4.2 out of 10 000, and it is

more commonly found in males. Microtia can be predisposed genetically or by the environment.

2.2.9.2 Genetics of microtia

It can be an autosomal dominant or recessive disease or it can be due to chromosomal aberrations. It is several genes that are responsible for the disorder.

2.2.10 Rectal prolapse

2.2.10.1 General information of rectal prolapse

A rectal prolapse is a protrusion of the rectal tissue through the exterior of the body. It usually begins as a small round area that sticks out when the lamb lays down or coughs. In extreme cases, the intestines can pass through the opening and the disease can in some cases be fatal. There are many predisposing factors causing rectal prolapses, including genetics, short tail docks, coughing, weather, stress, and high concentrate diets.

2.2.10.2 Treatment of rectal prolapse

Rectal prolapse is a serious defect most commonly associated with the meat-type sheep. It is most common among lambs fed a high-concentrate ration. It is believed that this weakness is due to inheritance. This condition is sometimes corrected by surgery, but affected animals often continue to prolapse after surgery, therefore the best way of resolving this problem is by culling the affected animals from the flock.

Usually, lambs with prolapsed rectums are prematurely slaughtered or sent to the market. But it is possible to repair a rectal prolapse by amputating the prolapsed part of the rectum or by putting the prolapsed rectum inside again and by making some stitches around the anus opening it holds it inside. These lambs should not be kept for breeding.

2.2.10.3 Genetics of rectal prolapse

Lambs in feedlots had higher incidence of rectal prolapse than in grazing lambs. It tend to occur more often in ewe lambs and it is more often seen in black-faced sheep than in white-faced sheep. The link between extreme tail docking and the incidence of rectal prolapses in grain-fed lambs has been scientifically established.

Genetic analysis of rectal prolapse using half-siblings indicated a low heritability factor of around 0.14.

2.2.11 Microphtalmia

2.2.11.1 General information of microphtalmia

Microphthalmia in sheep is an autosomal recessive inherited congenital anomaly. The disoder is characterized by extremely small or absent eyes and affected lambs are absolutely blind from birth.

Morphological studies showed that impaired lens formation seems to be the major cause of anophthalmia and microphthalmia, although the precise pathogenesis of these phenotypes remains unknown. Lens development is a critical embryonic period in a vertebrate eye development, during which many inductive signals are exchanged between the optic vesicle and surface ectoderm. This stage is characterized by formation of the lens placode, a thickening of the surface ectoderm that comes into contact with the optic vesicle. Coordinated invagination of the lens placode and the optic vesicle results in the formation of the lens vesicle and a double-layered optic cup and provides the first indication of the final shape of the eye.

2.2.11.2 Genetics of microphtalmia

A set of putative transcription factors required for the earliest step of eye development have been identified in vertebrates. The involvement of homologous proteins in the lens development is subsequently elucidated by the characterization of mutations that cause congenital human or murine ocular disorders and their comparison to mutations in model organisms.

Analyzing inherited isolated microphthalmia/anophthalmia in humans revealed a total of eight genes (SOX2, PAX6, OTX2, RAX, CHX10, FOXE3, PITX3, CRYBA4) carrying causative mutations. The role of the eight genes during lens development was confirmed by studying spontaneous mouse mutants and genetically engineered mice with more or less similar ocular phenotypes. Besides CRYBA4, encoding a lens specific structural protein, seven of these

genes encode transcription factors which are required for appropriate lens formation during eye development.

Isolated congenital microphthalmia occurs in various mammalian species, including the Texel breed of sheep. In Texel sheep, microphthalmia behaves as a monogenic autosomal recessive trait. An abnormal development of the lens vesicle has shown to be the primary event, but so far the underlying genetic defect has not been elucidated. In an initial analysis, it has been performed a partial genome scan and it has been observed genetic linkage to microsatellite markers on sheep chromosome. But the results have not been confirmed.

Therefore, it can only be hypothesized that a more comprehensive genome wide mapping strategy may lead to the identification of the microphthalmia locus in Texel sheep. The ability to assay 50 000 evenly spaced SNP across the sheep genome was recently made possible by development of Illumina's OvineSNP50 BeadChip. OvineSNP50 BeadChip allowed us to localize the causative mutation for microphthalmia to a 2.4 Mb interval on the sheep chromosome 22 by association and homozygosity mapping. The PITX3 gene is located within this interval and encodes a homeodomain containing transcription factor involved in vertebrate lens formation. An abnormal development of the lens vesicle was shown to be the primary event in ovine microphthalmia.

In a trial it was genotyped from 23 microphthalmia affected lambs and 23 control sheep. A genome wide significant association was shown for SNPs on sheep chromosome 22 (OAR 22). Of the remaining 44,865 SNPs with a genotyping rate over 99% and minor allele frequency over 5%, eights SNPs, over the region 24,529,089 to 28,147,610 on OAR 22, showed strongest association with the microphthalmia phenotype. No other region in the genome showed genome wide associated SNPs. The OAR 22 SNP at position 24,952,721 showed strongest association with the microphthalmia phenotype.

Subsequently, it was applied a homozygosity mapping approach to narrow the region containing the microphthalmia mutation. Based on the reported occurrence of microphthalmia, some generations after the introgression of Texel sheep from the Netherlands we hypothesized that the affected sheep most likely were inbred to one single founder animal. Under this scenario the affected lambs were expected to be identical by descent (IBD) for the causative mutation and flanking chromosomal segments. We analyzed the cases for extended regions of homozygosity with simultaneous allele sharing.

Only one genome region fulfilled those search criteria. On OAR 22 all 23 affected genotyped sheep were homozygous and shared identical alleles over 39 SNP markers.

In order to further examine the critical interval defined by using SNP data, we genotyped three microsatellite markers derived from the surrounding virtual genome sequence of OAR 22. The analysis of the microsatellite genotypes confirmed an increased homozygosity within the microphthalmia affected lambs compared to the controls. The observed microsatellite heterozygosity in the cases ranged from 3–52% compared to 84–92% in the controls. A total of 130 out of 134 microphthalmia affected lambs showed homozygosity at microsatellite INRA81 located at 24.9 Mb on the virtual genome map of OAR 22.

The selection against this candidate causative mutation can now be used to eliminate microphthalmia. Furthermore, the identification of a naturally occurring PITX3 mutation offers the opportunity to use the Texel sheep as a genetically characterized large animal model for human microphthalmia.

Historically, the development of genomic tools for the sheep genome has lagged behind those of other major livestock species such as the cattle and chicken. This has limited the ability to identify genes controlling specific traits of interest. The development of low density microsatellite based linkage maps has lead to the mapping of Mendelian diseases and subsequent discovery of mutations underlying at least three genetic diseases in sheep, however many others remain uncharacterized. The recent development of a set of SNP markers distributed across the sheep genome has changed the prerequisites for such gene mapping projects. For the first time, it was demonstrated the use of a genome-wide ovine SNP array for efficient positional cloning of a Mendelian trait in sheep. The result illustrates the power of genome-wide association analysis in domestic animals for the genetic dissection of trait loci.

2.3 Summary

There are a lot of different disorders that can be found in sheep, some more common than others. In Norway these disorders has been described in literature as common disorders:

- Spider Lamb Syndrome (SLS)
- Cryptorchidism
- Inverted eyelids, also called entropion.
- Brachygnathia
- Cleft palate
- umbilical/abdominal and scrotal/inguinal hernias
- Antresia ani
- Microtia
- rectal prolapse
- microphtamia

Most inherited disorders are recessive in nature. That means that for the animal to get the disorder, two copies are needed. The animals having one copy are carriers. Recessive disorders are especially important when there are few animals (small genome). It is also important when there are a small number of animals used to improve the production, like in selection.

The disorders can be genetically mapped to get a full overview over which genes causing the disorder and which animals that is carriers the disease genes. In this way the disorders can be avoided.

The recent development of a set of SNP markers distributed across the sheep genome has changed the prerequisites for such gene mapping projects. For the first time, it was demonstrated the use of a genome wide ovine SNP array for efficient positional cloning of a Mendelian trait in sheep. The result illustrates the power of genome-wide association analysis in domestic animals for the genetic dissection of trait loci.

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Tables

Genomic Resource	Description and reference
Draft Reference Genome	Launched here at PAG 2011: Cattle and Sheep workshop
Linkage Map	Microsatellite based linkage map produced through genotyping of the International Mapping Flock. See Maddox et al.,2001
Virtual Sheep Genome	Comparative analysis of BAC end sequences to generate a virtual assembly http//www.livestockgenomics.csiro.au/vsheep/
CHORI-243 BAC library	A 12 fold coverage BAC library constructed from a single male Texel. Sequencing contributed to the draft reference genome assembly
Illumina GA sequence and 76,000 SNP	Deep re-sequencing of pooled genomic DNA from 60 animals identified approximately 76,000 SNP
Roche 454 sequence and 595,000 SNP	Three-fold coverage of 454 sequences derived from six animals. Alignment identified approximately 595,000 SNP. http//isgcdata.agresearch.co.nz/
Ovine SNP50 BeadChip	Illumina Infinium based platform for genotyping 50,000 SNP
INRA 1200-rad RH Panel	RH panel used for the assignment of sequence tagged sites. See Laurent et al.,2007
USDA 5000-rad RH Panel	RH panel used for the assignment of sequence tagged sites.

"The International Sheep Genomics Consortium" has developed a range of resources for the ovine research community.

Table 1: http://www.sheephapmap.org/news/GWAS_announcement.pdf

Trait	Description	Map Location	Contacts and reference
Achondroplacia	Limb malformation in UK Cheviots	OAR3: Mb 153- 157	James Kijas/Steve Bishop
Chondroplasia	Dwarfism/ Limb malformation in Texel	OAR4: 1 MB region	Dorrian Garrick
Microphthalmia	Ocular abnormality in Texel	OAR22: PITX3 mutation	Cord Drogemuller
Poll	Absence of horn	OAR10: RXFP2 gene	John McEwen and Jon Slate
Juctional Epidermolysis	Skin disease of German Black Headed sheep	OAR12	Ottmar Distl
Yellow fat	Yellowing of adipose tissue in Perendale	OAR15	John McEwen

"The International Sheep Genomics Consortium" list of studies where genes have been mapped to small genomic regions.

Table 2: http://www.sheephapmap.org/news/GWAS_announcement.pdf