

The effect of the double muscling gene on production efficiency and meat quality

Introduction

In an earlier study, the Roslin Institute showed that the allele of the myostatin (growth differentiation factor 8, *GDF8*) gene with an 11 bp deletion (*del*11) was associated with increased muscling and calving difficulty, and decreased fat depth in the South Devon breed (Wiener et al 2002). The analysis was, however, limited by a relatively small sample size. The aim of the current study was to examine in more detail the effect of the double muscling allele on a range of traits of economic importance, including growth, carcass conformation and meat quality.

Objective

The objective of this project was to investigate the effects of alleles of the myostatin (“double muscling”) gene, both in the homozygous and heterozygous form, on beef production.

Materials and Methods

Calving data, including sex, weight at birth, calving score, whether the calf was a twin and if the animals died at birth were provided by SDHBS (South Devon Breed Society) members, along with blood samples of all parents and offspring. Calving scores were treated as a trait of the calf rather than of the dam. Data from 1314 calvings were provided by twelve breeders. In addition information from all registered South Devon individuals born in the UK between 1963 and 2005 was collected from Breedplan and Signet.

Carcasses were classified commercially using the “EUROP” conformation score and standard fat scores were recorded by slaughter plant Meat Hygiene Service staff. A subset of carcasses were classified using a standardized MLC 15-point numerical scale by a single professional inspector (Mr. Homer) from the Meat and Livestock Commission (MLC). Whole hot scale-weight of the carcass was estimated by adding the hot scale-weights of the left and right sides of the carcass.

Sensory traits and fatty acids. A subset of carcasses was measured for chemical, organoleptic (taste panel) and fatty acid composition. Analyses were done at the food lab in Bristol University using the protocols outlined in Appendix 1. Sensory analysis was carried out by a trained taste panel.

Statistical Analysis methods and models are described in Appendix 2. Briefly these adjust for the nuisance parameters inherent in the field study and address the impact of the “substitution effect” and whether there is a “dominance effect”. The analyses also address whether there are benefits from planned breeding to obtain heterozygous individuals, and if the different effects of the myostatin gene are related.

Results

PART 1. SOUTH DEVON

Calving score and birth weight. The myostatin allele with an 11 bp deletion (*MH*) is associated with more difficult calving. The effect is partially dominant such that heterozygotes (*MH/+*) had a lower calving score than would be expected by averaging *MH/MH* and wild type (*+/+*) individuals. The maternal genotype did not have a significant effect on either calving score or birth weight. Overall, male calves had higher calving scores than females and the *MH* allele had a greater effect on males than on females. A population composed of all *MH/MH* animals would have an average calving score of 0.6 higher than a population of all *+/+* animals (Table 1). Calving score also significantly increased with increasing birth weight at a rate of about 0.023 points per kilogram (Table 1). The *MH* allele was associated with significantly increased birth weight of calves, by approximately 2 kg per copy, again with a much greater effect on males than on females. Nevertheless there was clear evidence that the impact of the *MH* allele on calving difficulty was not mediated solely through birth weight.

Carcass traits. The *MH* allele was associated with increased hot weight at slaughter (~17 kg) and was additive, so *MH/MH* carcasses were ~34 kg heavier than *+/+* carcass. The *MH* allele was associated with increases in all six muscle score parameters (see Table 1), where the effects were very similar. All traits except top piece and EUROP score exhibited dominance. The direction of these dominance effects were all negative, such that the *MH/+* animals were more similar to the *+/+* than to the *MH/MH* animals. The *MH* allele was associated with significantly reduced fat and subcutaneous fat scores with additive effects of approximately -1.2 and -1.3 units (Table 1).

Organoleptic and Fatty acids analyses. This study represents the first time that the effects of myostatin alleles on taste and flavour have been investigated. Six traits were recorded: juiciness, texture, abnormal flavour, beef flavour, flavour liking and overall liking. Significant associations were observed between myostatin genotypes and the flavour traits and overall liking, but not with texture and juiciness or mechanical yield force (Table 1). The *MH* allele decreased the score for desirable traits (beef flavour, flavour liking and overall liking) and increased that for abnormal flavour. There was no evidence of dominance so that heterozygotes appeared intermediate. The *MH* allele was associated with reduced fat in the muscle and the effect was greatest for the neutral fatty acids. Significantly reduced total saturated fatty acids (SFA) and amounts of polyunsaturated fatty acids (PUFA) in the neutral fatty acids were associated with the *MH* allele. There was no evidence to support the view that the impact of the *MH* allele on flavour and liking were solely due to differences in fatness or specific fats.

Growth was approximately linear with mean weights of 64 kg (s.e. 13.6) at day 10 and 556 kg (s.e. 19.4) at day 504 (average slope ~ 0.996 kg/day, see Fig 1).

Although there was no evidence of initial differences in weight, by 252 days the animals with the *MH* allele had lower liveweights (-5.69 kg s.e. 1.82) with clear dominance, *+/+* and *+/MH* animals having similar weights while *MH/MH* animals were lighter. Liveweight gain for *MH/MH* genotype was also slower from 36 weeks of age onwards. See Fig. 1.

Interpretation of results

There are advantages and disadvantages associated with the *MH* allele. On the positive side, this allele is associated with increases in the hot carcass weight, muscle conformation of animals and the PUFA:SFA ratio, and in reduced fat levels. Although we did not have direct data on this, based on the *MH* allele being associated with lighter animals but heavier carcasses, it is likely that it also increases the saleable meat percentage. However, on the negative side the *MH* allele is also associated with increased calving scores (difficulty), reductions in desirable flavour characteristics. Furthermore, the *MH* allele is associated with reduced liveweight gain. The heterozygote benefited from favourable dominance in calving difficulty and liveweight gain, but dominance was unfavourable in muscle scores, whilst for all other traits there was no evidence for the heterozygote being different from the average of the homozygotes.

The differences in gain observed may be due to differences in nutrient requirements for the *MH* homozygotes compared to other animals, as a result of their increased muscling and consequently increased demand for protein at a comparable weight. This interpretation is supported by the observation and direction of dominance observed in both gain and muscling scores. If this is confirmed the sub-optimal nutrition will have an impact upon total nutrient excretion and environmental impact.

PART 2. ABERDEEN ANGUS

No *MH* homozygotes were identified in Aberdeen Angus samples. There were heterozygotes on one farm out of the ten surveyed (total of 367 animals genotyped). On that farm, there were four heterozygotes out of 201 animals. From the farm survey, the overall allele frequency was 0.5%; observing all samples from the single farm must be placed in the context that this farm provided 55% of all samples, and such an event whilst extreme would occur with probability >0.09 . Two heterozygotes were half-sibs. Also, there were 17 samples provided by AAHBS (16 of which were AI bulls) of which four were heterozygotes, again two were half-sibs. All heterozygous animals for which pedigree information was available had North American ancestry.

Future R&D resulting from this project

The Roslin Institute have reported previously that the effects of the *MH* allele on muscling are variable, with some homozygous individuals having an extreme phenotype, while others have little or no overt phenotype. The reason for this variation is not known and merits further investigation. Whether this variation is also found in the range of traits investigated in the current project is not known. The numbers available were insufficient to investigate the effects in detail. It is

possible that having a more complete understanding of the way myostatin regulates different traits may allow the breeding of animals displaying only the beneficial effects.

The data on the effects of the myostatin gene on meat quality traits in the South Devon are novel. However, the number of *MH/MH* homozygotes was lower than planned, as the breeders have clearly been selecting against animals with the *MH* allele. Additional samples from this genotype would allow the effects to be more thoroughly explored. The importance of these results is strengthened by the fact that double muscling is being positively selected for in other breeds, in Europe and also in the UK, and in some cases the *MH* allele has been introgressed into new breeds and is generally increasing in frequency. Given this scenario, the results from the South Devon form a first approximation to the impact of the *MH* allele in other breeds, but it is evident from gross comparisons of Belgian Blue, South Devon, Aberdeen Angus and Highland cattle which carry the 11 bp deletion *MH* allele that the genetic background influences the phenotypic expression. It is therefore possible that the set of impacts of the myostatin allele are not repeated in other breeds, and resolving this knowledge gap is important, so that UK breeders can make more informed decisions when selecting for these alleles.

Thus, the relationship between myostatin genotype and phenotype / performance is complex. The potential complexity of this relationship is increased by the existence of additional mutations in the cattle myostatin gene, a number of which have been identified in other breeds. Thus, this project poses questions not only about the most appropriate breeding objectives to exploit, control or eliminate myostatin mutations, but also questions of fundamental biological interest concerning the genetic control of complex traits.

The study suggested that *MH/MH* animals may have sub-optimal feeding in practice, due to their greater need for protein at the later stages of development, leading to greater nutrient wastage, with consequences for environmental impact and economic efficiency. Such a hypothesis suggests that the nutrient requirements for *MH/MH* carrying cattle should be reviewed, and if necessary new recommendations developed for this allele. If differences in requirements are confirmed then there is a complementary need for farmers to be made aware of the need for identification of genotype and the best practice for customised nutrition and associated benefits. It is possible that sub-optimal nutrition in practice might explain in part the differences in meat quality found, but such a hypothesis can only be considered if and when differences in nutritional needs are established.

Industrial relevance and plans for future commercial exploitation

The data, the analysis and breeding options developed in this project have been reported to the South Devon breeders, verbally and in a more detailed report. The report provides them with the information to define whether the net impacts of the *MH* allele on the Society's breeding goal are positive or negative. The project has also provided data to inform breeding strategies to manage the gene to address optimal breeding goals for growth, conformation, calving and

composition traits and to exploit any net benefits from heterozygotes. In reaching the decisions on breeding targets it will be necessary to weigh the different traits affected taking into account whether the effects of the gene are additive or dominant.

The beneficial effects on hot carcass weight associated with myostatin genotypes are additive, so the heterozygote express some of the benefits, however, *MH* appears dominant to the wild-type allele for muscle conformation traits so that the heterozygotes do not display the benefits. The adverse effects associated with the *MH* allele on calving score are dominant in a beneficial manner, i.e. heterozygotes behave as the non-carriers. The effect on flavour traits associated with myostatin genotypes, however, are additive so that heterozygotes have worse palatability compared to the wild-type. Growth rate exhibits beneficial dominance in that the heterozygotes grow as fast as wild-type animals.

The best way to exploit this gene may be a managed breeding scheme which maintains heterozygotes in the South Devon population, while limiting the number of homozygous *MH/MH* animals. The latter animals might be considered beneficial as animals for marketing to dairy breeders, since the offspring will all be heterozygotes. This strategy would avoid the more severe negative aspects associated with the homozygous genotype. Alternatively, the double muscling *MH* allele could be eliminated, which would remove the requirement for ongoing monitoring of allele frequencies and their distribution in the population. These options have been presented, but given the complexity of the results, the South Devon Society is taking time to consider its future strategy, after which time further assistance may be provided.

Trait	Significance level	Genotypic means			Substitution effect	Dominance	No. data points
		++	+/MH	MH/MH			
Calving and birth weight							
Calving Score (1-5)	P<0.001	1.514±0.103	1.755±0.097	2.387±0.176	0.312±0.061	-0.195±0.092*	755
Birth weight (kg)	P<0.001	39.42±0.870	41.07±0.835	44.34±1.267	1.918±0.356	NS	992
Carcass traits							
Hot carcass weight (kg)	P=0.003	339.6±6.17	356.7±5.592	367.8±13.046	15.88±4.693	NS	103
Muscle score (1-15)							
EUROP score	P<0.001	7.263±0.227	8.424±0.264	10.814±0.687	1.405±0.256	NS	133
fore rib	P<0.001	7.057±0.236	8.063±0.2742	10.789±0.714	1.348±0.2681	-0.8596±0.412*	133
loin	P<0.001	7.254±0.232	8.202±0.269	10.964±0.701	1.308±0.264	-0.907±0.405*	133
rump	P<0.001	6.937±0.291	8.031±0.280	11.667±0.754	1.602±0.297	-1.271±0.4689***	121
top piece	P<0.001	7.232±0.245	8.545±0.284	11.346±0.740	1.608±0.276	NS	133
overall	P<0.001	7.311±0.229	8.442±0.265	11.271±0.691	1.468±0.260	-0.849±0.340*	133
fat classification (1-15)	P=0.002	6.913±0.328	5.670±0.298	4.809±0.905	-1.167±0.330	NS	104
subcutaneous fat %	P<0.001	7.046±0.265	5.853±0.241	4.265±0.731	-1.271±0.267	NS	104
Sensory traits							
taste panel scores (1-8)							
abnormal flavour	P<0.001	3.345±0.098	3.593±0.124	4.293±0.240	0.377±0.113	NS	80
beef flavour	P<0.001	4.093±0.070	3.815±0.075	3.514±0.166	-0.285±0.076	NS	80
flavour liking	P<0.001	4.132±0.169	3.820±0.202	3.117±0.196	-0.412±0.089	NS	80
juiciness	NS	-	-	-	-	-	80
texture	NS	-	-	-	-	-	80
overall liking	P<0.001	4.267±0.086	3.956±0.106	3.133±0.206	-0.461±0.096	0.256±0.128*	80
terminal pH	NS	-	-	-	-	-	80
yield force	NS	-	-	-	-	-	80
Total fatty acids							
saturated (SFA)	P<0.001	862.80±51.12	947.40±62.66	512.10±122.51	-66.78±59.818	259.90**	80
polyunsaturated (PUFA)	P=0.110	-	-	-	-	-	80
n6:n3	P=0.848	-	-	-	-	-	80
3nPUFA	P=0.066	-	-	-	-	-	80
PUFA:SFA	P<0.001	0.131±0.007	0.119±0.008	0.207±0.017	0.018±0.009	-0.050±0.012***	80

Table1. Summary of statistical results for calving, carcass and meat quality traits

Figure 1. Weight as a function of age of animal for the three genotype classes. Typical s.e.s are ~ 13, 12 and 20 kg at days 10, 252 and 504 respectively.



Appendix 1: Methods employed for sensory traits and fatty acid composition (Bristol University)

On receipt, a section of loin was cut and trimmed of fat and connective tissue. The muscle portion was vacuum packed and frozen for subsequent analysis of fatty acid composition.

The remaining loin was conditioned to a total of kill + 10 days. Where the abattoir delivered the samples beyond kill + 10 days, the loin was sampled and frozen immediately.

Fatty acid composition.

Fatty acid analysis was carried out by direct saponification as described in detail by Teye et al. (2006), or Scollan et al. (2001). They were hydrolysed with 2M KOH in water:methanol (1:1) and the fatty acids extracted into petroleum spirit, methylated using diazomethane and analysed by gas liquid chromatography. Muscle fatty acid results are given as proportion times 100, or as mg of fatty acid per 100 g wet tissue quantified by reference to the internal standard (21:0). Only the major fatty acids and minor components readily identified and relevant to the study are reported, representing over 90 % of the total fatty acids present. The fatty acid reported as 16:1 cis consists of both the n-9 and n-7 isomers and contaminating branched 17 - carbon fatty acids

Texture

A 77mm section of loin muscle was thawed overnight and then cooked in vacuum bags in a water bath at 80°C to a centre temperature of 78°C, cooled and held on ice overnight. From each of these cooked sections, 10 replicate blocks (20 x 10 x 10 mm) were cut, parallel to the fibre direction and sheared with Volodkevitch jaws on a Stevens CR Texture Analyser to give a measure (kg) of toughness/tenderness at the point of first failure.

Sensory testing.

The remaining section of the conditioned loin was frozen and stored at -20°C prior to sensory analysis carried out by a 10- person trained taste panel (BSI, 1993). The sample was defrosted overnight at 4°C and then cut into steaks 20mm thick. Steaks were grilled to an internal temperature of 74°C (measured by a thermocouple probe) after which all fat and connective tissue was trimmed and the muscle cut into blocks 2cm³. The blocks were wrapped in pre-labelled foils and placed in a heated incubator. The samples were then given to the assessors in random order chosen by computer. Panelists assessed four samples per session and were asked to rate the samples on eight point category scales for texture, (tender to tough) juiciness (juicy to dry), flavour intensity (weak to strong , higher values denote more favourable responses), abnormal flavour intensity (weak to strong, lower values denote more favourable responses). Two

additional hedonic questions relating to flavour liking (like to dislike) and overall liking (like to dislike) were also used. Points in the scale ranged from extremely (e.g. tender) very, moderately, slightly, slightly (tough), moderately, very, extremely.

Appendix 2 Statistical analyses

All data was analysed with models using the restricted maximum-likelihood (REML) option of Genstat. Statistical significances for the fixed effects was determined by Wald tests and test statistics were compared to a χ^2 distribution with the appropriate degrees of freedom.

The general approach, described in detail below was:

- i. Model the effect of the MH gene by fitting a factor of 2 d.f. to the genotypes i.e. +/+, +/ MH, MH/MH.
- ii. The factor was then replaced by 2 covariates: the first the number of MH alleles carried i.e. 0, 1 and 2 for +/+, +/ MH, MH/MH respectively, and the second defined by 0 for +/+ and MH/MH and 1 for +/ MH. Fitting both these covariates together is an equivalent model to (i), but the regression coefficient for the second covariate is an estimate of dominance, and hence a statistical test for the presence of dominance is whether or not the regression coefficient is different from 0. In this model the regression coefficient for the first covariate estimates $\frac{1}{2}$ the difference between the two homozygotes.
- iii. The covariate for dominance was removed (irrespective of its significance) and only the regression on MH number was included in the model. The coefficient for this covariate estimates the α , average effect of an allele substitution, and estimates the impact of substituting a randomly chosen + allele for an MH allele. If q is the frequency of the MH allele in the population then the contribution to the EBV from the double muscling gene is given by $-2q\alpha$, $(1-2q)\alpha$, $2(1-q)\alpha$ for +/+, +/ MH and MH/MH respectively.

The statistical models used for of the traits described above are presented below. Once the full model was run, all non-significant factors were dropped from the model prior to estimating the effect of the myostatin gene on the traits. Also in order to increase the amount of data for the analysis, some factors such as age and sire were removed as many records did not have this data.

Calving score

Full Model tested

$$Y_{ijklmn} = \mu + S_i + P_j + G_k + B_l + F_m + D_n + M_n + Y_o + T_p + \sum_{ijklmnopq} + \Delta_{ijklmopr} + e_{ijklmnopqr}$$

S_i = Fixed effect of sex (male, female)

P_j = Fixed effect of Parity (0-12)

G_k = Fixed effect of the calf's genotype (0,1,2: ++, MH/+, MH/MH)

B_l = Fixed effect of Birth weight

F_m = Fixed effect of farm (1-12)

D_n = Fixed effect of dam maternal genotype

T_p = Fixed effect of multiple birth (twinning)

Y_o = Fixed effect of Year of birth

M_n = Fixed effect of season of birth

\sum_{ijklmn} = Random effect of sire

Δ_{ijklmo} =Random effect of dam
 $e_{ijklmnop}$ =Residual error

Full Final Model used to estimate effect of myostatin on trait:

$$Y_{ijklmn} = \mu + S_i + P_j + G_k + B_l + F_m + D_n + \Sigma_{ijklmnp} + \Delta_{ijklmq} + e_{ijklmnopq}$$
 S_i = Fixed effect of sex (male, female)
 P_j = Fixed effect of Parity (0-12)
 G_k =Fixed effect of the calf's genotype (0,1,2: ++, MH/+, MH/MH)
 B_l = Fixed effect of Birth weight
 F_m =Fixed effect of farm (1-12)
 D_n =Fixed effect of dam maternal genotype
 Σ_{ijklmn} =Random effect of sire
 Δ_{ijklmo} =Random effect of dam
 $e_{ijklmnop}$ =Residual error

Additional Model used to estimate effect of myostatin on trait:

$$Y_{ijklmn} = \mu + S_i + P_j + G_k + B_l + F_m + D_n + (SG)_{ik} + \Sigma_{ijklmnp} + \Delta_{ijklmq} + e_{ijklmnopq}$$
 S_i = Fixed effect of sex (male, female)
 P_j = Fixed effect of Parity (0-12)
 G_k =Fixed effect of the calf's genotype (0,1,2: ++, MH/+, MH/MH)
 B_l = Fixed effect of Birth weight
 F_m =Fixed effect of farm (1-12)
 D_n =Fixed effect of dam maternal genotype
 $(SG)_{ik}$ = Interaction between sex and genotype
 Σ_{ijklmn} =Random effect of sire
 Δ_{ijklmo} =Random effect of dam
 $e_{ijklmnop}$ =Residual error

Birth weight

Full Final model tested:

$$Y_{ijklmn} = \mu + S_i + M_j + G_k + F_l + T_m + P_n + D_o + \Sigma_{ijklmnop} + \Delta_{ijklmoq} + e_{ijklmnopq}$$
 S_i = Fixed effect of sex (male, female)
 M_j = Fixed effect season of birth (1-4)
 G_k =Fixed effect of the calf's genotype (0,1,2: ++, MH/+, MH/MH)
 F_m =Fixed effect of farm (1-12)
 T_m =Fixed effect of multiple births (twinning)
 P_n =Fixed effect of parity
 D_n =Fixed effect of Year of birth
 Σ_{ijklmn} =Random effect of sire
 Δ_{ijklmo} =Random effect of dam
 $e_{ijklmnop}$ =Residual error

Full Final Model used to estimate effect of myostatin on trait:

$$Y_{ijklmn} = \mu + S_i + Y_j + G_k + F_l + M_m + (SG)_{ik} + \Sigma_{ijklmo} + \Delta_{ijklmp} + e_{ijklmnop}$$

S_i = Fixed effect of sex (male, female)
 Y_j = Fixed effect season of birth (1-4)
 G_k = Fixed effect of the calf's genotype (0,1,2: ++, MH/+, MH/MH)
 F_m = Fixed effect of farm (1-12)
 M_m = Fixed effect of multiple births
 Σ_{ijklmn} = Random effect of sire
 Δ_{ijklmo} = Random effect of dam
 $e_{ijklmnop}$ = Residual error

Additional model used to estimate effect of myostatin on trait:

$$Y_{ijklmn} = \mu + S_i + Y_j + G_k + F_l + M_m + \Sigma_{ijklmo} + \Delta_{ijklmp} + e_{ijklmnop}$$

S_i = Fixed effect of sex (male, female)
 Y_j = Fixed effect season of birth (1-4)
 G_k = Fixed effect of the calf's genotype (0,1,2: ++, MH/+, MH/MH)
 F_m = Fixed effect of farm (1-12)
 M_m = Fixed effect of multiple births
 $(SG)_{ik}$ = Interaction between sex and genotype
 Σ_{ijklmn} = Random effect of sire
 Δ_{ijklmo} = Random effect of dam
 $e_{ijklmnop}$ = Residual error

Carcass traits (Bristol) and Organoleptic analysis

Full model tested for all traits:

$$Y_{ijklmn} = \mu + S_i + G_k + R_l + F_m + A_n + C_m + \Sigma_{ijklmnp} + e_{ijklmnopq}$$

S_i = Fixed effect of sex (male, female)
 G_k = Fixed effect of the calf's genotype (0,1,2: ++, MH/+, MH/MH)
 R_l = Fixed effect of sample age
 F_m = Fixed effect of farm (1-12)
 A_n = Fixed effect of Age (Slaughter date- birth day)
 C_m = Fixed effect of Fat as measured on conformation scale
 Σ_{ijklmn} = Random effect of sire
 Δ_{ijklmn} = Random effect of taste panel (*for sensory data only*)
 $e_{ijklmnop}$ = Residual error

Final model tested for all traits:

$$Y_{ijklmn} = \mu + S_i + G_k + R_l + F_m + A_n + \Sigma_{ijklmnp} + e_{ijklmnopq}$$

S_i = Fixed effect of sex (male, female)
 G_k = Fixed effect of the calf's genotype (0,1,2: ++, MH/+, MH/MH)
 R_l = Fixed effect of sample age
 F_m = Fixed effect of farm (1-12)
 A_n = Fixed effect of Age (Slaughter date- birth day)
 Σ_{ijklmn} = Random effect of sire
 Δ_{ijklmn} = Random effect of taste panel (*for sensory data only*)
 $e_{ijklmnop}$ = Residual error

The final models used to estimate the effects of myostatin on the traits are:

- i) Abnormal flavour: Calf genotype, farm and panel
- ii) Beef flavour: Calf genotype and representative
- iii) Flavour liking: Calf genotype, farm, sample age, sex and panel
- iv) Juiciness: Calf genotype, farm, sample age, sex and panel
- v) Overall liking: Calf genotype, farm and panel
- vi) Terminal pH: Calf genotype, farm and sex
- vii) Texture: Calf genotype, farm and sex
- viii) Conformation: Calf genotype and farm
- ix) Fat: Genotype

Carcass traits

A new factor called FA_AB(FArm and ABattoir) was created which was a combination of farm and abattoir because the two factors are confounded i.e. each farm sent their animals to the same abattoir apart from one farm which sent them to two

General Model tested:

$$Y_{ijklmn} = \mu + S_i + G_k + R_l + F_m + A_n + \sum_{ijklmnp} + e_{ijklmnopq}$$

S_i = Fixed effect of sex (male, female)

G_k = Fixed effect of the calf's genotype (0,1,2: ++, MH/+, MH/MH)

F_m = Fixed effect of farm and abattoir

A_n = Fixed effect of Age (Slaughter date- birth day)

\sum_{ijklmn} = Random effect of sire

$e_{ijklmnop}$ = Residual error

The final model used to estimate the effects of myostatin on the traits are:

- i) Hot carcass weights: Sex, Calf genotype, FA-AB and age
- ii) Frib: FA-Ab and calf genotype
- iii) Fat: Age, Calf genotype and FA_AB
- iv) Loin: FA_AB and calf genotype
- v) Overall: FA_AB and calf genotype
- vi) Subcutaneous fat percentage = Calf genotype, FA-AB and age
- vii) Conformation = FA_AB and calf genotype
- viii) Rump = Sex and calf genotype
- ix) Top = FA_AB and calf genotype

Fatty acids

Full model tested:

$$Y_{ijklmn} = \mu + S_i + G_k + R_l + F_m + A_n + \sum_{ijklmnp} + e_{ijklmnopq}$$

S_i = Fixed effect of sex (male, female)

G_k = Fixed effect of the calf's genotype (0,1,2: ++, MH/+, MH/MH)

F_m = Fixed effect of farm (1-12)

A_n = Fixed effect of Age (Slaughter date- birth day)

\sum_{ijklmn} = Random effect of sire

$e_{ijklmnop}$ =Residual error

The final model used to estimate the effects of myostatin on the traits are:

Neutral fatty acids

- i) SFA: Calf genotype and farm
- ii) PUFA: Calf genotype and farm
- iii) n-6:n-3 ration: Calf genotype, sex and farm
- iv) 3n PUFA: Calf genotype and farm
- v) P:S: Calf genotype and farm

Phospholipids and Total fatty acids

The models used were the same as those for neutral fatty acids above with the exception of P:S ratio where