

# Fatty acid composition and lipogenic enzyme expression in *semimembranosus* muscle of Limousin and Aberdeen Angus crossbred cattle.

R.Ward<sup>1</sup>, B.W.Woodward<sup>2</sup>, J.D.Nkrumah<sup>2</sup>, N.Otter<sup>3</sup>, and O.Doran<sup>4</sup>

<sup>1</sup>Dept of Clinical Veterinary Science, University of Bristol, Langford, Bristol, UK. <sup>2</sup>Meril Limited, Duluth, GA, USA. <sup>3</sup>Meril Animal Health Limited, Harlow, Essex, UK. <sup>4</sup>Institute of Biosensing Technology, University of the West of England, Coldharbour Lane, Bristol BS16 1QY, UK

## Introduction

- Intramuscular fat (IMF) is an important meat quality characteristic.
- The molecular mechanism controlling IMF deposition in cattle is still unclear.
- The enzymes acetyl CoA carboxylase (ACC) and fatty acid synthase (FAS) are known to play the key role in saturated fatty acid biosynthesis.
- Stearoyl CoA desaturase is associated with the monounsaturated fatty acids (MUFAs) biosynthesis
- The lipogenic enzymes delta-5-desaturase (D-5-D) and delta-6-desaturase (D-6-D) are associated with the regulation of longer chain polyunsaturated fatty acids formation (PUFAs)

**Aim:** The work presented here aimed to investigate the relationship between expression of the key lipogenic enzymes and IMF level in cattle of two breeds.

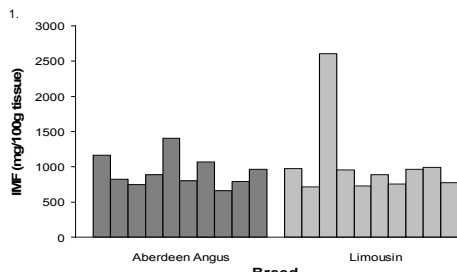
## Methods

**Animals:** Experiments were conducted on 10 Aberdeen Angus crossbred and 10 Limousin crossbred steers of approx. 25 months of age.

**Fatty Acid Analysis:** Fatty acids were isolated and extracted from *semimembranosus* muscle using the folch procedure. Free fatty acids were methylated with diazomethane, before being analysed by high resolution gas chromatography.

**Enzyme expression:** The isolation of microsomal and cytosolic fractions from muscles was performed by differential centrifugation. Proteins were separated by SDS-PAGE, and incubated with an appropriate primary antibody, followed by incubation with horseradish peroxidase linked donkey anti-goat IgG secondary antibody. The protein bands were visualised using ECL reagent and the intensity of signals was analysed using Image Quant software.

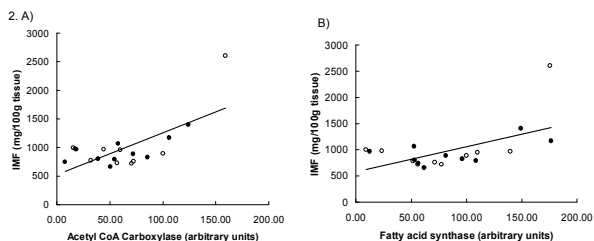
## Results



**Fig 1.** Total intramuscular fatty acid content (IMF) in *semimembranosus* muscle of Limousin and Aberdeen Angus crossbred cattle. Each bar represents an individual animal. Total IMF content was calculated by the sum weight of all fatty acids.

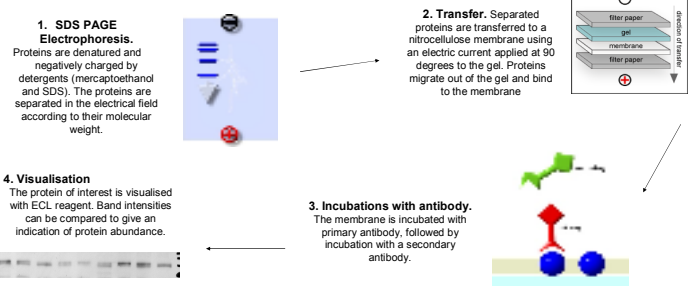
Parameter	Breed			
	Limousin		Aberdeen Angus	
	Average	SEM	Average	SEM
Intramuscular fat content	1036.74	177.646	930.494	71.180
Acetyl CoA Carboxylase	62.9	13.5	61.5	11.5
Fatty Acid Synthase	81.4	16.2	84.8	15.6
Stearoyl CoA Desaturase	106.8	17.0	85.8	9.5
Delta 6 Desaturase	50.1	11.4	51.8	12.0
Delta 5 Desaturase	100.3	20.7	75.4	25.6

**Table 1.** Total intramuscular fat content and lipogenic enzyme expression in *semimembranosus* muscle of Limousin and Aberdeen Angus crossbred cattle.

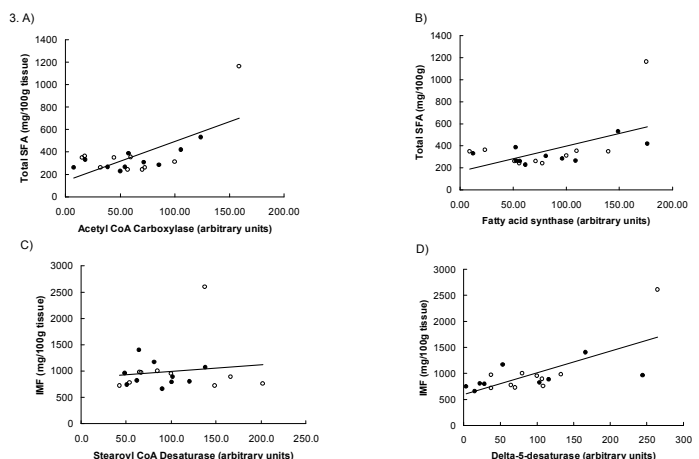


**Fig 2.** A) Relationship between acetyl CoA carboxylase protein expression and the total fatty acids in *semimembranosus* muscle (IMF). ○ represents Limousin crossbred cattle, and ● represents Aberdeen Angus crossbred cattle. The relationship can be described by the equation  $y = 7.3476x + 526.71$ ,  $r^2 = 0.4573$ ,  $P < 0.001$ .  
B) Relationship between fatty acid synthase protein expression and the total fatty acids in *semimembranosus* muscle (IMF). ○ represents Limousin crossbred cattle, and ● represents Aberdeen Angus crossbred cattle. The relationship can be described by the equation  $y = 4.8041x + 584.27$ ,  $r^2 = 0.3136$ ,  $P < 0.05$ .

### Main stages of western blotting



Pictures 1 & 3 were adapted from [www.westernblotting.org/THEORY-SDS-PAGE.htm](http://www.westernblotting.org/THEORY-SDS-PAGE.htm). Picture 2 was adapted from [www.bme.gatech.edu/civil/WesternBlotting/Background/transfer.htm](http://www.bme.gatech.edu/civil/WesternBlotting/Background/transfer.htm).



**Fig 3.**

- A) Relationship between acetyl CoA carboxylase protein expression and the total saturated fatty acid content (SFA) in *semimembranosus* muscle. The relationship can be described by the equation  $y = 3.5298x + 139.55$ ,  $r^2 = 0.4505$ ,  $P < 0.001$ .
- B) Relationship between fatty acid synthase protein expression and the total saturated fatty acid content (SFA) in *semimembranosus* muscle. The relationship can be described by the equation  $y = 2.2899x + 168.69$ ,  $r^2 = 0.3041$ ,  $P < 0.05$ .
- C) Relationship between stearoyl CoA desaturase protein expression and the total fatty acid content (IMF) in *semimembranosus* muscle. The relationship can be described by the equation  $y = 1.2793x + 860.4$ ,  $r^2 = 0.0178$ ,  $P > 0.05$ .
- D) Relationship between delta-5-desaturase protein expression and the total fatty acid content in *semimembranosus* muscle (IMF). The relationship can be described by the equation  $y = 4.1859x + 592.31$ ,  $r^2 = 0.4859$ ,  $P < 0.001$ .

In all figures ○ represents Limousin crossbred cattle, and ● represents Aberdeen Angus crossbred cattle.

## Conclusion

- Between-individual variations in IMF content were found within each breed.
- Significant positive relationships were found between ACC and FAS protein expression, and IMF and total SFA content.
- A significant positive relationship was found between D-5-D protein expression and IMF content.

- No relationship was found between the expression of the SCD enzyme and either monounsaturated fatty acid (MUFA) content (data not shown), or IMF content.
- It is suggested that the rate of SFA biosynthesis has a higher input in IMF formation in cattle than the rate of MUFA or PUFA biosynthesis. This is in contrast to monogastric animals (pigs) where MUFA biosynthesis plays the key role in IMF deposition.