

## Marbling biology – What do we know about getting fat into muscle?

### Key findings of research

- Intramuscular fat content (% fat) or marbling score is late maturing
- The expression of marbling is due to maintained fat synthesis combined with declining muscle growth as animals grow older
- A growth curve for the development of marbling found (i) a period up to approx. 200kg where intramuscular fat does not increase (ii) a period of linear development as carcass weight increases from 200-450kg and (iii) the suggestion that intramuscular fat content reaches a maximum at mature body size (approx 500kg carcass weight depending on breed)
- Expression of intramuscular fat after extended grain finishing is driven by three primary genes (i) overall fatness (ii) the degree of muscle development which also interacts with mature body size and (iii) the extent of fat distribution bias toward the intramuscular site. Further interaction of these genes will help in the final understanding of how they affect final marble score
- The level of intramuscular fat at the start of finishing is a key determinant of the final level of intramuscular fat after finishing supporting the use of ultrasound estimates as a means of selecting superior marbling animals
- Recent results from Beef CRC research in collaboration with International partners using modern biochemical and genomic tools suggest that (i) intramuscular fat cells are different to other fat depots (i.e. subcutaneous) and (ii) intramuscular fat cell development is determined relatively early in life (3-8 months of age)
- The major nutritional and/or management tool for increasing the development of marbling is to maximise the availability of net energy (and glucose) for fat synthesis during finishing
- Net energy available for fattening is the most likely reason why grain feeding (compared to grass) results in a higher marbling score at equal carcass weights
- In heavier 'British' type cattle (Live Weight  $\geq 540$ kg, P8=12mm) it is difficult to increase the net energy for fattening by reducing protein supply (that is these cattle have a low protein requirement) and this is clearly an avenue for reducing feed costs
- Increased processing of the ration (i.e. steam flaking versus dry rolling) will increase the net energy intake and glucose supply and increase marbling

### The role of intramuscular fat in meat palatability

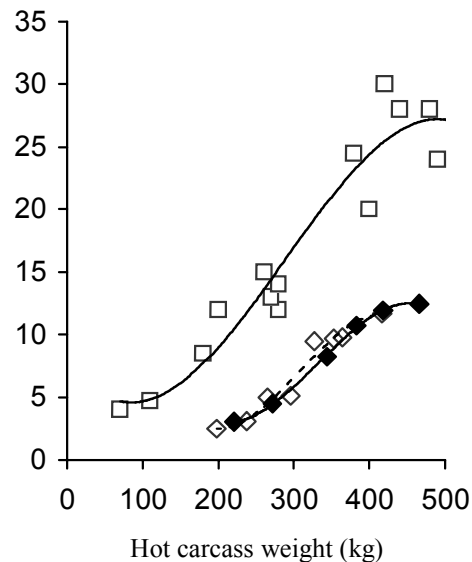
Despite all the hype, world literature suggests that marbling only explains 10 to 15% of the variance in palatability. Meat Standards Australia (MSA) research basically agrees and shows that the contribution of marbling to palatability was significant and important, but just one of many factors determining final palatability. However it has been suggested that as variations in tenderness are controlled by schemes such as MSA, marbling will become a more important determinant of palatability due to its specific contribution to juiciness and flavour.

There is concern that very low levels of intramuscular fat will lead to meat that is dry and less tasty. Such a situation has been found in young highly muscled lean cattle (e.g. double-muscled cattle genotypes, young bulls from Belgian Blue or Blonde d'Aquitaine) and in many cuts from modern pig genotypes. The minimum requirement for intramuscular fat in order to achieve acceptable consumer satisfaction for grilling 'red meat' cuts (beef and lamb) is quoted at 3-4% on a fresh uncooked basis.

### Development of intramuscular fat - Growth and development

Adipose tissue is deposited in specific depots primarily in the abdominal cavity (perirenal, mesenteric and omental) and intermuscular, subcutaneous and intramuscular sites. However the proportions differ between the species and are influenced by age. Intramuscular fat represents about 5-10% of the total fat in the live animal and so it is of a moderate to small size.

**Figure 1.** The relationship between carcass weight and intramuscular fat content of the striploin of American Angus x Hereford (◇), Australian Angus (◆, work by Alex Pugh, Beef CRC) and Japanese Black x Holstein cross cattle (□, Japanese work).



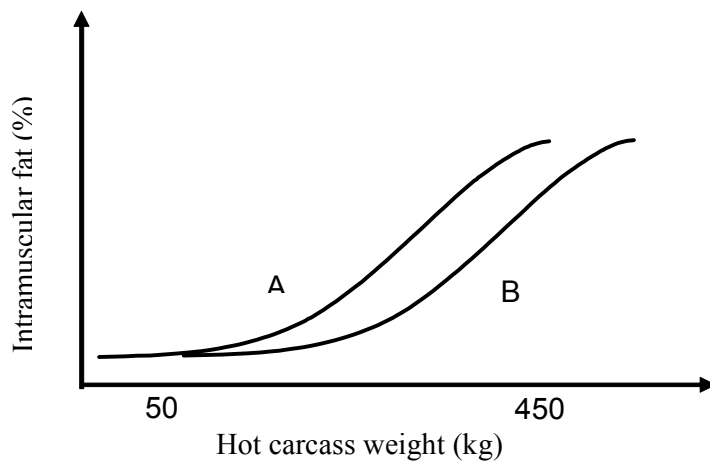
The development of intramuscular fat in beef cattle is shown in Figure 1. Based on this data the Beef CRC can hypothesise four drivers of intramuscular fat development (i) the potential for total carcass fat deposition (ii) the potential for muscle growth (iii) mature body size and finally (iv) the extent of fat distribution bias for intramuscular fat versus other carcass depots. Importantly these principles suggest that genetic selection (via traditional quantitative genetics or via gene marker assisted selection) for increased intramuscular fat alone could come about via alterations in any of the drivers described above.

Selection for high levels of muscularity is known to reduce both total carcass fatness and intramuscular fat at a given carcass weight. Recent work in sheep has shown that lambs produced from sires with a high estimated breeding value for post weaning eye muscle depth (using the Sheep Genetics Australia system) produce substantially leaner carcasses with reduced intramuscular fat when compared to lambs from sires with an elevated estimated breeding value for post weaning growth rate.

Therefore the Beef CRC propose that the developmental curve for intramuscular fat will be shifted to the right in animals with a high propensity to grow muscle or with a greater mature body size (and in these cases with the same propensity to marble at maturity, Figure 2). This 'right shift' would also occur in response to metabolic modifiers such as hormonal growth promotants,  $\beta$  agonists and organic chromium supplementation, all of which can increase muscle growth.

In very highly muscled animals it might be that intramuscular fat does not reach the 'linear accumulation' phase discussed in Figure 1 within normal commercial slaughter weights (that is the right shift described in Figure 2 is profound). This would appear to be the case for the modern pig genotypes where intramuscular fat % does not increase over a wide range of commercial carcass weights.

**Figure 2.** Hypothetical graph showing the development of intramuscular fat in cattle of different mature body weight and/or muscle development ( $B > A$ ).



The initial or 'starting' intramuscular fat content at  $\leq 200$ kg carcass weight is likely driven by the genetic predisposition for development of fat cells at the intramuscular site relative to other depots. Importantly there is a proportional developmental difference that is maintained when the American or Australian cattle are compared to the Japanese Black cross cattle such that the starting (2 vs. 4%) and final (13 vs. 27%) intramuscular fat contents are proportionally different at about 2 fold (Figure 1).

This suggests that the potential for development of fat cells is fixed relatively early in life and there after changes in either size and/or number of cells occurs in proportion to the initial cell number. This would clearly indicate that a measure of intramuscular fat content (perhaps by non invasive methods such as ultrasound) would be a good predictor of subsequent marbling performance.

#### **Development of intramuscular fat - Metabolic Understanding**

Recently some gene expression work has been undertaken by the Beef CRC using both Japanese and Australian cattle and the results confirm that developmental changes related to marbling occur quite early in life. Thus by 11 months of age Japanese Black cattle had a significantly greater expression of genes associated with intramuscular fat compared to other beef breeds and the key time of maximal difference in gene expression was between three and eight months of age (before the end of weaning).

Related work has established that marbling adipocytes show a preference for glucose carbon while subcutaneous adipose tissue uses mainly acetate as a source of acetyl units for lipogenesis. This may offer an opportunity to study the role of specific nutritional and hormonal intervention work in young (pre-weaning) cattle to 'set up marbling' for the finishing phase.

Another feature associated with the development of intramuscular fat is the muscle fibre type or metabolic pattern of energy use expressed by the muscle tissue. Within the one animal genotype the 'fast twitch' or white muscle types (e.g. eye round) marble less strongly. Across genotypes a similar response can be found.

In a Beef CRC study in collaboration with French scientists where two muscle types were contrasted across three breeds of cattle with disparate propensity to accumulate intramuscular fat, there was a strong correlation between intramuscular fat and 'slow twitch' or red muscle fibres.

#### **Nutritional modulation of intramuscular fat - Manipulating protein and energy**

Nutritional manipulation of intramuscular fat levels in pork via altering the dietary protein:energy ratio has been clearly shown in a number of studies. Of course the basic premise is that by restricting muscle development through a subtle protein deficiency, total carcass fatness will be increased sufficiently to elevate intramuscular fat.

The results of manipulating the protein:energy ratio in beef cattle diets is less conclusive. The conclusions from two studies Beef CRC were that diets which contain more or less protein than

recommended amounts for feedlot animals do not lead to significant differences in marbling or intramuscular fat. However, there was a trend for high protein diets to produce less and low protein diets more marbling than control diets in both experiments. Certainly the data suggests that a simple diet based on cereal grain and hay ( $\approx 10\text{-}11\%$  crude protein, with no additional protein source in the form of grain legumes or urea) fed to Angus steers at a starting live weight of 540 kg (P8 back fat = 12 mm) produced equal performance to more traditionally formulated rations containing additional protein sources at an extra cost.

\*\*\*\*\* **FIG 7** \*\*\*\*\*

### **Nutritional modulation of intramuscular fat - Fermentation pattern in ruminants**

It has been assumed for sometime that diets which promote both: (i) maximal fermentation in the rumen to produce glucose precursors (propionate), and (ii) which maximise starch digestion in the small intestine (to produce free glucose) might increase intramuscular fat deposition. Such diets are usually associated with high levels of processing which increase the accessibility of the dietary starch granule to both microbial and animal amylases and so maximise the availability of glucose to the fattening animal.

The logic behind this hypothesis was that (i) such diets would promote increased levels of anabolic hormones (insulin) which are known to stimulate lipogenesis; (ii) the logic parallels the observation in humans that diets with a high glycaemic index (i.e. diets that allow rapid glucose absorption and concomitant high insulin levels) promote obesity; (iii) Such diets will also deliver increased levels of net energy for lipogenesis (the reason why grain feeding promotes more intramuscular fat development compared with grass finishing and (iv) there is evidence that marbling adipocytes show a preference for glucose/lactate carbon while subcutaneous adipose tissue uses mainly acetate as a source of acetyl units for lipogenesis (see above).

### **Conclusions**

Intramuscular fat is clearly an important but not exclusive determinant of consumer response to beef. A major determinant of intramuscular fat content is the potential for mature size and muscle growth. Thus animals which display high muscle growth either have reduced expression or show no development of intramuscular during the so called fattening phase. In this scenario genetic approaches for increasing intramuscular fat will need to focus on changing fat distribution (toward intramuscular fat) if the potential for muscle growth is to be maintained.

Our knowledge of fat development at the intramuscular site is now being underpinned by new metabolic and genomic characterization of pathways for adipogenesis and lipogenesis and the hope is that this might allow for some nutritional management of marbling.

Nutritional manipulation of intramuscular fat independently from total carcass or whole body fat depots has proved difficult to achieve. However there is now a growing body of evidence that intramuscular adipocytes are metabolically different to at other depots (subcutaneous fat) and further research is needed to see if this offers the possibility for targeted stimulation of fat development within muscle.

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