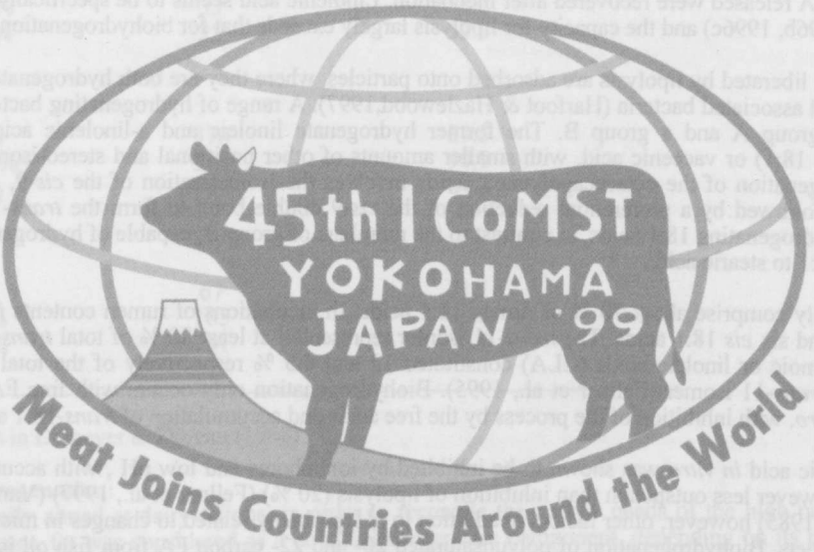
SESSION 2

MEAT PRODUCTION



2-L1

LIPID COMPOSITION OF BEEF IN RELATION TO FEEDING, NUTRITION AND TECHNOLOGY.

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Abstract:
Factors affecting rumen lipid metabolism, involving lipolysis, hydrogenation and fatty acid synthesis are reviewed, as well as technology to prevent dietary lipids from interaction with rumen micro-organisms. The interactions of fat feeding with breed and conformation effects on adipose and muscle tissue lipids is discussed, in relation to lipid content, lipid class and fatty acid composition. Special attention is given to the Belgian Blue double muscled breed. Finally, an integrated approach to the limits for modification of ruminant fat is proposed.

Introduction:
For many years, consumers in industrialized countries have been displaying an aversion to dietary fat (Claus, 1991) and, for health reasons, nutritional guidelines advocate a decrease in the consumption of (saturated) fat (Gormley et al., 1987). Nevertheless, total fat intake in Western-Europe continues to amount to about 40 % of dietary energy intake, equivalent to about 130 g/d in the UK (Mansbridge & Blake, 1997), Belgium (Demeyer et al., 1995) and Germany (Flachowski & Jahreis, 1997). Beef is estimated to contribute about 5 % of total fat intake only, but is often regarded as a rich source of saturated fat (Demeyer et al., 1995). For both social and ecological reasons, an intervention by the farmer rather than by the food technologist, to make beef more attractive for consumption as healthy food may be indicated. Several (bio)technologies supply the means for such interventions, aimed at target processes situated at both digestion and/or tissue metabolism (Mersmann, 1990). The use of feed additives and growth promotors is often involved, clashing with the strengthening requirements of consumers and regulation for more quality of animal production and products. This paper will therefore mainly discuss effects of feeding fat supplements on milk and beef fat content and composition. It summarizes information from other recent reviews (Demeyer et al., 1995) (Doreau et al., 1997) (Doreau & Chilliard, 1997) (Wood & Enser, 1997) (Demeyer & Doreau, 1998) and emphasizes data obtained in Belgium, a country characterized by very intensive animal production and the use of extremely lean double muscled cattle.

Rumen metabolism of lipids:
Lipolysis. Triacylglycerols, phospholipids and galactosyl lipids in ingested forage and concentrate feeds are rapidly hydrolyzed in the rumen by extracellular lipases demonstrated in a limited number of bacteria. Some activity could be associated with the protozoal fraction (Harfoot & Hazlewood, 1997). No intermediate production of partial acylglycerols can be demonstrated and free FA form the major lipid fraction in rumen and duodenal lipids, even when high amounts of fat are fed. Lipolytic activity is clearly inhibited by pH values below 6 (Van Nevel & Demeyer, 1996a) and by antibiotics such as ionophores (Van Nevel et al., 1996b). In incubations of rumen contents with soya bean oil, 92 % of the FA released were recovered after incubation. Linolenic acid seems to be specifically released over linoleic acid (Van Nevel & Demeyer, 1996b, 1996c) and the capacity for lipolysis largely exceeds that for biohydrogenation (Immig et al., 1993).

Biohydrogenation. Free FA liberated by lipolysis are adsorbed onto particles, where they are both hydrogenated and/or incorporated into the lipid fraction of the solid associated bacteria (Harfoot & Hazlewood,1997). A range of hydrogenating bacteria have been isolated and can be partitioned into a group A and a group B. The former hydrogenate linoleic and a-linolenic acids mainly to the *trans*-11 octadecanoic acid (*trans*-11 18:1) or vaccenic acid, with smaller amounts of other positional and stereo-isomers of the same acid. The initial step in the biohydrogenation of the octacecapolyenoic acids involves the isomerization of the *cis*-9, *cis*-12 isomer to the *cis*-9, *trans*-11 isomer, which is followed by a preferential reduction of the *cis*-9 double bond to form the *trans*-11 18:1. Group A bacteria appear to be incapable of hydrogenating 18:1 acids, in contrast to the members of group B, capable of hydrogenating a wide range of both *cis*- and *trans*-isomers of 18:1 to stearic acid.

Trans 18:1 isomers normally comprise about 70 % of rumen 18:1 acids. In incubations of rumen contents *in vitro* they were found to contain up to seven *trans* and six *cis* 18:1 acids. The *trans*-11 isomer represented at least 80 % of total *trans*-18:1 acids, and *trans*-18:1 and conjugated octadecadienoic or linoleic acids (CLA) constituted 13 and 0.5 % respectively of the total FA. CLA were shown to contain 33 % of the *cis*-9,*trans*-11 isomer (Fellner et al., 1995). Biohydrogenation only occurs with free FA, but the system is easily overloaded, especially *in vitro*, with inhibition of the process by the free acids and accumulation of *trans*-18:1 and CLA.

Biohydrogenation of linoleic acid *in vitro* was shown to be inhibited by ionophores and low pH , with accumulation of *trans* 18:1 and CLA. Such inhibition is however less outspoken than inhibition of lipolysis (20 %) (Fellner et al., 1997) (Van Nevel & Demeyer, 1996). As shown by Gerson et al. (1985) however, other factors in addition to pH, probably related to changes in microbial populations, must be involved with concentrate diets. Biohydrogenation of polyunsaturated 20- and 22- carbon FA from fish oil is controversial. Ashes et al. (1992) noted the absence of biohydrogenation *in vitro*, in contrast to results obtained *in vivo* by Doreau & Chilliard (1997) and *in vitro* by Van Nevel et al. (1998).

Formation of microbial FA. It is now clear that both bacteria and protozoa can synthesize and/or incorporate higher FA, the synthesis possibly including, at least under some conditions, linoleic acid (review in Demeyer, 1973) (Demeyer & Hoozee, 1984). Linoleic acid is incorporated in polar lipids or free FA. The balance of synthesis and incorporation seems to depend on the availability of FA. When free FA are largely available, incorporation surpasses synthesis, whereas the opposite obviously pertains in the absence of added lipids (Demeyer et al., 1978) (Bauchart et al., 1990). For the animal, bacterial lipids are more important than protozoal lipids, as protozoa are largely retained in the rumen. The bacterial lipids are characterized by a large proportion of branched chain and uneven numbered straight carbon chain FA, synthesised from propionate and/or amino acids. The main lipid fractions are free FA and polar lipids and the fatty acid composition reflects incorporation of dietary FA, as well as biohydrogenation and bacterial synthesis. Although considerable variability is apparent from the data reported, it is clear that a mixture of liquid and particle associated bacteria, when leaving the rumen, contains

between 10 and 20 % lipid in the DM, its FA containing between 5 and 15 % *trans* 18:1 isomers, as well as 2 to 20 % PUFA, the higher values being obtained with diets comprising PUFA rich lipid supplements (Demeyer & Doreau, 1998). These amounts are not to be neglected: per kg DM intake, a diet of corn silage contains about 400 g OM apparently digested in the rumen, equivalent to the production of about 120 g of bacterial DM, representing about 12g (10 % in DM) newly synthesized lipid. As, apart from hydrogenation, all dietary lipid (58 g/kg DM) leaves the rumen mainly unaltered it can be estimated that duodenal lipids may contain up to 17 % bacterial lipids on such diets. Similar estimates were made earlier (Jenkins, 1994) (Demeyer & Van Nevel, 1995) (Mansbridge & Blake, 1997).

Rumen outflow of FA:
The rumen outflow of lipids to the duodenum occurs mainly as free FA, showing a high degree of saturation and containing considerable amounts of *trans*-isomers of mono- and di- octadecenoic acids. Compared to rumen digesta, duodenal (Kobayashi et al., 1992) and abomasal (Van Nevel & Demeyer, 1994) digesta are slightly enriched in PUFA's, a finding possibly related to the sloughing off of enterocytes.

Recovery of dietary intake. Comparison of duodenal flows of FA with fatty acid intake in dairy cattle indicate that between 6 and 10 g of bacterial FA are synthesized per kg of DM intake, a value comparable with 12g/kg DM intake as derived above. Between 20 and 30 % of dietary FA, mainly those shorter than 14 carbons, are degraded in the rumen beyond hydrogenation and incorporation or are absorbed from the rumen. (Demeyer & Doreau, 1998).

Biohydrogenation and trans acids. The extent of hydrogenation of 18 carbon PUFA's observed *in vivo* is high; higher for linolenic (50 - 80 %) than for linoleic (35 - 60 %) acids and is lower for high concentrate than for low concentrate diets as concluded from the litterature (Doreau et al., 1997). A striking feature of duodenal lipid composition, is its high content of *trans*-isomers, increasing with the completeness of rumen biohydrogenation of added unsaturated oils, this being also determined by the balance between biohydrogenation and outflow of oils. (Table 1). Contents of stereo- and positional isomers of C18:1 are reported, but it is clear that *cis-trans* conjugated dienoic acids must also occur in duodenal contents.

Indeed, Noble et al. (1974) have shown that sheep rumen micro-organisms produce *trans*-11 C18:1 together with *cis*-9, *trans*-11 18:2 *in vitro*. The *trans* 11-18:1 isomer makes up at least 80 % of the *trans* 18:1 isomers in accordance with the hydrogenation pathways established for rumen bacteria.

Table 1. *trans* 18:1 content of duodenal FA in relation to oil feeding and biohydrogenation.

Reference[1]	*trans* 18:1 (mg/g total FA)	Biohydrogenation %[1]
Wu et al. (1991)		
Diet + -	42	67.3
+ Ca salt Palm oil 3%	33	58.3
+ Ca salt palm oil 6%	25	53.0
Enjalbert et al. (1994)		
Diet + soya oil SO 5.7%	370	89.5
+ emulsified SO 5.7%	349	90.9
+ Ca salts SO 6.7%	115	64.3
Kalscheur et al. (1997)		
Diet + -	67	71.1
+ 18:1 sunfl.oil 3.7%	185	79.5
+ 18:2 sunfl.oil 3.7%	201	80.1

[1] References and comments in Demeyer & Doreau (1998)

Ways to decrease biohydrogenation:
The use of fat was originally aimed at dairy rations, in order to reconcile the energy needs of the high producing dairy cow with its capacity for feed intake. Later, fat was considered as a potential concentrate component, depending on its price compared to other raw materials (Doreau & Chilliard, 1997).
It was realized early on that dietary fat levels had to be limited to about 5 % of the diet, in order to prevent negative effects on rumen metabolism, resulting in milk fat depression and, also, lower performance of fattening animals (Buysse, 1962). The effects in the rumen mainly relate to an inhibitory effect on the fibre degrading flora and fauna, particularly protozoa, resulting in shifts in the rumen fermentation, reflected in lowered productions of methane, acetate and butyrate (Van Nevel & Demeyer, 1995). The finding that only FA in the free acid form exhibited an anti-microbial effect (see e.g. Demeyer & Henderickx, 1967) has resulted in the development of "by-pass", "protected" or, better, "rumen inert" fats, protecting lipids from rumen metabolism. The four main processes used are (1)

encapsulation of triglycerides in a matrix of aldehyde treated protein, (2) formation of calcium salts of FA, (3) pelleting of saturated fats or hydrogenated oils and small amounts of starch to form prilled fat supplements and (4) extrusion of vegetable oil seeds. The effectiveness of protection can be estimated from the extent of lipolysis and biohydrogenation when incubated with rumen contents which was estimated to amount to < 20, 45, 65 and 70 % respectively, compared to 95 % for an untreated oil (Gulati et al., 1997). Processing for protection of fat does not guarantee a standard degree of inertness however, as illustrated by the use of calcium soaps of unsaturated FA. Such soaps dissociate more rapidly than those of saturated FA at the same pH, and dissociation followed by hydrogenation becomes important at pH values < 6 (Ferlay et al., 1992) (Van Nevel & Demeyer, 1996a).
The formaldehyde treatment offers efficient protection, but because of the cost of treatment, the variability in treatment efficiency and the refusal by the US FDA to approve formaldehyde in animal feed, interest has shifted to other processing technology such as the use of heat treated oil seeds.

Digestibility:
Fatty acid digestibility can only be measured in a reliable manner between duodenum and ileum or faeces. A litterature survey revealed data ranging between 55 and 92 %. Variability was not explained by level of fatty acid intake or FA composition of the diet. Digestibility does not differ between 16 and 18 carbon atoms and appears to be lower for 20 and 22 carbon FA. Mean digestibilities for 18 carbon FA are 77, 85, 83 and 76 % for 0,1,2 and 3 double bonds respectively (Doreau & Chilliard, 1997).

Absorption and incorporation into adipose tisue lipids:
Transport. The epithelial cells are the site for esterification into triacylglycerols and phospholipids, transported into the lymph as chylomicrons and VLDL, and further into the blood where these lipoproteins are found together with LDL and HDL. LDL, VLDL and chylomicrons contain approximately 10-15, 60 or 85 % triacylglycerols respectively. HDL account for approximately 90 % of blood lipids and consist largely of phospholipids, cholesterol and cholesterol esters, in ruminants containing the major fraction of PUFA (Mansbridge & Blake, 1997). Lipoproteins transport FA mainly to the mammary gland in dairy cattle and mainly to adipose and muscle tissue in fattening animals.

Uptake by adipose tissue. During the the passage into the tissues, triacyl glycerols are largely or completely hydrolysed by a lipoprotein lipase. There is little further modification of preformed FA within these tissues, except for mild to extensive desaturation of medium- and long-chain FA in, particularly 18:0, to ensure fluidity of tissue and milk. Both, adipocytes and mammary gland tissue incorporate chylomicron derived FA into triacylglycerides, using glycerol formed from glucose. The specificity of FA distribution within the triglycerides is determined during the esterification of glycerol-3-phosphate with 2 FA, followed by the incorporation of a third FA into the dephosphorylated 1,2-diacyl-3-phosphate. The FA distribution in adipose triacylglycerols conforms to the general rules formulated by Brockerhoff (1966) for mammalian depot triglycerides i.e. saturated FA predominantly in the position sn-1, shorter chain and unsaturated FA largely in position sn-2 and C18 and longer chain FA in position sn-3. The branched chain FA are mainly found in position sn-2 whereas the *trans*-18:1 isomers are distributed in a different manner from the *cis*-isomers (sn-2 position) and are mainly found in the sn-3 position (Christie, 1981). Phospholipids are formed through reactions between diacylglycerols and cytidine phoshate components, involving specificity of the acyl*trans*ferases involved.

Turnover and the balance between FA synthesis and incorporation:
Biosynthesis of adipose tissue and milk lipids both, uses diet derived FA as well as FA synthesized *de novo* from acetate. The effect of the former on tissue FA composition is limited however as synthesis *de novo* and direct incorporation are regulated and balanced in relation to optimal fluidity of both adipocyte and milk lipids and membranes. Adipose tissue is the major site of fatty acid synthesis in ruminants except during lactation when the mammary gland becomes the predominant site. Synthesis of FA up to palmitic acid takes place in the cytoplasm, from acetyl-CoA and ß-hydroxybutyrate derived from mitochondrial oxidation. Mitochondria elongate palmitic acid to longer FA with up to 22 carbons, whereas microsomes are capable of elongation as well as desaturation of FA with 18 or more carbon atoms. Ruminants, as all mammals, and in contrast to plants are however incapable of the introduction of double bonds between carbon 9, counted from the carboxyl group and the methyl end of the chain (Gurr & James, 1980). Fatty acid CoA esters inhibit acetyl CoA carboxylase in adipose tissue and mammary gland.

As with all living tissues, amounts of adipocyte lipids are the net result of both synthesis and degradation. Turnover time of adipose tissue lipids varies with level of feeding, physical activity and lactation and may be up to hundreds of days. FA are liberated from adipose tissue for transportation as albumin bound non esterified FA (NEFA) to tissues in need for oxidative energy. Complex regulation of lipolytic activity occurs by both short and long term mechanisms, the former involving the action of a hormone sensitive lipase, activated by adrenalin and inhibited by insulin. An increased flux of circulating exogenous FA within adipocytes may decrease *de novo* FA synthesis from acetate. An increased PUFA flux may however stimulate the adrenergic lipolytic response. These two effects could explain why ruminants are less reponsive tot dietary fat for increased fatness than non ruminants (Doreau & Chilliard, 1997).

Preferential retention of PUFA:
Ruminants attempt to specifically retain the small amounts of PUFA that escape hydrogenation by preferentially incorporating them into plasma cholesterol esters via the lecithin cholesterol acyl transferase pathway. It is also clear that they selectively incorporate polyenoic acids into membrane phospholipids. The ruminant has however probably developed other mechanisms to preferentially retain PUFA's. Vernon & Flint (1988) suggested a limited oxidation of linoleic acid in ruminants, based on the finding that malonyl CoA is much more effective in inhibiting carnitine acyltransferase when linoleyl CoA is the substrate than when palmityl CoA is substrate in sheep liver, whereas the converse pertains in rat liver. Nevertheless, because of extensive rumen biohydrogenation, it has been suggested that ruminants can be delicately balanced in respect of their essential fatty acid status (Ashes et al., 1995).

Effect of fat feeding on adipose and muscle tissue lipids:
Nature and development of fat depots. In ruminants, lipids are mainly accumulated as triacylglycerols in adipocytes, located in subcutaneous, inter - and intramuscular adipose tissue, and abdominal, e.g. perirenal and omental fat depots. Total dissectable fat in a beef carcass varies roughly between 10 and 40 %, about half of which is subcutaneous and intermuscular fat. With increasing animal weight, the proportion of subcutaneous fat increases more than that of intermuscular tissue, i.e. it has a higher relative (allometric) growth rate.

Different regions of sucutaneous fat also grow at different rates and the rate of increase of intramuscular lipid is less than that of dissectable fat (Wood, 1984). Young fat tissue has a high proportion of water and connective tissue and a low proportion of lipid, contained in small cells. When the animal gets older, fat depots, especially internal depots, increase mainly through increase of cell size, with a decrease of the proportion of water and connective tissue in the tissue and a colour change from gray to more white. The fat cell diameter in adult animals may vary between 100 and 250 mm, meaning more than an eightfold difference in storage capacity (Lawrence & Fowler, 1997). Female animals deposit more fat than males and castration increases fat deposition so that there is a great interest to produce entire males, especially in southern and Western continental Europe.

Intramuscular fat. Triacylglycerols in intramuscular adipocytes, associated with the epi- and endomysium, determine the "marbling" of meat, which increases with the age of the animal. In muscle fibers and at higher concentrations in red than in white muscle, lipid is also present as intracellular lipid droplets. Besides triacylglycerols in adipocytes and in oil droplets, intramuscular lipid also comprises the membrane phospholipids of the muscle fibers. Whereas extraction of the former can be done using non polar solvents such as ether, extraction of total lipids, including membrane phospholipids, requires the use of more polar solvents such as chloroform/methanol. Mean values $\pm$ SE calculated from data reported by Christie (1981), Larick & Turner (1989), Ashes etal (1995) and Gandemer (1997) show that FA in phosphatidyl choline and phospatidyl ethanolamine make up 48.1 $\pm$ .7 % and 25.3 $\pm$ 2.0 respectively of total membrane phospholipid bound FA, 20 - 30 % in each fraction being recovered as plamologen analogues. It is claimed that total intramuscular lipid content is very important for sensory evaluation of beef,especially of texture. Meat preparation and/or cultural differences considerably affect sensory evaluation however, as minimal intramuscular fat contents for optimal sensory quality vary between 1.5 and 3 % for Denmark and the US respectively (Wood, 1990). With decreasing total intramuscular lipid content,the relative importance of phospholipids in total lipids increases. Indeed, as adipocyte size is lowered or less adipocytes are present, amounts of muscle fibre membranes remain relatively constant.

Breed and conformation effects on adipose tissue distribution. There are important breed differences in the proportions of body fat in the various depots. Breeds noted for beef production have a higher ratio of dissectable/intramuscular fat than breeds noted for milk production, as shown in Table 2, also illustrating the low intramuscular fat content of double muscled Belgian Blue cattle compared to Holstein cattle.

Furthermore, the ratio subcutaneous fat/ dissectable fat increases with meat yield. For the same degree of subjective EU carcass fatness classification (e.g.2), based to a large extent on visual inspection of the subcutaneous fat cover, the carcass dissectable fat content, estimated from dissection of a one-rib cut, decreased with increasing conformation from 20.0 to 12.3 %. This suggests that the relative importance of subcutaneous tissue in total adipose tissue increases with increasing muscularity (Van de Voorde et al., 1997).

Fatty acid composition. Beef lipid composition, as well as milk fat, reflects rumen metabolism of dietary lipids. Proportions of "unusual" odd-chain, branched chain and *trans* monoenoic acids of 3.3 , 2.7 and 4.2 moles % respectively have been reported. The multi-branched FA pristanic and phytanic acids are present, whereas the *trans* 18:1 series is dominated by the *trans*-11 isomer (Christie, 1981).

Table 2. Carcass and intramuscular fat content of Holstein, Double-muscled (DM) and mixed (M) Belgian Blue (BB) cattle, fed different diets[1].

Reference	% dissectable fat in carcass	% intramusc. fat in Longissimus Thoracis[a]
DM BB bulls ca. 700 kg		
High energy	13.1	.93
Low energy	12.3	.86
Bulls ca. 500 - 550 kg		
Holstein	25.2	4.64
Mixed BB	24.2	2.20
DM BB	11.6	.75
DM BB bulls ca. 600 kg		
control	13.4	.68
+ milk fat	13.7	.88
+ veg.fat	14.9	1.12
DM BB cows ca. 700 kg	12.6	1.12

[1] From Demeyer & Doreau (1998)

Beef meat and beef tallow display slightly higher *trans* 10 + *trans* 11 18:1 relative proportions ($\pm$ 65 % of total *trans* 18:1 isomers which represent 4.9 $\pm$ 0.9 % of total FA), compared to butter fat (55 - 60 %), the opposite being found for 13 and 16 isomers. This finding would indicate that some subtle differences exist in the metabolic selectivity for individual *trans* 18:1 acids between the mammary gland and muscles or adipose tissue (Bayard & Wolff, 1996). In earlier work, the same group reported higher *trans* 18:1 contents in total FA for beef tallow (4.6 %) than for meat (1.95 %). Intramuscular *trans* fatty 18:1 was concentrated in triacylglycerol FA (2.53 %), and much less in polar lipid FA (0.76%) (Wolff, 1995). Saturated and unsaturated aldehydes, possibly metabolically derived from FA, have been detected in adipose tissue from ruminants and some may be important in flavor determination of meat and meat products (Christie, 1981). Although it is generally recognized that beef fat has a more saturated nature than pork and poultry fat, because of the biohydrogenation processes in the rumen, there are e.g. clear indications of specific deposition and/or metabolism (desaturation) of absorbed FA in the

various triacylglycerol depots. As in all large animals, internal fat depots in mature animals are characterized by high proportions of saturated FA. Saturation, characterized by the 18:0/18:1 ratio, decreases with decreasing distance from the animal's exterior. Effects of temperature differences between these sites on desaturase activity are held partly reponsible for this finding. In contrast to pigs, the concentration of saturated acids in fat depots decreases with age and body fat content in cattle (Wood, 1984). This finding seems to be limited to domesticated cattle however as adipose tissue triacylglycerols of lean African wild ruminants were found to contain higher proportions of unsaturated FA, possibly because of dietary differences (Sinclair & O'Dea, 1990). Effects of breed are illustrated by the more saturated character of Hereford cow subcutaneous adipose tissue compared to Brahman cows (Huerta-Leidenz et al., 1993). The effect of site is illustrated for double muscled animals in Table 3. The data in Table 3 also illustrate that the triglyceride fraction of intramuscular lipids shows a fatty acid composition close to that of subcutaneous fat. The phospholipid fraction on the other hand is probably fairly representative of the membrane structures and is characterized by high levels of PUFA, particularly linoleic and arachidonic acids. In fact, skeletal muscle appears to be a major repository for these essential FA in ruminants (Christie, 1981). Concentrations of total FA in the M.longissimus thoracis increased with increasing carcass fat content ($r = 0.41$), the increase being limited to the TAG fraction, a finding also illustrating the separate nature of the polar lipid fraction. The concentration of FA in the latter amounts to 3.1 mg/g wet tissue, a value similar to those reported by Larick & Turner (1988) and Gandemer (1997). The PUFA content in the polar lipids (32.6 %) is similar to recent data reported for beef (21.5 - 38.7 %), as well as for pork (29.9 - 38.7 %) (Demeyer et al., 1995).

Because of the low content of total intramuscular lipid, its fatty acid composition is determined to a large extent by that of the polar lipid fraction and is similar to that of pork intramuscular lipids, characterized by high and low contents of PUFA's and *trans* acids respectively, an important finding from a nutritional point of view. In summary, dissectable fat of lean animals is more saturated than that of fat animals, whereas the opposite is true for intramuscular fat.

Effects of fat feeding. Unlike milk production, lean beef production is not limited by energy intake. Fats may however be cheaper alternatives in feed formulation and their use allows modification of beef fatty acid composition, providing other incentives for the increasing practice of fat supplementation in beef production. From a litterature survey, Clinquart et al. (1995) conclude that, in general, fat addition to a finishing diet results in improved weight gain at lower intakes. At incorporation levels higher than 5% however, growth may be impaired, possibly related to impairments of both digestion and intake. Whatever the nature of the fat, an improved feed conversion, associated with a 5 to 15 % higher fat content of the carcasses, is generally observed. The authors cite two non published experiments from their laboratory however, in which decreases in carcass fat content of up to 28 % were observed. This finding might be explained by inhibitions of *de novo* fatty acid synthesis as reported with dairy cattle. From their own results,summarized in Table 4, it is clear that unsaturated oils provoke effects comparable to those observed in milk fat: a decrease in SMCFA content, and an increase in LCFA, mainly 18:1. These effects are less outspoken in intramuscular lipids and with heat treated beans vs. free oils. With free oil, 18:2 is preferentially retained in intramuscular lipids, again reflecting the separate metabolic importance of muscle polar lipids. Heat treatment of beans seems to protect 18:2 from biohydrogenation, as discussed earlier. As with dairy cattle, evidence is available suggesting that fat supplementation induced effects beyond changes in the proportion of dietary and newly synthesized FA as substrates for fat synthesis. Van Eenaeme et al. (1991) have shown e.g. that fat supplementation increased plasma insulin levels with reduction of blood glucose levels.

Table 3. Fatty acid composition of muscle and adipose tissue lipids of Double-Muscled Belgian Blue Cows (Webb et al.,1998).

FA	Intram. Fat[a]		Interm. fat		Perir. fat	Subc.fat
(mg/g)	PL (3.1)[b]	TAG (8.1)[b]	Ser.	Transv.		
14:0	2.5	21.5	25.2	21.7	22.1	26.2
16:0	146.1	271.9	259.8	254.3	237.1	274.1
16:1	28.7	33.5	28.6	18.8	14.4	39.9
18:0	125.8	161.2	180.8	257.4	308.2	127.9
18:1	191.1	431.0	421.7	366.7	340.2	448.0
18:2	231.0	24.4	23.3	24.9	24.1	22.1
18:3	35.7	6.3	7.3	7.9	7.1	7.1
20:4	58.8	0.0	-	-	-	-
other	180.3	50.2	53.4	48.2	46.9	54.6

[a] Intram. = intramuscular; interm. =intermuscular; perir. = perirenal; subc. = subcutaneous; ser. = musculus serratus; transv. = musculus transversalis; PL = polar lipids; TAG = Triacylglycerols; FA are represented as number of carbons: number of double bonds. [b] () = mg of FA/g wet tissue.

Table 4. Percentage change in fatty acid composition of perirenal and intramuscular (Longissimus thoracis) fat by dietary addition of soya oil (3.3 %) or heat treated soya beans (15 %) (Clinquart et al., 1995).

FA (mg/g)	Soybean oil		Heat treated soybeans	
	Perirenal	Intramusc.	Perirenal	Intramusc.
SMCFA	- 21.2	- 10.8	- 3.8	- 1.5
18:0	+ 2.0	+ 3.8	- 2.2	- 2.2
18:1	+ 34.1	+ 2.3	+ 5.5	+ 2.1
18:2	- 8.7	+ 7.1	+ 4.5	+ 3.8

It is more difficult to justify dietary interventions to change fatty acid composition of beef fat than for milk fat. Indeed, beef dissectable fat is not consumed directly and used as tallow in food and non - food industries. Furthermore, intramuscular fatty acid composition was shown to display a favourable PUFA content with very lean animals (Webb et al.,1998). The further development of lean animals with low intramuscular fat content seems to deserve more support than dietary interventions. Management of feeding may also contribute to a better partitioning of fat deposition however. Feeding animals at low intake levels, followed by realimentation, increases the proportion of subcutaneous lipid, at the expense of intramuscular lipid (Hornick et al., 1998). However, as it seems that the very low intramuscular lipid levels of about 1 % may impair sensory quality, a higher content of intramuscular triacylglycerols, enriched in PUFA through dietary means may be a goal to pursue further. Also, in the intramuscular lipid of very lean animals, an improvement of the *n-3/n-6* PUFA ratio may be indicated. In contrast to milk, where very low transfer efficiencies of dietary *n-3* fish oil FA were observed (Mansbridge & Blake, 1997), sheep muscle intramuscular lipids specifically retained 20:5 and 22:6 *n-3* PUFA, added in the diet as a rumen inert fish oil preparation (Ashes et al., 1992). The *n-3/n-6* ratio was increased from a control value of 0.20 to 0.54. It should be stressed however that such changes are observed in the phospholipid fraction only, whereas no *n-3* PUFA were retained in the triacylglycerols, in contrast to pigs (Wood & Enser, 1997). In more detailed work, Ashes et al. (1995) discuss the specific substitution of 20:5 for 20:4 in the phosphatidyl inositol of membrane phospholipids. They refer to the associated changes to be expected on the nature and effects of PI derived secundary messenger molecules. In a more general approach, these include possible changes in gene expression (Jump et al., 1996) and immune reactions (Miles & Calder, 1998), mediated trough altered prostaglandins and leukotrienes, when muscle membrane PUFA are modied.

Limits to the modification of ruminant fat:

Processing for fat protection may not be acceptable by consumers or governments, as illustrated by the refusal to accept formaldehyde treatment, in spite of extensive experimentation (McDonald & Scott, 1977). The same argument may hold for newer technologies, involving the preparation of fatty acyl amides (Jenkins & Thies, 1997). The further use of feed antibiotics, (also) resulting in modulation of fat digestion is, at least for the EU, out of the question. A good example is Monensin, shown to increase duodenal flow of unsaturated FA (Kobayashi et al., 1992). A definite ban of its use could be supported by the finding that it is absorbed from the intestinal tract and elicits a series of metabolic effects in the animal when infused intravenously (Armstrong & Spiers, 1988). Consumer and governmental aversion is even greater against the use of production modifiers active at the animal tissue level. Also, they may adversely affect sensory meat quality (Fiems et al., 1990). A proposal for immunomodulation of adipocyte development (Flint et al., 1994) should certainly be subjected to social acceptance.

The limitation on the use of tallows in the UK because of the risk of transmission of factors linked to bovine spongiform encephalopathy (BSE) is perhaps the best example of the care that should be taken when proposing dietary or other interventions. The use of supplements should be evaluated for environmental effects related to e.g. methanogenesis (Dong et al., 1997) whereas the role of pasture vs. concentrate feeding in providing more acceptable unsaturation should perhaps receive more attention in relation to the presence of PUFA derived flavor compounds as discussed for dairy products (Urbach,1990), related also to the need for extensivation. It is clear that a constant concern relates to the greater susceptibility to oxidation and possible flavor defects of meat products enriched in PUFA (Gandemer, 1997). In analogy with an excellent paper, dealing with milk production but that should inspire the whole of animal production (Fredeen, 1996), modification of beef fat composition via nutrition should not be approached simply from the point of view of economic advantage. Environmental, welfare and moral considerations should also be considered, analogous to a general quality concept for animal production (Demeyer, 1997). Similar considerations should indeed apply for further selection and breeding of double-muscled cattle (Demeyer et al., 1995a).

Relations between classic nutritional strategies mainly involving increase of the concentrate/forage ratio and beef fat composition should be more precisely defined for application in practice. Such relations are indeed complex and do not allow precise predictions of the effects of a dietary change, also because of animal variability. As proposed for the dairy cow, the use of mechanistic models covering complete nutrient utilisation may improve precision (Sutton, 1988) whereas variability may be reduced if the use of cloned cattle is accepted. Finally, it should be realized that simple feeding practices may go a long way in optimizing beef fatty acid composition. From data presented by Wood & Enser (1997), *n-3/n-6* ratio's for PUFA in intramuscular lipids of 0.12 to 0.77 can be calculated for concentrate and grass fed cattle respectively.

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