# INFLUENCE OF β-HYDROXY-β-METHYL BUTYRATE ON PERFORMANCE, CARCASS QUALITY, LIPID DEPOSITION AND TENDERNESS OF LONGISSIMUS MUSCLES FROM SERIALLY SLAUGHTERED FEEDLOT STEERS

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"Graduate school is like an endurance race, you don't run across the finish line, you fall over it!"

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#### Format of Dissertation

This dissertation is presented in the Journal of Animal Science style format, as outlined by the Oklahoma State University graduate college style manual. The use of this format allows for independent chapters to be prepared suitable for submission to scientific journals. Three papers have been prepared from the data collected for research to partly fulfill the requirements for the Ph.D. degree. Each paper is complete in itself with an abstract, introduction, materials and methods, results and discussion, implications and literature cited section.

#### CHAPTER I

#### **GENERAL INTRODUCTION**

Ruminants are a diverse group of animals that consist of many different species scattered across a variety of different climatic and vegetative zones. Ruminants vary in color, shape and size; ranging from the mouse deer (300 mm tall weighing between 2 and 5 kg) to the giraffe (3.5 m tall weighing 2 metric tons; Church, 1988). Ruminants in general have played an important role in human agriculture, being hunted for 750,000 years and domesticated for 8,500 years (Church, 1988).

Ruminants can eat low quality forage and convert it into high quality products such as meat, milk and fiber; this unique ability sets ruminants above other animals because 13.6 billion acres of the earth's land mass is more suited for grazing than for cultivation. Thus, if ruminants did not possess this unique ability, the competition for food resources that sustain both animals and humans would be more vigorous.

The US cattle industry is one of this country's oldest industries; it has obtained this lofty position by adapting to meet consumer demands. The end product of the cattle industry is beef with approximately 24 million head of cattle slaughtered each year. The ability of the cattle industry to make progressive and responsive changes in such a large number of animals is an amazing accomplishment. Recently, consumers have become more aware of nutritional value in their diets. They have begun to reduce their intake of foods that are presumed to be rich in saturated fat and cholesterol. This, in part, has caused the per capita beef consumption to increase only 7% in the past 20 years. During this time period, per capita consumption of chicken and turkey has increased 72% and 62%, respectively. Thus, the beef industry must search for ways to produce beef that will satisfy consumer demands.

Even though consumers may want leaner beef with less trimmable s.c. fat, most consumers still prefer a moderate degree of intramuscular fat (marbling); this contributes to tenderness, taste, juiciness and eating acceptability of beef. Another important factor, which has been studied for years but has only recently become a top priority is improving the consistency in the tenderness of beef products. To meet consumer demands, one should increase or maintain the intramuscular fat while decreasing the amount of s.c. fat deposited, lower the cholesterol content of beef tissue, and improve the consistency in tenderness of beef products.

## CHAPTER II REVIEW OF LITERATURE

#### **EFFECT OF MUSCLE MYOFIBERS ON CARCASS QUALITY**

Characterization of Myofibers. Muscle and its composition, has been studied for centuries. Cassens and Cooper (1969) indicated that the earliest work in muscle histochemistry (by Lorenzini in 1678) described muscles based upon color. Subsequently, Ranvier in 1874 showed that muscles with increased redness were associated with slower contractions. Ashmore et al. (1972) summarized that two types of primary fibers exist,  $\alpha$  and  $\beta$ , that differ in their myosin ATPase activity, speed of contraction and metabolism. Myosin ATPase activity is low in the  $\beta$  fiber but high in the  $\alpha$  fiber. Contraction speed is slower in  $\beta$  than in  $\alpha$  fibers. The  $\beta$  fibers exhibit the highest succinic dehydrogenase/glycogen phosphorylase activity ratio; therefore, they are adapted to aerobic metabolism. In contrast,  $\alpha$  fibers exhibit a low succinic dehydrogenase/glycogen phosphorylase activity ratio; therefore, they are adapted for anaerobic metabolism. Ashmore et al. (1972) stated that  $\beta$  fibers can be consistently classified as "red" fibers; whereas,  $\alpha$  fibers could be classified as "red", "white", or "intermediate" fibers, based on their enzyme patterns. Thus, these authors re-designated fiber types as:  $\beta$  fibers as  $\beta$  Red  $(\beta R)$  and the  $\alpha$  fibers as  $\alpha$ -Red ( $\alpha R$ ) or intermediate, and  $\alpha$  White ( $\alpha W$ ).

In 1965, Beecher et al. conducted the first comprehensive study in pigs using the red and white muscle fiber concept with direct application to meat science. This was followed by an effort to understand the properties of meat based on histochemistry. Muscle biology researchers have become convinced that the myofibril composition of meat is of great importance, stating that "the

properties of muscle, be they visual appearance, physiological parameters, or biochemical characteristics, are a reflection of the proportions of types of myofibers present." Data obtained from extracts or homogenates of whole muscle" whether they be enzyme activity, compositional information, or characteristics of specific proteins, are merely a reflection of the proportion of red and white myofibers present." (Cassens, 1977).

#### Antemortem Effects on Myofiber Distribution and Diameter

Gender. Many antemortem factors can influence the distribution and diameter of muscle myofibers. Animal gender can affect the proportion of different muscle fibers as well as the diameter of the fibers. Dreyer et al. (1977) reported that meat from bulls had a higher percentage of BR muscle fibers and a lower percentage of  $\alpha W$  muscle fibers as compared to meat from steers. Similarly, steers have a higher percentage of  $\beta R$  fibers and fewer  $\alpha W$  fibers than heifers (Johnston et al., 1981). However, West (1974) reported that heifers had a lower percentage of  $\alpha$ W fibers than steers. Drever et al.(1977) and West (1974) also reported that bulls had larger fiber diameters of all three fiber types than steers, while heifers had smaller fiber diameters than steers. Although the gender of an animal appears to alter the ratio of muscle fibers, it is not believed to be the major factor involved in the influence gender has on meat tenderness. Generally, meat from bulls is less tender than the meat from steers (Field, 1971; Seideman et al., 1982). This difference has been attributed primarily to differences in fiber diameter and intramuscular collagen content (Dreyer et al., 1977; Cross et al., 1984).

**Breed.** Different breeds of cattle have been used advantageously in production systems; ranchers and feedlots in warmer climates utilize the heat resistant characteristics of *Bos Indicus* cattle. However, *Bos Indicus* cattle have lower meat tenderness.

Wheeler et al. (1990a) reported that LM steaks taken from Brahman steers were less tender than that of purebred Hereford steers after aging for either 7, 14, 21, 28 or 35 d postmortem. Differences with aging were believed to be caused by an increase in calcium-dependent protease (CDP)-I activity on d 0 together with a concurrent decrease in CDP inhibitor. In their study, no differences were found in CDP-II or cathepsin B or B+L activities. Using electrophoresis data, the authors concluded that meat from Hereford steers had a more advanced state of proteolysis as early as 1 d postmortem, and that the inherent difference between breeds in CDP activity and inhibitor play a major role in tenderness. This work is supported by the findings of Shackelford et al. (1991); shear force was greater in 5/8 Brahman versus Angus x Hereford steers. In contrast to Wheeler et al. (1990a), Shackelford did not find any changes in CDP-I activity; however, they found greater CDP inhibitor activity with 5/8 Brahman steers but no difference between breeds in cathepsins B and B+L activity. Shackelford et al. (1991) concluded that the decreased tenderness in the 5/8 Brahman steers was due to the increased CDP inhibitor activity. Although the meat from Brahman cattle is less tender than meat from European breeds, this problem may be alleviated postmortem by electrical stimulation and blade tenderization (Wheeler et al., 1990b).

Piedmontese cattle were introduced into the United States because of superior muscularity and leanness. Some Piedmontese have a unique characteristic called "double-muscling" or muscular hypertrophy. Increased muscularity is believed to come from a 46% increase in the number of muscle

fibers; most of this increase is in  $\alpha$ W fibers (Holmes and Ashmore, 1972). Tatum et al. (1990) compared carcass composition and quality of steers sired by Piedmontese, Gelbvieh and Red Angus bulls. In their study, steers sired by Piedmontese bulls had carcasses with less s.c. fat, greater longissimus muscle (LM) area, lower numerical yield grades, and the higher muscle to bone ratios. These steers also had LM with the highest percentage of  $\alpha$ W fibers, and the lowest percent of  $\alpha R$  fibers, plus the smallest cross-sectional area of  $\beta R$  fibers. Concentrations of total and soluble collagen were not different between these breeds. Tenderness, measured by Warner-Bratzler shear force, as well as myofibrillar tenderness and palatability attributes for Piedmontese steers were greater than for Gelbvieh steers, but not different than for Red Angus steers. The increased percentage of  $\alpha W$  fibers most likely is responsible for the enhanced muscularity and palatability attributes, while the smaller crosssectional area of the  $\beta R$  fibers may be responsible for the improved tenderness. In general, Piedmontese steers possessed a LM with a larger percent of  $\alpha W$ fibers and smaller cross-sectional area of BR fibers; hence, they and produced larger amounts of lean meat with desirable eating attributes.

**Days on Feed.** As time on feed increases, tenderness often increases (Dolezal et al., 1982; Miller et al., 1987); however, this increase may be limited in time to a maximum of 139 d (Epley et al., 1968) or between 150 and 180 d (Zinn et al., 1970b); after this time, animal age may exert a greater influence and decrease tenderness. May et al. (1992) fed steers between 0 and 196 d and reported that shear force values were lowest at 112 d; shear force values at 28 and 196 d both were greater than at 112 d. Steers that have been fed a high concentrate diet for 120 d have more of their collagen in the soluble fraction when compared to steers coming directly off from grass (Wu et al., 1981). This

may explain why cattle fed for approximately 140 d have more tender meat than cattle short-fed off grass or fed for an extremely long time. In general, the work by Johnston et al. (1981) demonstrated that as the energy level of the diet was increased,  $\alpha$ R fibers were replaced by  $\alpha$ W fibers resulting in an increase in the average fiber diameter. Johnston et al. (1975) also reported that mean fiber diameter of  $\beta$ R fibers in steers was greater for those fed 233 versus 153 d. Marsh (1977) reported that the intermolecular cross links within collagen increase with animal age; they become more thermally resistant which results in less collagen breakdown during cooking. Together with factors that increase the fiber diameter, e.g., fiber transformation due to dietary energy levels or increased days on feed, collagen solubility may play a role in the decrease in tenderness as animals age.

#### Fiber Type and Diameter in Relation to Tenderness

Brady (1937) studied correlations, between fiber diameter and shear force; he found these correlations to be low but to increase when the muscle was cooked. He reported a high negative correlation (r = -0.81) between the number of fibers in a muscle bundle and mechanical shear force when considering with four different bovine muscles. Later Tuma et al. (1962) studied cattle ranging in age from 6 to 90 months. Steaks from these animals were aged at 2<sup>o</sup> C for either 48 h or 14 d. Correlations, uncorrected for animal age, were significant across both aging periods, implying that as fiber diameter increased, shear force increased. Herring et al. (1965) also found a high correlation between fiber diameter and tenderness (r = .73). When animal age was included by Tuma et al. (1962), correlations between fiber diameter and shear force existed when steaks were aged for 48 h but for steaks aged for 14 d, these correlations disappeared. This would agree with the work of Crouse et al. (1991) who

reported that fiber diameter affected shear force in steaks aged for 1 or 3 d, but not in steaks aged between 6 and 14 d. The research by Tuma et al. (1962) and Crouse et al. (1991) suggests that prior to steak aging, which is believed to alter postmortem proteolysis, fiber diameter can alter tenderness.

Calkins et al. (1981) reported that in "A" maturity carcasses, the percent  $\alpha$ R fiber area was negatively correlated (r = -.46; P<.05) to shear force while the ratio of  $\alpha W: \alpha R$  was positively correlated (r = .43; P<.05) to shear force. Hence, with the transformation from  $\alpha R$  to  $\alpha W$  occurs in less active muscle during their maturation, shear force increases (Ashmore et al. 1972). In the same study, averaged across carcass maturities from A to E, shear force was related positively to the percentage of  $\alpha W$  fibers (r = .35; P<.01) and the percent  $\alpha W$ area (r = .31; P<.05) but negatively to the percent  $\alpha R$  area (r = -.25; P<.05). Thus, red fibers appear more highly correlated with carcass quality attributes than white fibers. This supports the work of Crouse et al. (1991), who also reported that the percent  $\alpha W$  fiber area was significantly correlated to shear force (r = .60) when steaks were aged for 1 d; however, this was not true for steaks aged between 3 and 14 d. This suggests that as the percentage of  $\alpha W$ fibers increase, shear force increases if the muscle is not aged. Although  $\alpha W$ fibers generally are larger than  $\beta R$  fibers, as the percentage of white fibers increases the average diameter of the fibers will increase, thus causing shear force to increase.

Romans et al. (1965) reported low fiber diameter and tenderness correlations, for steaks aged for 10 d. Melton et al. (1974) worked with muscle biopsies, for which no aging would have occurred; they found that  $\beta$ R fiber area was correlated significantly with live weight (r = .59), hot carcass weight (r = .64), marbling (r = .49), quality grade (r = .54), s.c. fat thickness (r = .75) and yield grade (r = .67), but  $\beta$ R fiber area was not correlated with shear force or taste

panel tenderness. Intermediate ( $\alpha$ R) and  $\alpha$ W fibers were correlated significantly to taste panel flavor score (r = .45),but were not significantly related to tenderness. When these data were analyzed with the  $\beta$ R and  $\alpha$ R fibers being combined, most correlation coefficients decreased; however, when  $\alpha$ R and  $\alpha$ W fibers were combined, those same correlation coefficients increased. Thus, the  $\alpha$ R fibers appeared to be more closely related to the  $\alpha$ W fibers than  $\beta$ R fibers. This work is similar to a later study reported by Melton et al. (1975) were muscle biopsies once again were obtained; fiber type as a percentage had no relation to shear force, but taste panel palatability scores tended to decrease as the percentage of  $\beta$ R fibers increased. Similar to his work in 1974, he found that  $\alpha$ W fibers were positively correlated with taste panel attributes.

Lewis et al. (1977) found no correlation between fiber diameter and shear force or taste panel tenderness when examining LM aged for 7 d; however, when the Psoas major (PM; considered to be predominately  $\beta$ R) was aged for 7 d and examined the uncooked fiber diameter was correlated to shear force (r = .37; P,.01) and taste panel tenderness (r = -.24; P,.05). The effect of aging was expected to be less dramatic on the PM than the LM, because  $\beta$ R fibers have a much thicker Z line which makes it less labile to postmortem enzymes (Goll et al., 1970, 1974). They also reported that even though breed had an effect on fiber diameter, breed had little effect on tenderness. Therefore, Lewis et al. (1977) suggested that cattle should be selected for larger fiber diameters and increased muscling and that such selection would not affect tenderness. This may be true, as long as muscle is aged prior to cooking.

In summary, fiber type and diameter or area play some role in meat tenderness. An increase in fiber diameter has been correlated with an increased shear force prior to postmortem aging. The negative correlations between  $\alpha W$  fibers and tenderness may be a result of the  $\alpha W$  fibers being larger than the  $\beta R$ ; thus, fiber diameter again may be related to tenderness.

#### Fat Deposition and Myofibers

Generally, "red" fiber types metabolize and store more lipid than "white" fiber. George and Jyoti, (1955) first reported histochemical data indicating that red muscle fibers contain more lipid than do white muscle fibers. More recently, Calkins et al. (1981) reported similar findings within A maturity carcasses, in which both the percentage of  $\alpha W$  fibers (r = -.46; P<.05) and the percent  $\alpha W$ area (r = -.45; P<.05) were correlated negatively with marbling. In contrast, the percent  $\alpha R$  (r = .45; P<.05) area was correlated positively to marbling. Melton et al. (1974) reported that correlation coefficients between the area of  $\beta R$  fibers and all measures of fat deposition, both s.c. and intramuscularly were positive when bulls fatten, the  $\beta R$  fiber increases in size;  $\alpha R$  and  $\alpha W$  fibers were not related to fat deposition. Melton et al. (1975) also reported that as vascularity (capillaries per fiber) of a tissue increases, the amount of lipid deposition also increased. Ashmore et al. (1972) reported that  $\beta R$  fibers have a higher capillary to fiber ratio than  $\alpha W$  fibers do; increased blood flow allows for greater aerobic metabolism with less fatigue. Hence, lipid deposition within a muscle, such as the LM, would be greater if more  $\beta R$  fibers are present. Because the LM is predominately composed of  $\alpha$ W fibers which, generally are not associated with lipid deposition, lipid deposition in the LM may be a poor index of carcass quality.

#### **Differences in Collagen Content Between Myofibers**

Data concerning the amount of connective tissue present with different myofibers is limited. Beatty (1966, 1967) used histochemical staining procedures and a biochemical method to determine hydroxyproline content, one index of collagen. In primates, white muscles generally contain higher concentrations of connective tissue. However, the similarity between white muscles of adult primates and A maturity bovine carcasses could be questioned.

#### Effects of Aging on Muscle fiber types

Generally, postmortem aging increases the tenderness of muscles due to postmortem proteolysis by several enzymes. However, muscle fiber types respond differently to aging. Lewis et al. (1977) found no correlation between fiber diameter and shear force or taste panel tenderness when examining the LM (predominately  $\alpha$ W fibers) which had been aged for 7 d. However, the Psoas major (PM; considered to be predominately  $\beta R$ ) was aged equally for 7 d and examined; the uncooked fiber diameter was positively correlated to shear force (r = .37; P < .01) and correlated negatively to taste panel tenderness (r = .24;P<.05). This suggests that the two muscles did not age at equal rates. The effect of postmortem aging would be expected to be less significant on the PM as compared to the LM, because  $\beta R$  fibers have Z lines which are 2 to 3 times thicker than  $\alpha W$  fibers; the  $\alpha W$  fibers are less labile to postmortem enzymes (Goll et al., 1970, 1974). Crouse et al. (1991) reported that average fiber size (proportionally based) and tenderness were correlated when steaks were aged for 3 and 6 d but only percent  $\alpha$ W area was correlated with tenderness after 1 d of postmortem aging. In summary,  $\alpha W$  fibers are degraded faster than  $\beta R$  fibers during postmortem aging; this is believed to be due to thinner Z lines.

# Effects of $\beta$ -Adrenergic Agonist and Somatotropin on Myofibers and Tenderness

Consumer demands for lean products has led to the investigation of compounds both of which increase protein accretion and decrease fat synthesis such as  $\beta$ -adrenergic agonists and somatotropin (Moseley et al., 1990; Wheeler and Koohmaraie, 1992; Lanna et al., 1992). These compounds have increased production of lean meat in cattle (Carroll et al., 1990; Fabry and Sommer, 1990; Moseley et al., 1990), sheep (Hamby et al., 1986), swine (Jones et al., 1985) and poultry (Gwartney et al., 1992).

Although they increase the amount of lean product,  $\beta$ -adrenergic agonists and somatotropin, reduce tenderness. An increased force is required to shear muscle from cattle (Fabry et al., 1990; Vestergaard et al., 1990; Wheeler and Koohmaraie, 1992), sheep (Hamby et al., 1986; Pringle et al., 1991), swine (Jones et al., 1985; Solomon et al., 1991; Warriss et al., 1991, Chang et al., 1992;) and poultry (Morgan et al., 1989; Gwartney et al., 1992). In both cattle (Vestergaard et al., 1990) and swine (Aalhus et al., 1992) administration of  $\beta$ adrenergic agonists alter the myofiber distribution, decreasing the percentage of  $\alpha$ R and increasing the percent of  $\alpha$ W. This transformation from  $\alpha$ R to  $\alpha$ W is similar to that described by Ashmore et al. (1972). The shift from "red" to "white" fibers also may explain the decrease in intramuscular fat deposition. These changes have caused dramatic increases in shear force with no increase in the amount of total or soluble hydroxyproline (Aalhus et al., 1992). Concurrent with the increased percentage of  $\alpha$ R and  $\alpha$ W fibers came an increase in diameter of these fibers.

Administration of  $\beta$ -adrenergic agonists also stimulate a transformation between fiber types and may reduce the action of proteolytic enzymes such as calpains and cathepsins due to increased calpastatin (Morgan et al., 1989; Pringle et al., 1991; Wheeler and Koohmaraie, 1992). The combination of increased fiber diameter and reduced postmortem proteolytic activity may explain the decrease in tenderness.

Alterations in the size and distribution of muscle fibers associated with the use of porcine somatotropin (pST) have been documented by Solomon et al. (1990 ;1991). In both studies, they administered pST to boars, barrows and gilts. The pST decreased the percentage of  $\alpha$ W fibers while both  $\alpha$ R and  $\beta$ R muscle fibers tended to increase. Even more pronounced than the shift in fiber distribution was the increase in fiber area; this was significant for all three different fiber types. Thus, the increased muscularity observed with the administration of pST came primarily from an increase in fiber diameter. This increase in fiber diameter concurrently increased shear force in barrows, but less consistently in boars and gilts.

The  $\beta$ -adrenergic agonists and somatotropin cause opposite shifts in fiber type distribution, with  $\beta$ -adrenergic agonists increasing the percentage of  $\alpha$ W fibers and somatotropins increasing the percentage of  $\alpha$ R and  $\beta$ R fibers. However, both increase fiber diameter leading to enhanced muscularity and decreased tenderness.

#### **General Description of Metabolism and Biochemical Pathways**

The three BCAA, leucine (LEU), isoleucine and valine are among the nine amino acids that cannot be synthesized by animal tissues and must be supplied in the diet (Harper et al., 1984). The initial step in the metabolism of the amino acid leucine is a reversible transamination reaction; it produces the ketoacid,  $\alpha$ ketoisocaproate (KIC). This transamination reaction is catalyzed by branchedchain aminotransferase (BCAT) which is general for all BCAA, or by leucine transaminase, which acts solely on LEU (Ichihara, 1975). Subsequent to transamination, the ketoacid, KIC, is irreversibly decarboxylated primarily (approximately 90%) by branched-chain  $\alpha$ -ketoacid dehydrogenase (BCKAD); BCKAD is located in the mitochondria and produces isovaleryl CoA (Wohlhueter and Harper, 1970). Isovaleryl CoA is sequentially converted in the mitochondria to  $\beta$ -methylcrotonyl CoA,  $\beta$ -methylglutaconyl CoA and finally to  $\beta$ -hydroxy- $\beta$ methylglutaryl CoA (HMG CoA). Hydroxymethylglutaryl-CoA lyase cleaves HMG CoA to acetoacetate and acetyl CoA. Acetoacetate can cross the mitochondrial membrane and can be converted into acetone and β-hydroxybutyrate or acetoacetyl CoA and  $\beta$ -hydroxy- $\beta$ -methylglutaryl CoA (HMG CoA) (Rawn, 1989). In the cytosol, HMG CoA can be converted to mevalonic acid by HMG CoA reductase; this is the first committed step in cholesterol synthesis.

The remaining KIC (approximately 10%) is metabolized by  $\alpha$ ketoisocaproate oxygenase (KIC oxygenase) is located in the cytosol. During this reaction, KIC is decarboxylated and hydroxylated to form  $\beta$ -hydroxy- $\beta$ methylbutyrate (HMB) (Sabourin and Bieber, 1981b, 1982a). The fate of HMB is not well established, but in both lambs and pigs approximately 34% of an exogenous dose of HMB is excreted via the urine; thus, HMB is presumably metabolized by the body (Van Koevering and Nissen, 1992).

In summary, KIC can be metabolized both in the cytosol and in the mitochondria by the mitochondrial BCKAD or KIC oxygenase. Both enzymes release carbon dioxide during decarboxylation. The oxidative decarboxylation of KIC by mitochondrial BCKAD produces the branched-chain acyl CoA derivative, isovaleryl CoA as an end product (Rawn, 1989). In the rat and human liver, KIC oxygenase produces  $\beta$ -hydroxyisovalerate, otherwise known as  $\beta$ -hydroxy- $\beta$ -methylbutyrate or HMB, as the major end product (Sabourin and Bieber 1981a, 1982a, 1983). HMB also has been isolated from skeletal muscle, heart, diaphragm and kidney of the rat (Wagenmakers et al., 1984; Hokland and Bremer, 1988) and from the urine of humans, rats, lambs and pigs (Tanaka et al., 1968; Landaas, 1974, 1975; Finnie et al., 1976; Van Koevering and Nissen, 1992). Thus, KIC oxygenase activity may not be limited to the liver.

#### Subcellular Distribution of Branched-Chain Amino Transferase

Although BCAT activity occurs both in the cytosol and in the mitochondria, the proportion of activity in each fraction varies between organs and species. In swine, sheep, cattle and rats, BCAT activity is primarily cytosolic in skeletal muscle and adipose tissue (Busboom et al., 1984a). However, BCAT activity in the liver and kidney is cytosolic in swine but mitochondrial in sheep, cattle and rats (Busboom et al., 1984a). Within the rat, BCAT activity occurs solely in the mitochondria of heart and kidney, whereas cytosolic activity is predominant in the brain (70%) and the liver (100%) (Hutson, 1988; Hutson et al., 1988).

Hutson (1988) found that rat skeletal muscle varies in the subcellular location of BCAT. In the soleus muscle, considered to be a predominately red muscle (89%  $\beta$ R fibers and 11%  $\alpha$ R fibers), BCAT activity was completely in the mitochondria. The plantaris muscle, composed of a mixture of fibers (53%  $\alpha$ W fibers, 40%  $\alpha$ R fibers, and 7%  $\beta$ R fibers), had 65% of the BCAT activity in the

mitochondrial fraction. In the white gastrocnemius muscle (91%  $\alpha$ W fibers and 9%  $\alpha$ R fibers), 70% of the BCAT activity was accounted for in the cytosol (Hutson, 1988). The differences in BCAT distribution between muscle fiber types could be a result simply of metabolic preference. Red fibers ( $\beta$ R), as mentioned previously, have an oxidative type of metabolism. These fibers typically have more mitochondria; this would enhance the development of BCAT activity in the mitochondria. In contrast, the  $\alpha$ W fibers have primarily a glycolytic type of metabolism and have fewer mitochondria to support BCAT activity in the cytosolic fraction.

Increased BCAT activity in the cytosolic fraction of muscles consisting predominately of  $\alpha$ W fibers, would suggest that increased amounts of KIC are produced in the cytosol of these muscles as compared to muscles with predominantly  $\beta$ R fibers. In light of the fact that KIC concentrations in the cytosol most likely are the rate limiting factor in HMB synthesis, due to the higher Km of KIC oxygenase (~10 X) than of mitochondrial BCKAD (Sabourin and Bieber, 1979), muscles that consist of predominately  $\alpha$ W fibers may have greater concentrations of KIC present in the cytosol; this could lead to increased synthesis of HMB.

HMB synthesis in muscle consisting predominately of  $\beta$ R fibers could only occur if KIC, which was produced in the mitochondria, was transported across the inner mitochondrial membrane to the cytosol were it could be oxidized by KIC oxygenase. Patel et al. (1980) in rat liver demonstrated, that KIC must be transported actively from the cytosol across the inner mitochondria membrane; even at elevated concentrations, passive diffusion does not occur. This work was supported by May et al. (1980) who observed that addition of carnitine to rat liver homogenates stimulated the oxidation of KIC and increased the export of acylcarnitines from the mitochondria. This supports the theory that KIC must be

transported from the cytosol across the inner mitochondrial membrane and that KIC can not move freely; however, this transportation is believed to proceed from the cytosol into the mitochondria only and not vice versa. Transportation of KIC out of the mitochondria is unlikely due to the location of BCKAD, on the inner surface of the inner mitochondrial membrane, and BCKAD's great affinity for KIC (Km of .02 mM; Harper et al., 1984; Paxton and Harris, 1982). Thus, if KIC Oxygenase activity exists in the cytosolic fraction of peripheral tissues, which likely is due to HMB isolation in the skeletal muscle, heart, diaphragm and kidney of the rat (Wagenmakers et al., 1984; Hokland and Bremer, 1988), and in the cytosolic fraction of the liver (Sabourin and Bieber, 1982a, 1982b), then muscles consisting predominately of  $\alpha$ W fibers should produce the majority of HMB.

#### Effects of $\alpha$ -Ketoisocaproate in Ruminants

#### Growth and Performance:

Dietary supplementation of beef cattle with KIC has been studied in three experiments. In the first experiment (Flakoll et al., 1987), KIC was administered to 72 steers in a dose titration design with 0, 0.02, 0.07, or 0.20% of diet DM being KIC. These cattle were fed a moderate energy diet consisting of 75% concentrate and 25% roughage. During the first 56 days, KIC supplemented at .02% produced the greatest increase in growth rate (20%, P<.07), and improvement (13%, P<.05) in feed conversion. After 161 days on feed, cattle fed KIC at .02% had made 14% greater growth (P<.01) with an 8% improvement in feed conversion .

The second experiment was conducted using 156 steers (232 kg), and KIC was fed at 0, 0.005, 0.02, or 0.07% of the diet. Simultaneously, a third study was conducted with 76 steers initially weighing 258 kg; it was designed to evaluate the effects of KIC together with implants and ionophores. In both of these studies, steers were fed a growing ration (30% corn silage and 70% concentrate) for 90 d and a high energy finishing diet (7% corn silage and 93% concentrate) for the duration of the 192 d study (Van Koevering et al., 1989b).

Cattle performed well in both experiments 2 and 3; however, there were no significant responses in growth or feed conversion regardless of the treatment (Van Koevering et al., 1989b). Why the same effects in the first experiment were not observed in the second and third experiment is not known. Tischler and Goldberg (1980) proposed that in vitro, KIC may decrease pyruvate utilization and spare glucose. When a moderate energy diet was fed, the glucose sparing effect of KIC may have decreased the catabolism of amino acids for energy, allowing more amino acids to be utilized for growth. In the second and third experiments, a higher energy diet was fed, so the glucose sparing effect of KIC on amino acid catabolism would not have been as useful for growth.

#### Alterations in Carcass Composition:

Effects of KIC on carcass composition have been consistent. Repeatedly, administration of KIC to feedlot cattle has increased the amount of intramuscular fat deposited within the LM. An increased percentage of cattle have graded choice, and more fat is present in LM even though less cholesterol is present in the LM (Van Koevering et al., 1989a, 1989b). Carcasses from steers fed KIC had less external fat (fewer yield grades of 4 or greater) and tended to have a thinner s.c. fat layer.

#### **Possible Mechanisms for Alterations in Tissue Composition**

An increase in intramuscular lipogenesis together with a decrease in s.c. fat and cholesterol deposition seem to be contradictory effects. Separate mechanisms must be employed to explain each phenomena. Whether these mechanisms are intricately related is unknown, but they appear to originate simultaneously from the same compound. The combination of these mechanisms may explain the biological responses to HMB feeding.

Alterations in Marbling. KIC can decrease proteolysis without increasing protein synthesis and decrease the oxidation of glucose (Tischler and Goldberg, 1980). This demonstrates the ability of KIC, or possibly HMB, to direct energy away from protein and glucose metabolism and toward fat synthesis. Thus, HMB may act on peripheral tissues to decrease proteolysis and glucose oxidation. This allows muscle (predominately  $\beta R$  fibers) to direct more energy into fat deposition as marbling.

Alterations in Subcutaneous Fat. The mechanism behind the decreased s.c. fat is elusive. In ruminant adipose tissue, BCAT activity is 7 fold greater in the cytosol than in the mitochondria; hence, the majority of the KIC produced in adipose tissue is in the cytosol (Busboom et al., 1984a, 1984b). The activity of BCKAD, the enzyme which catabolizes KIC, is very low in ruminant adipose tissue and may be rate limiting. Thus, KIC may accumulate in the cytosol and stimulate KIC Oxygenase activity to produce HMB. In turn, when HMB is fed, this exogenous source may inhibit adipose tissue which may decrease fat synthesis.

Alterations in Cholesterol. Cholesterol biosynthesis occurs in the cytosol of the cell; HMB is the only metabolite of KIC that exists in the cytosol. HMB has direct access to the compartment of the cell to competitively inhibit  $\beta$ hydroxy-β-methyl glutaryl Coenzyme A (HMG CoA) reductase, the committed step enzyme in cholesterol synthesis. The similarity in structure between HMB and HMG, differing only by the presence of an extra carboxyl group on HMG, gives support to the theory of competitive inhibition. Volumes of data exit about HMG CoA reductase inhibition. Several compounds competitively inhibit HMG CoA reductase. These include compactin and ML-236B, isolated fungal metabolites, and lavastatin, pravastatin, fluindostatin, BMY-21950 and BMY-22089. These synthetic and non-synthetic agents that have been shown to competitively inhibit HMG CoA reductase (Brown et al., 1978; Parker et al., 1990). Whether the mechanism of cholesterol reduction by HMB is similar to such compounds is not yet known, but the current evidence strongly supports the theory that competitive inhibition of HMG CoA reductase can decrease cholesterol synthesis and concentration in both blood and tissue

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# EFFECTS OF $\beta$ -HYDROXY- $\beta$ -METHYL BUTYRATE ON PERFORMANCE AND CARCASS QUALITY OF FEEDLOT STEERS

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### ABSTRACT

Beta-hydroxy- $\beta$ -methyl butyrate (HMB), a compound formed in vivo during catabolism of leucine, was fed to 256 crossbred steers as 0 or .03% of diet dry matter. Effects on performance, carcass characteristics and tissue composition were measured. Groups of 32 steers per diet were slaughtered after 105, 119, 133 and 147 days on feed. HMB was fed only during the final 82 days on feed. Averaged across slaughter date, animal performance was not altered by HMB; however, an interaction between HMB and time on feed was detected. Daily gain was greater (P<.01) for steers fed HMB when slaughtered at 105 d but depressed (P<.01) by HMB when steers were fed for 147 d. Steers fed HMB had numerically higher marbling scores resulting generally in higher carcass quality grades. Steers receiving HMB had less (P<.08) s.c. fat and fewer steers with yield grades of 4 or greater (1.56 vs 4.69%). Supplementation of HMB to feedlot steers tended to increase (P<.07) the intramuscular fat to subcutaneous fat ratio. Steers fed HMB had higher (P<.001) plasma concentrations of HMB (3.06 vs 1.70 mg/l) and lower (P<.03) concentrations of cholesterol (108.4 vs 118.7 mg/dl). Total lipid content within the longissimus muscles (LM) tended (P<.10) to be greater for steers fed HMB when slaughtered after 105 d on feed.

(Key Words: Feedlot Steers, Marbling, Fat.)

### Introduction

To improve productivity of cattle, producers in the U.S. have utilized various anabolic implants to improve rate and efficiency of growth. Over 90% of the cattle slaughtered annually in the US in 1989 were treated with an anabolic implant (NCA, 1989). However, anabolic implants depress marbling score and the percentage of cattle with U.S. Choice quality grades (Foutz et al., 1990; Trenkle, 1990; Wagner et al., 1990). Current trends in US beef consumption favor products containing less fat, particularly trimmable waste fat; in contrast, moderate or high amounts of intramuscular fat or marbling have been related to improved taste and tenderness.

HMB, a compound produced in vivo during catabolism of the amino acid leucine, is in an experimental stage and has not yet been approved for commercial use by the U.S. Food and Drug Administration. In lambs and pigs, HMB was produced directly from the oxidation of either leucine or its keto acid,  $\alpha$ -ketoisocaproate (KIC; Van Koevering and Nissen ,1992). KIC feeding did not alter plasma levels of KIC but increased plasma concentrations of HMB suggesting that HMB is an intermediate and active metabolite.

Carcasses from feedlot cattle fed KIC have had increased marbling scores, more U.S. Choice quality grades, and less excess fat (yield grades of 4 or greater; Van Koevering et al., 1989a, 1989b) than carcasses from control cattle. When steers were fed KIC, cholesterol concentration (mg/100g of wet tissue) in the longissimus muscle (LM) was reduced (P<.08) even though intramuscular fat deposition was increased (P<.07; Van Koevering et al., 1989b).

Because HMB is formed during oxidation of KIC the biological alterations observed from KIC may be the result of its metabolite, HMB. Based on this theory, a serial slaughter feedlot study was designed to determine the effect of dietary HMB on performance, carcass quality, and tissue composition of feedlot steers.

### **Materials and Methods**

Animals and Diets. Two hundred and fifty-six crossbred steers (329 kg) were selected from a larger group (n=570) based on uniformity in size, weight and breed-type. Steers visually appraised as having greater than 25% Bos Indicus or Angus characteristics were removed, leaving steers of primarily British x Continental breed-type. This was done to reduce variability in marbling scores. Steers were processed by routine feedlot practices and implanted with an estrogenic implant (24 mg estradiol; Compudose®) at a commercial feedlot prior to arrival at Panhandle State University in Goodwell, OK. Upon arrival, individual steers were weighed, identified, and blocked into groups based on initial weight. Steers were assigned randomly to pens and allotted to treatments in a serial slaughter 2 x 4 factorial arrangement. One-half the pens (16) of cattle received the basal diet (controls); the other 16 pens of cattle received the same diet supplemented with HMB at .03% of the diet dry matter. Sets of four pens were assigned to be fed for a different number of days; these steers were fed for either 105, 119, 133 or 147 d after arrival. HMB was fed for only the final 82 d of each feeding period; HMB was removed from the diet 5 d prior to slaughter, the withdrawal time specified by the U.S. Food and Drug Administration for this experimental compound.

Steers were given ad libitum access to their high concentrate diets for the entire feeding period. Cottonseed hulls and chopped alfalfa, used as roughage sources, were removed stepwise from the diet to adapt cattle to their final diet. Diet compositions and analyses are shown in Table 1. Steers were receiving their final ration by day 19 of the study.

Cattle were weighed initially directly off the truck; these weights were used for allocation. Weight gain and feed efficiency were calculated based on initial shrunk weight and final live weights which were calculated from hot carcass weight /.6495, the mean dressing percentage for all cattle. Net energy values were calculated for each treatment using the 1977 yearling steer equation as reported by Hays et al. 1986. Cattle were trucked to Dodge City, Kansas for slaughter. At slaughter, livers were examined for the presence and severity of abscesses.

*Plasma Sampling.* Plasma was obtained 16 h post-feeding on the last day that HMB was fed to each of the respective slaughter groups. Plasma was collected in non-silicone coated Na<sub>2</sub>EDTA tubes and stored at -20<sup>o</sup>C until analyzed. HMB and KIC concentrations were determined using the method described by Nissen et al. (1990). Cholesterol concentrations were determined using Sigma Kit # 352 (Sigma Chemical, St. Louis, MO.).

*Carcass Data and Longissimus Muscle Sampling.* Carcass data for all slaughter groups were obtained approximately 48 hr postmortem; yield and quality grades were determined (USDA, 1989). A 20 cm thick section of the LM corresponding to the 9 through 12th rib section was removed from the left side of each carcass; it was vacuum packaged, and shipped to the Oklahoma State University Meat Laboratory. LM sections were aged at 2°C for 14 d postmortem. Ribeye samples subsequently were frozen (-30°C) and faced (uneven portion removed from the posterior end) before being fabricated into steaks for

determining composition. A 1.3 cm thick LM steak was removed from the posterior end of each LM section; it was denuded of exterior fat and epimysial connective tissue, and stored for proximate analysis. Immediately anterior to the steak used for proximate analysis, the remaining LM was cut into 2.5 cm thick steaks to be used for cholesterol analysis.

Longissimus Muscle Chemical Analysis. Samples were prepared in duplicate for chemical analysis by immersing them in liquid nitrogen and powdering them in a Waring Commercial Blendor® (Model 34B122; Waring, New Hartford, CN). A frozen 3 g sample of powdered LM was subjected to proximate analysis according to procedures outlined by AOAC (1984). Cholesterol content was determined through a modification of Lepage and Roy (1986).

Data Analysis. Data were analyzed on a pen basis using least squares analysis (SAS, 1988) with a linear model that included the main effects of HMB presence (df = 1), weight block (df = 3), slaughter date (df = 3) and all two way interactions. Least squares means were calculated and these were compared using T-tests. Three steers were removed from the data set for reasons not related to dietary treatments.

### **Results and Discussion**

Steer Performance. Main effects of HMB feeding on animal performance are shown in Table 2. Averaged across days on feed, animal performance was not altered by treatment; however controls tended (P<.10) to have heavier final live weights. Significant treatment by slaughter date interactions were detected for final live weight, daily gain, feed intake, feed efficiency and calculated net energy content of the diet indicating that slaughter groups had reacted differently to HMB feeding. These interactions were examined further (Table 3).

For steers fed a total of 105 d, those fed HMB had higher (P<.01) daily gains, feed intakes (P<.10) and improved feed efficiencies (P<.10) when final live weight was calculated from carcass weight (Table 3). However, for steers fed a total of 147 d, those that received HMB the final 82 d (day 60 to 142) had lower daily gains (P<.01), higher feed intakes (P<.10) and poorer (P<.01) feed efficiencies (carcass adjusted basis) than control steers. Note that steers allotted to the HMB treatment in the 147 d period also had 11% lower (P<.05) daily gains during the pre-HMB period; this indicated that steers allocated to HMB were lower even before HMB was fed. Steers fed HMB and slaughtered at 105 d, gained 8% faster during the pre-HMB period. When Pre-HMB values were included as a covariate in the statistical analysis of overall ADG, the main effect of HMB was not significant, but the HMB X days fed interaction was still significant. Improved ADG and feed efficiency of steers fed HMB for 105 d were similar to results of Flakoll et al. (1987) for feedlot steers fed KIC, the precursor of HMB. Flakoll et al. (1987) reported that ADG and feed/gain ratios were improved (20% and 13% respectively) during the first 58 d of a 161 d trial, with an overall increase in ADG of 14%.

Calculated net energy values were not significantly altered by HMB when averaged across days on feed (Table 2), but calculated net energy values were lower (P<.01) for steers fed HMB and slaughtered after 147 days on feed (Table 3); this reflects a poorer feed efficiency. The interaction between HMB with slaughter date is difficult to explain. Perhaps effects of HMB may differ with animal age or carcass fatness.

*Carcass Traits.* The main effect of HMB averaged across days on feed are presented in Table 4. Carcass weight was lower (P<.05) for HMB steers (Table 4); however, again an interaction of slaughter date and HMB was detected (P<.01). Carcass data for treatment by slaughter date subclasses are

presented in Table 5. Among steers fed for 105 d, those fed HMB had heavier (P<.05) carcass weights, whereas for steers fed 147 d, those fed HMB had lighter (P<.01) carcass weights. These effects match the treatment effects on live animal performance. Averaged across slaughter dates (Table 4), carcasses from steers supplemented with HMB had less thick (P<.08) s.c. fat, this effect was consistent for all slaughter dates as illustrated in Figure 1. When analyzed within slaughter dates for steers fed 147 d, those fed HMB had less (P<.10) s.c. fat. Consumers are demanding beef cuts with less s.c. fat; this has resulted in large surpluses of trimmed beef fat. Feeding HMB to feedlot steers may help decrease the amount of s.c. fat present on beef carcasses, and reduce the amount of external fat that needs to be trimmed from carcasses.

Figure 2 illustrates the effects of HMB on the percentage of carcasses in each yield grade for each slaughter group. Carcasses from steers fed HMB tended to have more (31% vs 26%) yield grade 1's (YG1) and fewer yield grades of 4 or greater (YG4). For steers fed 147 d, those fed HMB tended to have a greater (47 vs 31; P<.10) number of carcasses with yield grades of 2 (YG2) and fewer (16 % vs 3%; P<.05) YG4 (Table 6). Fat thickness has a large influence on yield grades; the decrease (P<.10; Table 5) in s.c. fat thickness with steers fed HMB during the 147 d slaughter group may be responsible for the increase in YG2 and decrease in YG4 carcasses. Within a given U.S. quality grade (Table 7), carcasses from steers fed HMB had more YG1 and fewer YG4 than control carcasses. This upward shift in yield grades further demonstrates that steers fed HMB had leaner carcasses than controls.

Ribeye area was not altered by HMB, except for the 147 d slaughter group in which steers fed HMB had smaller (P<.05; Table 5) ribeye areas than controls. This may reflect our concern that smaller framed, earlier maturing animals happened to be assigned randomly to HMB. The treatment by slaughter

group interaction was not significant for the percentage of condemned livers; however, animal groups with higher daily gains tended to have a higher percentage of condemned livers (r=.31; P<.09). No differences due to HMB were detected in dressing percentage, percentage of kidney, pelvic and heart fat, overall maturity or average USDA yield grade (Table 4).

Marbling scores were numerically higher (432 vs 422) for steers fed HMB (Table 8). This resulted in fewer (P<.01) HMB cattle receiving the quality grades of U.S. Standard (1.6 vs 6.0%; Table 8). Cattle receiving HMB tended to have a greater number of U.S. Select quality grades (42 vs 34%; P<.10). There were no significant differences in the percentage of steers with U.S. Choice or U.S. Prime quality grades. When quality grades were divided into thirds (Table 8), steers supplemented with HMB were more prevalent in the upper one-third of the Select (15.6 vs 8.6%; P<.09) and Choice (8.6 vs 5.5%) quality grades. When the high Choice and low Prime steers were combined into one category classified as "High Quality", HMB fed steers were more prevalent than controls within this category (9.5 vs 5.5). Table 9 illustrates the marbling scores and quality grades for HMB by slaughter date subclasses. In contrast to the animal performance data, no significant HMB by slaughter group interactions were apparent for marbling score or quality grade, except for U.S Standards. Trends with HMB followed those observed previously with KIC, the precursor of HMB. Van Koevering et al. (1989b) noted that marbling score increased linearly with increasing dietary KIC. In that study, KIC increased the percentage of cattle grading U.S. Choice or higher.

Changes in s.c. fat thickness and marbling may indicate that the rate and site of fat deposition is altered by HMB. The slope of the regression of marbling score on s.c. fat thickness was altered (P<.01) by HMB (Figure 3). Based on confidence intervals, the Y intercepts were similar with and without

HMB but the slope of the line was greater (P<.07) for steers fed HMB. The rate of deposition of intramuscular fat was greater for steers fed HMB indicating that more fat was deposited intramuscularly per unit increase in s.c. fat when HMB was fed.

Using the regression equations in figure 3, all steers were adjusted to constant quality grade of low U.S. Choice (marbling score = 400); following adjustment, steers fed HMB had less (.78 vs .82 cm) s.c. fat than controls. When all steers were adjusted to a constant s.c. fat thickness of 1.27 cm, steers fed HMB had higher (473 vs 440) marbling scores . Based on the mean marbling score, standard deviation, and adjusted marbling scores (constant s.c. fat thickness ;1.27 cm) for control and HMB, the percentage of steers grading U.S. Choice were calculated to be 57% and 66% respectively. Intramuscular fat, which is advantageous to carcass quality, normally is considered to be deposited only after a substantial amounts of s.c. fat is deposited. The ability of HMB to alter fat partitioning between these sites seems unique and cannot be explained metabolically at present.

Plasma and Tissue Composition. The main effects of HMB averaged across slaughter date is illustrated in Table 10. Plasma concentrations of HMB were greater (P<.001) in samples obtained 16 h post-feeding for steers fed HMB; concentrations of KIC were not different due to treatment. Plasma cholesterol concentrations were lower (108.4 vs 118.7; P<.03) for steers fed HMB. Data for HMB and slaughter date subclasses are presented in Table 11. Plasma concentrations of HMB were higher (P<.05) for every slaughter group except when steers were fed for 133 d. Plasma concentrations of cholesterol tended to be lower in every slaughter group except steers fed for 105 d.

For steers fed 105 d, those fed HMB had higher (P<.10; Table 11) amounts of total lipid within the LM. No other differences due to HMB were detected in tissue concentrations of total lipid, protein or moisture. However when averaged across slaughter dates (Table 10), no differences in LM composition due to HMB were detected. Steers fed HMB tended to have more (4%) lipid deposited within the LM as noted previously by Van Koevering et al. (1989b) with KIC. In contrast to results of Van Koevering et al. (1989b) where KIC reduced LM cholesterol content, the cholesterol content of the LM was not significantly affected by HMB. In the former study, KIC was fed until steers were slaughtered. The 5 d withdrawal period for HMB in this study may have relieved any inhibition of  $\beta$ -hydroxy- $\beta$ -methylgutaryl CoA reductase by HMB and permitted cholesterol biosynthesis to increase so that values were normal.

### Implications

Beta-hydroxy-β-methylbutyrate enhance carcass quality by altering the site of fat deposition and enhancing the intramuscular/subcutaneous fat ratio in feedlot steers. This change could improve quality grade while reducing the amount of trimmable s.c. fat. The effects of HMB on live animal performance remain unclear. With an increased consumer demand for high quality but lean beef, feeding HMB to steers may prove beneficial.

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	Diet Sequence							
Ingredient	1	2	3	4	Final			
			(%)					
Corn, rolled	40.20	50.20	60.20	70.20	82.20			
Alfalfa hay, pelleted	25.00	20.00	15.00	10.00	4.00			
Cottonseed hulls	25.00	20.00	15.00	10.00	4.00			
Molasses, cane	4.00	4.00	4.00	4.00	4.00			
Pelleted supplement <sup>a</sup>	5.80	5.80	5.80	5.80	5.80			

# Table 1. Composition of diets (dry matter basis)

Calculated Composition:

	Final	Diet
Nutrients	DM %	As Fed %
Dry matter, % NEm, Mcal/kg NEg, Mcal/kg Crude protein, % K, % Ca, % P, %	100.00 2.09 1.33 11.90 .70 .54 .32	87.80 1.84 1.17 10.45 .62 .48 .28

<sup>a</sup> Supplement composition: Cottonseed meal, 65.9%; calcium carbonate, 16.6%; urea, 9.49%; salt, 6.04%; dicalcium phosphate, 1.22%; β-hydroxy-β-methyl butyrate 0 or .52%; vitamin A, D, E, .20%; Manganese Dioxide .02%.

				Observed L	l Significance evel (P<)
	Control	НМВ	SEM	НМВ	HMB x Days fed
Pens, no. Steers, no. <sup>b</sup>	16 126	16 127			
Live weight, kg Initial Final <sup>C</sup>	329 508	329 503	.07 1.61	.10	.01
ADG, kg Pre HMBd Overall	1.64 1.42	1.62 1.39	.03 .01		.09 .01
DMI, kg/day Pre HMB <sup>d</sup> Overall	10.32 10.18	10.28 10.11	.08 .08		.04 .04
Feed/Gain Pre HMB <sup>d</sup> Overall	6.38 7.21	6.41 7.27	.11 .06		.01
Calc. energy in diet, over ME, Mcal/kg DM NEm, Mcal/kg DM NEg, Mcal/kg DM	rall 2.79 1.77 1.17	2.77 1.75 1.15	.01 .01 .01		.02 .02 .02

# Effects of $\beta$ -Hydroxy- $\beta$ -Methylbutyrate (HMB) on performance of feedlot steers averaged across days on feed<sup>a</sup> Table 2.

 a Least squares means; SEM with n = 16.
 b Two control steers and one HMB steer were removed for reasons unrelated to treatments.

<sup>c</sup> Calculated as hot carcass weight/.6495 (mean dressing percentage for all steers). d Pre HMB = Time before HMB was fed.

Time on feed, d	1	05		119	1	33	· · · · · · · · · · · · · · · · · · ·	147		
Diet	Control	HMB	Control	HMB	Control	HMB	Control	HMB	SEM	
Pens, no. Steers, no. <sup>b</sup>	4 30	4 32	4 32	4 31	4 32	4 32	4 32	4 32		
Live weight, kg Initial wt Final wt <sup>c</sup>	330 464e	330 479 <sup>f</sup>	329 504	330 498	330 515	329 518	329 548 <sup>e</sup>	329 519 <sup>f</sup>	.14 3.23	
ADG, kg Pre HMBd Overall	1.45 1.30	5 1.55 9 1.431	1.80	) 1.72 7 1.42	1.62 1.40	1.72 1.42	1.66 1.51	9 1.48 <sup>h</sup> e 1.31 <sup>f</sup>	.06 .03	
DMI, kg/day Pre HMB <sup>d</sup> Overall	10.48 9.68	10.88 10.16	10.36 10.26	9 9.75 <sup>1</sup> 9 9.81j	10.21 10.28	10.48 10.46	10.21 10.51	10.02 i 10.02j	.16 .16	
Feed/Gain Pre HMB <sup>d</sup> Overall	7.32 7.48	2 7.10 7.12	5.77 7.00	7 5.66 ) 6.94	6.30 7.37	6.12 7.38	6.13 6.97	i 6.76j e 7.65 <sup>f</sup>	.22 .12	
Calc. energy of diet ME, Mcal/kg NEm, Mcal/kg NEg, Mcal/kg	2.70 1.68 1.10	2.76 1.74 1.14	2.81 1.79 1.19	2.84 1.81 1.20	2.76 1.74 1.15	2.76 1.73 1.14	2.88 1.85 1.23	e 2.73 <sup>f</sup> e 1.72 <sup>f</sup> e 1.13 <sup>f</sup>	.03 .03 ·02	

Table 3. Effects of  $\beta$ -Hydroxy- $\beta$ -Methylbutyrate (HMB) on performance of feedlot steers<sup>a</sup>

а

Least squares means; SEM with n = 4. Two control steers and one HMB steer were removed for reasons unrelated to treatments. Calculated as hot carcass weight/.6495 (mean dressing percentage for all steers). Pre HMB = Time before HMB fed. b

С

d

efghij Means within a slaughter group with different superscripts differ (ef, P<.01; gh, P<.05; ij, P<.10).

				Observed Significance Level (P<)		
	Control	НМВ	SEM	НМВ	HMB x Days fed	
Pens, no. Steers, no. <sup>b</sup>	16 126	16 127				
Carcass weight, kg <sup>C</sup> Dressing percent <sup>d</sup> Longissimus area, cm <sup>2</sup> Fat thickness, cm KPH, % Overall maturity <sup>e</sup> Yield Grade Condemned liver %	330 65.21 85.0 1.07 1.78 145 2.46 10.27	327 65.15 83.6 .99 1.77 144 2.42 12.50	1.07 .18 .85 .03 .04 1.04 .06 3.81	.05 .08	.01	

# Effects of $\beta$ -Hydroxy- $\beta$ -Methylbutyrate (HMB) on carcass characteristics averaged across days on feed<sup>a</sup> Table 4.

 Least square means; SEM with n = 16.
 Two control steers and one HMB steer were removed for reasons unrelated to treatments.

c Carcass weight adjusted for trimming loss.

Calculated by dividing (final shrunk live weight by hot carcass weight)x100.
 Calculated by averaging lean and skeletal maturities; 100="A"; USDA, 1989.

Time on feed, d	10	)5	11	9	133	3	147	7	
Diet	Control	HMB	Control	HMB	Control	HMB	Control	HMB	SEM
Pens, no.	4	4	4	4	4	4	4	4	
Steers, no. <sup>b</sup>	30	32	32	31	32	32	32	32	
Carcass weight, kg <sup>C</sup>	303h	312 <sup>i</sup>	328	323	335	336	356 <sup>f</sup>	3379	2.14
Dressing percent <sup>d</sup>	64.70	65.45	65.44	65.12	64.85	64.77	65.86	65.24	.35
Longissimus area, cn	n <sup>2</sup> 83.0	82.1	83.7	84.1	85.4	86.4	87.8 <sup>h</sup> .	81.9 <sup>i</sup>	1.70
Fat thickness, cm	.91	.81	1.02	.97	1.12	1.07	1.24	1.09 <sup>k</sup>	.06
KPH. %	1.48	1.47	1.61	1.61	2.00	2.06	2.02	1.95	.07
Overall maturity <sup>e</sup>	137	139	147	143	148	147	149	148	2.08
Yield Grade. %	2.11	2.13	2.42	2.31	2.56	2.50	2.76	2.73	.12
Condemned liver, %	3.57	21.88	9.38	12.50	6.25	12.50	21.88	3.13	7.62

Effects of β-Hydroxy-β-Methyl Butyrate (HMB) on carcass characteristics of feedlot steers<sup>a</sup> Table 5.

а b

Least square means; SEM with n = 4. Two control steers and one HMB steer were removed for reasons unrelated to treatments.

С Carcass weight adjusted for trim loss.

d

d Calculated by dividing (final shrunk live weight by hot carcass weight)x100.
 e Calculated by averaging lean maturity and skeletal maturities; 100="A"; USDA, 1989).
 fghijk Means within a slaughter group with different superscripts differ (fg, P<.01; hi, P<.05; jk, P<.10).</li>

Time on feed, d	105		119		133		147		
Diet	Control	HMB	Control	HMB	Control	HMB	Control	HMB	SEM
Pens, no.	4	4	4	4	4	4	4	4	
Steers, no. <sup>b</sup>	30	32	32	31	32	32	32	32	
Yield Grade 1, %	34.82	40.63	18.75	32.14	21.88	37.50	28.13	15.63	6.92
Yield Grade 2, %	58.04	53.13	56.25	45.09	50.00	40.63	31.25 <sup>e</sup>	46.88 <sup>f</sup>	5.93
Yield Grade 3, %	7.14	3.13	21.88	22.77	21.88	21.88	25.00	34.38	7.53
Yield Grade 4, %	0	3.13	0	0	3.13	0	15.63 <sup>c</sup>	3.13 <sup>d</sup>	3.90

Table 6. Effects of β-Hydroxy-β-methyl Butyrate (HMB) on yield grades of feedlot steers<sup>a</sup>

а

Least square means; SEM with n = 4. Two control steers and one HMB steer were removed for reasons unrelated to treatments. Means within a slaughter group with different superscripts differ (<sup>Cd</sup>, P<.05; <sup>ef</sup>, P<.10). b

cdef

U.S. Quality Grade	Choice	e or >	Sele	ect	Star		
Diet	Control	HMB	Control	НМВ	Control	НМВ	SEM
Steers, no.	76	71	42	54	7	2	
Yield Grade 1, % Yield Grade 2, % Yield Grade 3, % Yield Grade 4, %	7.60 56.00 28.12 5.87	14.95 52.32 30.29 2.56	48.53 42.21 6.10 2.90	50.77 40.93 7.72 .35	80.00 15.58 .49 3.37	100 -14.90 16.18 -1.45	11.62 14.15 11.03 4.61

# Table 7. Effects of U.S. quality grades and $\beta$ -Hydroxy- $\beta$ -Methyl Butyrate (HMB) on yield grades of feedlot steers<sup>a</sup>

<sup>a</sup> Least square means; SEM averaged across quality grade and treatment.

				Observed Le	Significance evel (P<)
, ,	Control	HMB	SEM	НМВ	HMB x Days fed
Steers, no. <sup>b</sup> Pens, no.	126 16	127 16			
Marbling Score <sup>C</sup> Prime %	422	432	8.12		
Low, % Total	<u>0.00</u> 0.00	<u>0.89</u> .89	.63 .63		
Choice, % High Quality <sup>d</sup> , % High, % Average, % Low, % Total	5.47 5.47 13.39 <u>41.07</u> 59.93	9.49 8.59 8.71 <u>37.95</u> 55.25	2.02 2.15 2.54 3.47 3.91		
Select, % High, % Average, % Low, % Total	8.59 14.73 <u>10.38</u> 34.04	15.63 11.83 <u>14.84</u> 42.30	2.45 3.28 2.37 3.14	.09 .10	
Standard, % High, % Total	<u>6.03</u> 6.03	<u>1.56</u> 1.56	1.04 1.04	.01 .01	.02 .02

# Effects of $\beta$ -Hydroxy- $\beta$ -Methyl Butyrate (HMB) on marbling score and U.S. quality grades of steers averaged across days on feed<sup>a</sup> Table 8.

 a Least squares means; SEM with n= 16.
 b Two control steers and one HMB steer were removed for reasons unrelated to treatments.

c 300 to 399, Slight; 400 to 499, Small.
 d High Quality = a combination of High Choice and Low Prime.

Time on feed, d	10	)5	1	19	13	3	14	47		
Diet	Control	HMB	Control	HMB	Control	HMB	Control	HMB	SEM	
Pens, no. Steers, no. <sup>b</sup> Marbling Score <sup>c</sup>	4 30 371	4 32 383	4 32 428	4 31 438	4 32 453	4 32 451	4 32 437	4 32 454	16.2	
Prime, Low, % Total	<u>0.00</u> 0.00	<u>0.00</u> 0.00	<u>0.00</u> i 0.00i	<u>3.57j</u> 3.57j	<u>0.00</u> 0.00	<u>0.00</u> 0.00	<u>0.00</u> 0.00	<u>0.00</u> 0.00	1.26 1.26	
Choice, High Quality <sup>d</sup> , % High, % Average, % Low, % Total	3.13 3.13 6.70 <u>20.54</u> 30.36	3.13 3.13 3.13 <u>31.25</u> 37.50	3.13 3.13 15.63 <u>46.88</u> 65.63	9.82 6.25 12.95 <u>33.04</u> 52.24	6.25 6.25 18.75 <u>53.13</u> i 78.13	12.50 12.50 12.50, <u>34.38</u> J 59.38	9.38 9.38 12.50 <u>43.75</u> 65.63	12.50 12.50 6.25 <u>53.13</u> 71.88	4.04 4.29 5.08 6.94 7.82	
Select, High, % Average, % Low, % Total	10.71 21.43 <u>16.52</u> 49.55	15.63 12.50 <u>28.13</u> 56.25	12.50 12.50 <u>9.38</u> 34.38	15.63 12.95 <u>15.63</u> 44.20	3.13 <sup>9</sup> 15.63 <u>3.13</u> 21.88	21.88 <sup>h</sup> 9.38 <u>9.38</u> 40.63	9.38 9.38 <u>12.50</u> 31.25	9.38 12.50 <u>6.25</u> 28.13	4.89 6.57 4.74 6.29	
Standard, High, % Total	<u>20.98</u> e 20.98e	<u>6.25</u> 6.25	<u>0.00</u> 0.00	<u>0.00</u> 0.00	<u>0.00</u> 0.00	<u>0.00</u> 0.00	<u>3.13</u> 3.13	<u>0.00</u> 0.00	2.07 2.07	

Table 9. Effects of β-Hydroxy-β-Methyl Butyrate (HMB) on marbling score and U.S. quality grades of steers<sup>a</sup>

a Least squares means; SEM with n = 4.
b Two control steers and one HMB steer were removed for reasons unrelated to treatments.
c 300 to 399 = Slight; 400 to 499 = Small.
d High Quality = a combination of High Choice and Low Prime.
efghij Means within a slaughter group with different superscripts differ (ef, P<.01; gh, P<.05; ij, P<.10).</li>

				Observed Le	Significance evel (P<)
,	Control	НМВ	SEM	НМВ	HMB x Days fed
Pens, no. Steers, no. <sup>b</sup>	16 126	16 127			
<u>Plasma:</u> HMB, ug/ml KIC, ug/ml Cholesterol, mg/dL	1.70 10.73 118.7	3.06 10.80 108.4	.22 .47 2.86	.001 .03	
<u>Longissimus Muscle:</u> Proximate Analysis, % Total lipid Protein Moisture	3.53 22.51 73.34	3.68 22.36 73.28	.13 .08 .09		
Cholesterol, mg/100g wet tissue	48.98	49.71	.57		

# Effects of $\beta$ -Hydroxy- $\beta$ -Methyl Butyrate (HMB) on composition of plasma and longissimus muscle averaged across days on feed<sup>a</sup> Table 10.

 Least square means; SEM with n = 16.
 Two control steers and one HMB steer were removed for reasons unrelated to treatments.

Time on feed, d	105		119	·	133		147			
- Diet	Control	НМВ	Control	НМВ	Control	НМВ	Control	НМВ	SEM	-
Pens, no. Steers, no.	4 30	4 32	4 32	4 31	4 32	4 32	4 32	4 32		
<u>Plasma:</u> HMB, ug/ml KIC, ug/ml Cholesterol, mg/dL	1.36 <sup>C</sup> 11.96 94.88	3.12 <sup>d</sup> 12.29 98.22	1.80 <sup>c</sup> 10.58 124.96	3.45 <sup>d</sup> 10.25 111.35	2.18 11.08 125.01 <sup>c</sup>	2.60 11.44 106.46 <sup>d</sup>	1.47 <sup>C</sup> 9.29 129.90	3.06 <sup>d</sup> 9.23 117.45	.44 .95 5.73	
Longissimus Muscle: Proximate Analysis Total Lipid Protein Moisture	, % 2.69 <sup>e</sup> 22.83 73.84	3.33 <sup>f</sup> 22.57 73.44	3.69 22.42 73.23	3.63 22.46 73.23	3.93 22.12 73.23	3.58 22.12 73.57	3.81 22.69 73.06	4.19 22.31 72.87	.25 .16 .17	
Cholesterol mg/100g wet tissue	48.65	45.85	46.31	47.69	50.01	51.60	50.96	53.69	1.14	

Table 11.	Effects of β-Hydroxy-β-Methyl Butyrate (HMB) on composition of plasma and longissimus muscle
	averaged across slaughter dates. <sup>a</sup>

a b

Least square means; SEM with n = 4. Two control steers and one HMB steer were removed for reasons unrelated to treatments. Means within a slaughter group with different superscripts differ (cd, P<.05; ef, P<.10).

cdef

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# Legend for Figure

Figure 1. Effects of  $\beta$ -Hydroxy- $\beta$ -Methylbutyrate (HMB) on s.c. fat thickness of feedlot steers.

Figure 2. Effects of  $\beta$ -Hydroxy- $\beta$ -Methylbutyrate (HMB) on calculated yield grades of feedlot steers averaged across days on feed.

Figure 3. Relationship between s.c. fat thickness (12 th rib) and marbling deposition with in the longissimus muscle (marbling score: 100 = Practically Devoid; 800 = Moderately Abundant).







# EFFECTS OF $\beta$ -HYDROXY- $\beta$ -METHYLBUTYRATE ON THE TENDERNESS OF RIBEYE STEAKS FROM FEEDLOT STEERS

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### ABSTRACT

Beta-hydroxy- $\beta$ -methylbutyrate (HMB), a compound formed during in vivo catabolism of leucine, was fed to 256 crossbred steers at a rate of 0 or 0.03% of diet dry matter. Groups of 32 steers per diet were slaughtered after 105, 119, 133 and 147 days on feed. Effects HMB on the cooking properties, tenderness, and composition of ribeye steaks were evaluated. Averaged across slaughter date, the cooking properties of ribeye steaks were not altered due to HMB feeding; however, Warner-Bratzler shear force values were reduced (P<.004) by HMB feeding. Fewer (P<.02) steaks from HMB fed steers were considered tough (> 4.54 kg force/ 1.27 cm core). Steaks from steers fed HMB for 133 and 147 d were reduced most dramatically, being 8.6% (P<.08) and 9.6% (P<.05) lower than for control steers, respectively. This resulted in fewer HMB steaks being considered tough, with the largest difference (P<.05) at 147 d. Averaged across slaughter group, total lipid and moisture content within the longissimus muscles (LM) was not changed by HMB feeding; however, the protein content tended (P<.09) to be lower when steers were fed HMB. Compared with control steers, steers fed HMB for 105 d tended (P<.07) to have a greater amounts of total lipid within the LM, while steers fed HMB for 147 d had less (P<.05) protein (%) within the LM. Correlation coefficients were low and could only account for a maximum of 20% of the variation in shear force. (Key Words: Tenderness, Fat, Feedlot Steers.)

### INTRODUCTION

Increased consumer demand for leaner products, has lead into the investigation of compounds that increase protein accretion and decrease fat synthesis (Moseley et al., 1990; Lanna et al., 1992; Wheeler and Koohmaraie, 1992). The use of compounds like  $\beta$ -adrenergic agonists and somatotropin in various species has increased production of lean meat in cattle (Carroll et al., 1990; Fabry and Sommer, 1990; Moseley et al., 1990), sheep (Hamby et al., 1986), swine (Jones et al., 1985) and poultry (Gwartney et al., 1992). However, the use of  $\beta$ -adrenergic agonists and somatotropin, generally has decreased muscle tenderness. Increased amounts of shear force (kg force/1.27 cm core) are required to shear muscle of cattle (Fabry et al., 1990; Vestergaard et a;., 1990; Wheeler and Koohmaraie, 1992), sheep (Hamby et al., 1986; Pringle et al., 1991), swine (Jones et al., 1985; Solomon et al., 1991; Chang et al., 1992) and poultry (Morgan et al., 1989; Gwartney et al., 1992) fed these products; this is believed to be caused primarily by the reduced proteolytic enzyme activity due to increased calpastatin activity and increased fiber diameter (Morgan et al., 1989; Pringle et al., 1991; Solomon et al., 1991; Wheeler and Koohmaraie, 1992).

HMB is a compound produced during in vivo catabolism of the amino acid leucine (LEU). Van Koevering and Nissen (1992) found that in lambs and pigs, HMB was produced directly from the oxidation of either leucine or its keto acid,  $\alpha$ ketoisocaproate (KIC). More specifically, the oxidation of KIC to HMB has been demonstrated to occur in the liver via the cytosolic enzyme KIC oxygenase (Sabourin and Bieber 1982a, 1982b). The effect HMB has on muscle tissue has not yet been assessed; therefore, the objective of this study was to determine if feeding HMB to feedlot steers would alter the tenderness of ribeye steaks.

## MATERIALS AND METHODS

Animals and Diets. Two hundred and fifty-six crossbred steers (329 kg) were selected from a larger group (n=570) based on uniformity in size, weight and breed-type. Steers visually appraised as having greater than 25% Bos Indicus or Angus characteristics were removed, leaving steers of primarily British x Continental breed-type. This was done to reduce variability in marbling scores. Steers were processed by routine feedlot practices and implanted with an estrogenic implant (24 mg estradiol; Compudose®) at a commercial feedlot prior to arrival at Panhandle State University in Goodwell, OK. Upon arrival, individual steers were weighed, identified, and blocked into groups based on initial weight. Steers were assigned randomly to pens and allotted to treatments in a serial slaughter 2 x 4 factorial arrangement. One-half the pens (16) of cattle received the basal diet (controls); the other 16 pens of cattle received the same diet supplemented with HMB at .03% of the diet dry matter. Sets of four pens were assigned to be fed for a different number of days; these steers were fed for either 105, 119, 133 or 147 d after arrival. HMB was fed for only the final 82 d of each feeding period; HMB was removed from the diet 5 d prior to slaughter, the withdrawal time specified by the U.S. Food and Drug Administration for this experimental compound.

*Carcass Data and Longissimus Muscle Sampling.* Carcass data for all slaughter groups were obtained approximately 48 hr postmortem; yield and quality grades were determined (USDA, 1989). A 20 cm thick section of the LM corresponding to the 9 through 12th rib section was removed from the left side of each carcass; it was vacuum packaged, and shipped to the Oklahoma State University Meat Laboratory. LM sections were aged at 2°C for 14 d postmortem. Ribeye samples subsequently were frozen (-30°C) and faced (uneven portion removed from the posterior end) before being fabricated into steaks for determining

composition. A 1.3 cm thick LM steak was removed from the posterior end of each LM section; it was denuded of exterior fat and epimysial connective tissue, and stored for proximate analysis. Immediately anterior to the steak used for proximate analysis, the remaining LM was cut into 2.5 cm thick steaks to be used for Warner-Bratzler shear force determinations.

Longissimus Muscle Cooking Properties and Shear Force. Cooking properties and shear force determinations were conducted as described in AMSA (1978). LM steaks used to determine shear force were thawed at 2°C for 24 h, trimmed of s.c. fat, weighed and broiled on a Faberware® (Faberware, Bronx, NY) open hearth broiler to a final internal temperature of 70°C. Cooking time to a medium degree of doneness (minutes/100 g raw steak) and cooking shrinkage (percentage weight loss) were calculated for each steak. Steaks were allowed to cool to 25°C, after which six cores (1.27 cm diameter) were removed parallel to the longitudinal direction of the muscle fibers. Cores then were individually sheared using a Instron® Model SD-50 Warner-Bratzler shear apparatus(Instron, Canton, MA) to determine the peak force required.

*Data Analysis.* Data were analyzed on a individual animal basis using least squares analysis (SAS, 1988) with a linear model that included the main effects of HMB presence (df = 1), weight block (df = 3), slaughter date (df = 3) and all two way interactions. Least squares means were calculated and treatment means were compared using T-tests. Three steers were removed from data set for reasons not related to dietary treatments.

### **RESULTS AND DISCUSSION**

*Cooking Properties of Ribeye Steaks.* Averaged across slaughter date no differences due to HMB with respect to the cooking properties of ribeye steaks were significant (Table 1). Steaks form control steers fed for 147 d (Table 2) tended to have greater (P<.08) raw weights than steers fed HMB. This increase in

raw weight may have be a result of the increase in carcass weight and in REA in control steers fed for 147 d. Cooking shrink, which would be a indicator of relative changes, was not affected by HMB for steers fed 147 d. Cooking time (min./100 g raw tissue) was less (P<.05) for steaks taken from HMB steers fed for 105 d (Table 2), and may be the result of an increased amount of total lipid within these steaks. Cooking properties also were determined for dietary treatment by quality grade subclasses (Table 3). No differences due to HMB were observed between steaks of different quality grades.

Tenderness of Ribeye Steaks: Tenderness of the LM in this study was measured by determining Warner-Bratzler shear force. Averaged across days on feed, shear force (Table 1) was much lower (P<.004) in steaks from HMB-fed steers. The nonsignificant HMB x days fed interaction indicates that this effect was consistent across all slaughter groups as is illustrated in Table 2. Although the HMB x days fed interaction was nonsignificant, comparisons within each slaughter group were made as orthogonal contrasts. Shear force in steaks from HMB-fed steers were numerically lower at each slaughter date (Table 2), with the difference being most dramatic for steers fed for 133 d (P<.08) and 147d (P<.05). The decrease in shear force averaged across days fed (Table 1) resulted in steaks from HMB fed steers being more predominantly classified as very tender (< 3.86 kg force/1.27 cm core) and tender (3.86 < tender < 4.54 kg force/1.27 cm core), while fewer (P<.02) steaks from HMB fed steers were considered to be tough (> 4.54 kg force/1.27 cm core; P<.02). Similar to shear force, the HMB x days fed interaction was not significant for the percentage of very tender, tender and tough steaks. The treatment by slaughter date subclasses are illustrated in table 2; the effects were most dramatic among steers fed for 147 d, with the steaks from the control steers having a greater (P<.05) percentage of tough steaks. Several factors may affect the tenderness of the LM. These include differences in fiber diameter, amount and

solubility of collagen, and the activity of postmortem proteases. Without additional measurements, it is difficult to determine how HMB increased tenderness of the LM steaks.

Table 3 illustrates the HMB by quality grade subclasses. Carcasses grading U.S. Choice or greater from steers fed HMB had lower (P<.04) shear force values as compared to the controls. A similar pattern existed for carcasses grading U.S. Select and Standard (P<.04) to have lower shear force values. This decrease in shear force resulted in an concomitant increase (P<.04) in the number of steaks be considered very tender (< 3.86 kg force/1.27 cm core) from steers fed HMB and grading both U.S. Choice and Standard. These data would suggest that feeding HMB to feedlot steers could reduce the amount of variation in and improve the consistency of tenderness between ribeye steaks of different U.S. quality grades.

*Chemical Composition of Longissimus Muscle:* Averaged across slaughter date (Table 1), there was a tendency for HMB to increase in lipid content of the LM; however, the trend was not consistent enough to reach statistical significance. The percentage of protein within the LM tended (P<.09) to be decreased for steers fed HMB. The negative correlation (r = -.28; P<.01) between lipid and protein content of the LM supports the directional increase in lipid deposition. The moisture content of the LM was not significantly changed by HMB. For steers fed 105 d, those fed HMB had higher (P<.08; Table 2) amounts of total lipid and numerically lower amounts of protein within the LM. Similarly for steers fed 147 d, those fed HMB had lower (P<.05) amounts of protein and numerically more lipid deposited within the LM. Once again, the moisture content of the LM was not significantly altered by HMB. Dietary HMB did not alter the chemical composition of steaks from steers within the different U.S. quality grades (Table 3).

*Correlation Coefficients:* Correlation coefficients averaged across HMB and slaughter group are illustrated in Tables 4 and 5. Shear force and the cooking
properties of ribeye steaks were low but significantly correlated (Table 4; P<.01); cooking shrink (%) accounted for the largest (19%) amount of the variation in shear force. Shear force and LM composition also were correlated (Table 4), with the total amount of lipid being associated negatively with shear force in contrast to a positive association with protein and moisture. However, when carcass characteristics and daily gains were averaged across HMB and slaughter group, shear force was correlated with marbling score (P<.01) and overall maturity (P<.05).

When correlation coefficients were separated by HMB, and averaged across slaughter group (Tables 6 and 7), responses between shear force and the cooking properties and composition of LM steaks were similar as when compared to the overall average. However, when correlation coefficients for shear force and carcass characteristics were determined for HMB treatments (Tables 8 and 9), responses were found to differ. Shear force values from HMB fed steers were correlated (P<.01) to marbling score, LM area and yield grade whereas for control steers, no correlation was detected. Shear force values from control steers were found to be correlated (P<.05) more closely with overall carcass maturity, whereas in HMB-fed steers, no such correlation was detected.

Although certain correlations between shear force and various traits were found to be significant, no more than 20% of the variation in shear force values could be attributed to any single trait such as cooking shrink. Even when all independent class variables and carcass traits were combined, only about 20% of the total variation in shear force could be accounted for.

## IMPLICATIONS

Feeding HMB to feedlot steers enhanced the quality of ribeye steaks by increasing tenderness; fewer steaks were classified as tough. HMB enhanced the tenderness of ribeye steaks from carcasses of different quality grades. Because tenderness is an important factor in the overall acceptability of beef products, addition of HMB to feedlot diets may improve the quality of beef produced.

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				Observed Significance Level (P<)		
	Control	НМВ	SEM	НМВ	HMB x Days fed	
Steers, no. <sup>b</sup>	126	127				
Cooking Properties Raw weight, g Cooked weight, g Cooking Shrink, % Cooking Time <sup>C</sup>	305.97 217.66 28.92 7.02	304.43 217.91 28.42 6.85	2.79 2.19 .28 .15	.70 .94 .21 .42	.23 .25 .44 .21	
Shear Force, kg Very Tender, % <sup>d</sup> Tender, % <sup>e</sup> Tough, % <sup>f</sup>	4.44 30.07 28.66 41.27	4.13 39.51 32.37 27.32	.08 4.16 4.07 4.08	.004 .11 .52 .02	.90 .68 .67 .61	
Proximate Analysis, % Total lipid Protein Moisture	3.53 22.51 73.34	3.68 22.36 73.28	.12 .06 .11	.41 .09 .71	.19 .25 .42	

# Table 1.Effects of $\beta$ -Hydroxy- $\beta$ -Methyl Butyrate (HMB) on Cooking<br/>Properties, Warner-Bratzler shear force and longissimus muscle<br/>composition of feedlot steers averaged across days on feed<sup>a</sup>

<sup>a</sup> Least square means; SEM averaged across treatments.

b Two control steers and one HMB steer were removed for reasons unrelated to treatments.

Cooking time = min./100 g raw tissue.

d Very Tender < 3.86 kg force/1.27 cm core.

e Tender = 3.86 <tender <4.54 kg force/1.27 cm core.

f Tough > 4.54 kg force/1.27 cm core.

Time on feed, d	10	5	11	9	133	3	147		
Diet	Control	HMB	Control	HMB	Control	HMB	Control	НМВ	SEM
Steers, no. <sup>b</sup>	30	32	32	31	32	32	32	32	******
Cooking Properties Raw weight, g Cooked weight, g Cooking Shrink, % Cooking time <sup>b</sup>	290.32 202.15 30.43 8.07 <sup>f</sup>	297.43 210.29 29.31 7.199	292.49 208.95 28.57 7.02	297.48 213.61 28.22 6.81	333.42 240.80 27.80 6.38	328.79 235.59 28.30 6.71	307.66 <sup>h</sup> 218.74 28.90 6.60	293.99 <sup>i</sup> 212.16 27.85 6.68	5.59 4.37 .56 .30
Shear Force, kg Very Tender, % <sup>C</sup> Tender, % <sup>d</sup> Tough, % <sup>e</sup>	4.57 23.41 24.00 52.60	4.34 34.38 25.00 40.63	4.52 28.13 37.50 34.38	4.29 26.78 38.88 31.17	4.30 <sup>n</sup> 34.38 28.13 37.50	3.93 <sup>1</sup> 53.13 25.00 21.88	4.37 <sup>†</sup> 34.38 25.00 40.63 <sup>†</sup>	3.959 43.75 40.63 15.639	.33 8.26 8.08 8.10
Proximate Analysis, % Total Lipid Protein Moisture	% 2.69h 22.83 73.83	3.33 <sup>i</sup> 22.57 73.44	3.69 22.42 73.23	3.62 22.46 73.24	3.93 22.12 73.23	3.58 22.12 73.57	3.81 22.69 <sup>f</sup> 73.06	4.19 22.319 72.87	.25 .12 .23

Table 2.	Effects of $\beta$ -Hydroxy- $\beta$ -Methyl Butyrate (HMB) and slaughter group on cooking properties, Warner	r-
	Bratzler shear force, and longissimus muscle composition of rib steaks from feedlot steers <sup>a</sup>	

a b c d e fghi

Least square means; SEM averaged across treatment and slaughter group. Cooking time = min./100 g raw tissue. Very Tender < 3.86 kg force/1.27 cm core. Tender = 3.86 <tender <4.54 kg force/1.27 cm core. Tough > 4.54 kg force/1.27 cm core. Means within a slaughter group with different superscripts differ (<sup>fg</sup>, P<.05; <sup>hi</sup>, P<.08).

U.S. Quality Grade	Choice	or >	Sele	ct	Standar	d		
Diet	Control	HMB	Control	HMB	Control	HMB	SEM	
Steers, no.	76	71	42	54	7	2		
Cooking Properties Raw weight, g Cooked weight, g Cooking Shrink, % Cooking time, <sup>b</sup>	305.81 217.16 29.08 7.32	301.45 215.58 28.50 6.90	305.07 217.58 28.70 6.57	309.44 221.48 28.41 6.76	312.56 222.99 28.61 6.78	275.07 204.45 25.73 7.08	8.92 7.00 .90 .49	
Shear Force, kg Very Tender, % <sup>C</sup> Tender, % <sup>d</sup> Tough, % <sup>e</sup>	4.39 <sup>f</sup> 29.94 <sup>f</sup> 30.06 40.01	4.049 45.029 27.18 27.61	4.50 31.61 26.02 42.43	4.28 29.75 39.80 28.89	4.62 <sup>f</sup> 23.10 <sup>f</sup> 29.37 47.19	3.129 106.579 17.26 -25.36	.24 13.19 13.04 13.07	
Proximate Analysis, % Total Lipid Protein Moisture	4.18 22.41 72.83	4.40 22.23 72.67	2.84 22.66 73.93	2.78 22.53 74.04	1.56 22.68 75.03	2.12 22.94 74.47	.34 .20 .32	

#### Table 3. Effects of U.S. quality grades and β-Hydroxy-β-Methyl Butyrate (HMB) on cooking properties, Warner-Bratzler shear force, and longissimus muscle composition of ribeye steaks from feedlot steers<sup>a</sup>

<sup>a</sup> Least square means; SEM averaged across quality grade and treatment.

b Cooking time = min./100 g raw tissue.
c Very Tender < 3.86 kg force/1.27 cm core.</li>
d Tender = 3.86 <tender <4.54 kg force/1.27 cm core.</li>

e Tough > 4.54 kg force/1.27 cm core.

fg Means within a quality grade with different superscripts differ P<.04.

Simple correlation coefficients for cooking properties, Warner-Bratzler shear force, and longissimus muscle composition averaged across treatment and slaughter group<sup>a</sup> Table 4.

	Shear	Raw	Cooked	Cooking	Cooking	a Pr	oximate Ana	alysis
	Force	Weight	Weight	Shrink	Time	Fat	Protein	Moisture
Marbling Score Moisture, % Protein, % Fat, % Cooking Time <sup>b</sup> Cooking Shrink, % Cooked Weight, g Raw Weight, g	-0.16 0.17 0.19 -0.22 0.22 0.44 -0.13 0.04	-0.04 0.10 0.04 -0.11 -0.37 -0.02 0.92	-0.03 0.05 0.03 -0.06 -0.51 -0.41	-0.03 0.08 0.02 -0.09 0.42	0.02 -0.01 0.01 0.03	0.70 -0.88 -0.28	-0.30 -0.10	-0.62

a Correlation coefficients  $\ge 0.13$  and  $\le 0.15$  differ from zero (P<.05),and  $\ge 0.16$  differ from zero (P<.01); n = 252. b Cooking time = min./100 g raw tissue.

Simple correlation coefficients Warner-Bratzler shear force, carcass characteristics, and daily gain averaged across treatment and slaughter group<sup>a</sup> Table 5.

	Shear	Daily	Yield	LM	S.C. Fat	Overall	Dressing	Carcass
	Force	Gains	Grade	area	Thickness	Maturity	Percent	Weight
Marbling Score Carcass Weight, kg <sup>b</sup> Dressing, % <sup>C</sup> Overall Maturity <sup>d</sup> S.C. Fat thickness, cm Longissimus area, cm <sup>2</sup> Yield Grade Daily Gain, kg	-0.16 0.07 -0.03 -0.13 -0.08 0.07 -0.06 0.10	0.08 0.95 0.26 -0.28 0.23 0.41 0.13	0.47 0.17 -0.14 0.13 0.85 -0.74	-0.24 0.40 0.35 -0.20 -0.36	0.48 0.27 0.04 0.12	0.13 -0.25 0.04	0.10 0.26	0.15

a Correlation coefficients ≥ 0.13 and ≤ 0.14 differ from zero (P<.05),and ≥ 0.15 differ from zero (P<.01); n = 252.</li>
b Carcass weight adjusted for trimming loss.
c Calculated by dividing (final shrunk live weight by hot carcass weight)x100.
d Calculated by averaging lean and skeletal maturities; 100=A; USDA, 1989.

Table 6. Simple correlation coefficients for cooking properties, Warner-Bratzler shear force, and longissimus muscle composition for control steers averaged across slaughter group<sup>a</sup>

	Shear	Raw	Cooked	Cooking	Cooking	g Pr	oximate Ana	alysis
	Force	Weight	Weight	Shrink	Time	Fat	Protein	Moisture
Marbling Score Moisture, % Protein, % Fat, % Cooking Time <sup>b</sup> Cooking Shrink, % Cooked Weight, g Raw Weight, g	-0.12 0.10 0.22 -0.20 0.20 0.40 -0.22 -0.08	-0.01 -0.01 0.07 0.01 -0.33 -0.07 0.92	-0.02 -0.07 0.14 0.05 -0.43 -0.45	0.04 0.15 -0.20 -0.12 0.35	0.03 -0.02 -0.06 0.06	0.65 -0.85 -0.22	-0.27 -0.17	-0.56

a Correlation coefficients ≥ .20 and ≤ .21 differ from zero (P<.05), and ≥ .22 differ from zero (P<.01); n = 125.</li>
 b Cooking time = min./100 g raw tissue.

Simple correlation coefficients for cooking properties, Warner-Bratzler shear force, and longissimus muscle composition for steers fed  $\beta$ -Hydroxy- $\beta$ -methyl Butyrate (HMB) averaged across slaughter group<sup>a</sup> Table 7.

	Shear	Raw	Cooked	Cooking	Cooking	) Pr	oximate Ana	alysis
	Force	Weight	Weight	Shrink	Time	Fat	Protein	Moisture
Marbling Score Moisture, % Protein, % Fat, % Cooking Time <sup>b</sup> Cooking Shrink, % Cooked Weight, g Raw Weight, g	-0.22 0.26 0.12 -0.25 0.27 0.44 -0.03 0.16	-0.10 0.18 0.03 -0.19 -0.47 0.02 0.92	-0.06 0.15 -0.04 -0.15 -0.64 -0.37	-0.07 0.04 0.17 -0.07 0.52	0.02 -0.02 0.01 0.03	0.74 -0.90 -0.30	-0.28 -0.07	-0.66

a Correlation coefficients ≥ .18 and ≤ .22 differ from zero (P<.05),and ≥ 0.23 differ from zero (P<.01); n = 127. b Cooking time = min./100 g raw tissue.

#### Table 8. Simple correlation coefficients Warner-Bratzler shear force, carcass characteristics, and daily gain for control steers averaged across slaughter group<sup>a</sup>

	Shear	Daily	Yield	LM	S.C. Fat	Overall	Dressing	Carcass
	Force	Gains	Grade	area	Thickness	Maturity	Percent	Weight
Marbling Score Carcass Weight, kg <sup>b</sup> Dressing, % <sup>C</sup> Overall Maturity <sup>d</sup> S.C. Fat thickness, cm Longissimus area, cm <sup>2</sup> Yield Grade Daily Gain, kg	-0.12 0.01 0.04 -0.19 -0.02 -0.11 0.07 0.06	0.17 0.96 0.21 -0.22 0.35 0.31 0.27	0.43 0.31 -0.16 0.14 0.88 -0.74	-0.25 0.29 0.35 -0.17 -0.41	0.38 0.38 -0.01 0.16	0.11 -0.19 -0.08	-0.07 0.18	0.22

a Correlation coefficients ≥ .19 and ≤ .21 differ from zero (P<.05),and ≥ .22 differ from zero (P<.01); n = 125.</li>
 b Carcass weight adjusted for trimming loss.
 c Calculated by dividing (final shrunk live weight by hot carcass weight)x100.
 d Calculated by averaging lean and skeletal maturities; 100=A; USDA, 1989.

Simple correlation coefficients Warner-Bratzler shear force, carcass characteristics, and daily gain for steers fed  $\beta$ -Hydroxy- $\beta$ -Methyl Butyrate (HMB) averaged across slaughter group<sup>a</sup> Table 9.

	Shear	Daily	Yield	LM	S.C. Fat	Overall	Dressing	Carcass
	Force	Gains	Grade	area	Thickness	Maturity	Percent	Weight
Marbling Score Carcass Weight, kg <sup>b</sup> Dressing, % <sup>C</sup> Overall Maturity <sup>d</sup> S.C. Fat thickness, cm Longissimus area, cm <sup>2</sup> Yield Grade Daily Gain, kg	-0.21 0.10 -0.07 0.02 -0.14 0.23 -0.21 0.12	0.01 0.95 0.34 -0.34 0.10 0.52 -0.03	0.50 0.01 -0.13 0.10 0.83 -0.73	-0.22 0.52 0.35 -0.21 -0.31	0.56 0.15 0.01 0.06	0.13 -0.31 0.09	0.21 0.36	0.08

a Correlation coefficients = 0.21 differ from zero (P<.05),and ≥ 0.22 differ from zero (P<.01); n = 127.</li>
 b Carcass weight adjusted for trimming loss.
 c Calculated by dividing (final shrunk live weight by hot carcass weight)x100.
 d Calculated by averaging lean and skeletal maturities; 100=A; USDA, 1989.

## EFFECT OF SLAUGHTER DATE ON PERFORMANCE, CARCASS CHARACTERISTICS, AND TISSUE COMPOSITION OF FEEDLOT STEERS

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## ABSTRACT

Two hundred and fifty-six (256) crossbred yearling steers initially weighing 329 kg were used to study the effect of slaughter date on live performance, carcass characteristics, tissue composition and tenderness. Steers were divided into four slaughter groups (64 steers) and fed for either 105, 119, 133 or 147 d. Daily gains (carcass adjusted basis) tended (P<.07) to increase in a quadratic manner while feed intake increased (P<.03) linearly as cattle were fed more days. Efficiency of feed conversion (carcass adjusted basis), greatest (P<.05) for steers fed 119 d, responded in a cubic fashion (P<.01) across slaughter groups. Carcass weight, s.c. fat thickness, KPH, overall carcass maturity, and yield grade increased linearly (P<.01) with slaughter date. Marbling score and the percentage of cattle grading U.S. Choice increased (linearly; P<.01) with time on feed but at a decreasing rate (quadratic; P<.05). Cholesterol and total lipid concentrations increased linearly (P<.01) in longissimus muscle as time on feed increased. While the percentage of protein and moisture decreased (L; P<.10 and P<.01). Tenderness of ribeye steaks tended to increase linearly (P<.07) with slaughter date.

(Key Words: Feedlot Steers, Cholesterol, Tenderness.)

#### INTRODUCTION

The length of time cattle are fed a high concentrate diets is dependent primarily upon economics. Seasonal changes in feed and cattle costs dictate the length of time cattle are fed. Longer feeding periods for cattle of a given starting weight will increase final live weight, hot carcass weight, longissimus area, s.c. fat thickness, yield grade and quality grades (Zinn et al., 1970a; Hicks et al., 1987; Dolezal et al., 1982), only some of which increase the value of cattle. Increases in s.c. fat thickness and yield grade are not conducive to increases in carcass quality and consumer interest. Additional quality factors include cholesterol content and tenderness of ribeye steaks. Tenderness increases with time to a point (139 d Epley et al., 1968; 150 to 180 d Zinn et al., 1970b), after which animal age may have a greater influence, resulting in reduced tenderness. The objective of this study was to evaluate the effects of different slaughter dates on performance, carcass quality, tenderness and cholesterol content.

#### **Materials and Methods**

Animals and Diets. Two hundred and fifty-six crossbred steers (329 kg) were selected from a larger group (n=570) based on uniform size, weight and breed-type. Steers with greater than 25% Bos Indicus or Angus characteristics were removed, leaving steers of primarily British x Continental breed-type. Steers were processed routinely and implanted with an estrogenic implant (24 mg estradiol; Compudose®) at a commercial feedlot prior to arrival at Panhandle State University in Goodwell, OK. Upon arrival, steers were individually weighed, identified, and blocked into 4 weight groups based on initial weight.

Sixteen (16) steers from each weight group were randomly assigned to pens (8 steers/pen) and pens were assigned to specific slaughter dates. Eight pens (2 from each weight group) were assigned to be fed for a different number of days. Steers were fed for either 105, 119, 133 or 147 days after arrival.

Steers were given ad libitum access to their high concentrate diets for the entire feeding period. Cottonseed hulls and chopped alfalfa, used as roughage sources, were removed stepwise from the diet to adapt cattle to their final diet. Diet compositions and analyses are shown in Table 1. Steers were receiving their final ration by day 19 of the study.

Cattle were weighed initially directly off the truck; these weights were used for allocation. Weight gain and feed efficiency were calculated based on initial shrunk weight and final live weights which were calculated from hot carcass weight /.6495, the mean dressing percentage for all cattle. Net energy values were calculated for each treatment using the 1977 yearling steer equation as reported by Hays et al. 1986. Cattle were trucked to Dodge City, Kansas for slaughter. At slaughter, livers were examined for the presence and severity of abscesses.

*Plasma Sampling.* Plasma was obtained 16 h post-feeding 5 d prior to slaughter for each respective slaughter group. Plasma was collected in non-silicone coated Na<sub>2</sub>EDTA tubes and stored at -20<sup>o</sup>C until analyzed. Cholesterol concentrations were determined using Sigma Kit # 352 (Sigma Chemical, St. Louis, MO.).

*Carcass Data and Longissimus Muscle Sampling.* Carcass data for all slaughter groups were obtained approximately 48 hr postmortem; yield and quality grades were determined (USDA, 1989). A 20 cm thick section of the LM corresponding to the 9 through 12th rib section was removed from the left side of each carcass; it was vacuum packaged, and shipped to the Oklahoma State

University Meat Laboratory. LM sections were aged at 2°C for 14 d postmortem. Ribeye samples subsequently were frozen (-30°C) and faced (uneven portion removed from the posterior end) before being fabricated into steaks for determining composition. A 1.3 cm thick LM steak was removed from the posterior end of each LM section; it was denuded of exterior fat and epimysial connective tissue, and stored for proximate analysis. Immediately anterior to the steak used for proximate analysis, the remaining LM was cut into 2.5 cm thick steaks to be used for cholesterol analysis and shear force determination.

Longissimus Muscle Chemical Analysis. Samples were prepared in duplicate for chemical analysis by immersing them in liquid nitrogen and powdering them in a Waring Commercial Blendor® (Model 34B122; Waring, New Hartford, CN). A frozen 3 g sample of powdered LM was subjected to proximate analysis according to procedures outlined by AOAC (1984). Cholesterol content was determined through a modification of Lepage and Roy (1986).

Longissimus Muscle Cooking Properties and Shear Force. Cooking properties and shear force determinations were conducted as described in AMSA (1978). LM steaks used to determine shear force were thawed at 2°C for 24 h, trimmed of s.c. fat, weighed and broiled on a Faberware® (Faberware, Bronx, NY) open hearth broiler to a final internal temperature of 70°C. Cooking time to a medium degree of doneness (minutes/100 g raw steak) and cooking shrinkage (percentage weight loss) were calculated for each steak. Steaks were allowed to cool to 25°C, after which six cores (1.27 cm diameter) were removed parallel to the longitudinal direction of the muscle fibers. Cores then were individually sheared using a Instron® Model SD-50 Warner-Bratzler shear apparatus(Instron, Canton, MA) to determine the peak force required.

Data Analysis. Beta-hydroxy- $\beta$ -methyl butyrate (HMB), a metabolite of leucine, was imposed across this experiment to evaluate the effects of HMB on performance, carcass characteristics and tissue composition. Data were analyzed on a pen basis using least squares analysis (SAS ,1988) with a linear model that included effects of HMB presence (df = 1), weight block (df = 3), slaughter date (df = 3) and all two way interactions being included in the model. Carcass data, chemical composition and shear force values of LM were regressed against the mean carcass weight and s.c. fat thickness. When a slope was significant, but the interaction was not significant, the adjusted means were reported. Least squares means were calculated and slaughter group means were compared using T-tests and linear, quadratic and cubic contrasts.

#### **Results and Discussion**

Steer Performance. The effects of slaughter date on live animal performance are presented in Table 2. Initial weights were not different between slaughter group, even though a linear decrease (L; P<.01) was detected. Final live weight increased (P<.05) resulting in a L increase (P<.01) across slaughter group, but weight increased at a decreasing rate giving a quadratic (Q; P<.03) response. Hicks et al. (1987) fed steers of similar weight for either 100, 114, 128 or 142 d, and reported similar L increases in final live weight. Zinn et al. (1970a) fed steers and heifers between 0 and 270 d with slaughter dates at 30 d intervals. They reported that final live weights increased from 90 to 210 d. May et al. (1992) found similar results, with L increases (P<.01) in slaughter weight from steers fed between 0 and 196 d and slaughter dates at 28 d intervals. Carcass adjusted ADG (Table 2) was the greatest for steers fed 119 d, being 5.7 % greater (P<.05) than steers fed for 105 d but not significantly different from ADG of steers fed for 133 or 147 d. This was similar to the findings of Zinn et al. (1970a) where ADG increased with increasing time on feed; however, as in our

study ADG was not significantly increased after 120 d on feed. In contrast, Hicks et al. (1987) found that ADG on a live weight basis decreased (L; P<.05) as time on feed increased. May et al. (1992) found no differences in ADG among slaughter groups. In the present study, daily feed intake of steers appeared to increase (L; P<.03) across slaughter groups, with steers fed 105 d having lower (P<.05) feed intakes than those fed for 133 d. Hicks et al. (1987) found no differences in feed intake of cattle fed a comparable amount of time. Because feed intake generally is lower the first month than thereafter (Hicks et al. 1990), longer feeding times will dilute this effect and give a higher mean feed intake for the total feeding period.

Steers fed for 119 d had superior (P<.05) efficiency in feed conversion (carcass adjusted basis) as compared to all other groups (Table 2). The cubic (C; P<.01) effect observed in this trait is difficult to explain. Hicks et al. (1987) reported that feed efficiencies became poorer (L; P<.05) across a similar feeding period. Dietary calculated net energy tended to increase (L; P<.08) with slaughter groups; however, the best fit appeared to be C (P<.01) as for feed efficiency.

*Carcass Traits.* The effects of time on feed on carcass traits are illustrated in Table 3. Hot carcass weights increased L (P<.01) but less at later times (P<.05) with slaughter date, which is similar to linear responses previously reported (Zinn et al., 1970a; Hicks et al., 1987; May et al., 1992). Once again, as with final live weights, hot carcass weight increased at a decreasing rate, resulting in a Q (P<.06) response across slaughter group. Dressing percentage was not altered by slaughter group in this study. In contrast Hicks et al. (1987) reported that dressing percentage increased L between 100 and 142 d on feed. Williams et al. (1989) fed medium framed, crossbred steers for 84, 112, or 142 d and found that dressing percent was constant until 142 d. With cattle fed for

longer amounts of time (196 or 270 d) dressing percent has increased with time on feed (Zinn et al., 1970a; May et al., 1992). When dressing percent was adjusted to a mean carcass weight, dressing percent actually decreased (Q; P<.04) with increasing time on feed. This means that the effect of weight on dressing percentage differs from the effect of time on feed on dressing percentage.

Although carcass weights increased, LM area was not altered by slaughter group. These data are not in agreement with the results of Hicks et al. (1987), Williams et al. (1989) and May et al. (1992); they all observed an increase in LM area with an increase in time on feed. Subcutaneous fat thickness (Table 3), however, increased (L; P<.01) together with mean USDA yield grades and percentage of yield grades of 4 or greater (P<.07). This is similar to other reports (Hicks et al., 1987; Miller et al., 1987; May et al., 1992)... The percentage of kidney, pelvic and heart fat (KPH) increased (L; P<.01) with time on feed; with steers fed for less than 119 d having less (P<.05) KPH than steers fed more than 133 d (Table 3). Hicks et al. (1987) found similar results, with steers being fed for 114 d or less having lower percentage of KPH than steers being fed for 128 d or greater. However, the C effect (P<.01) of KPH we detected is difficult to explain.

The maturity of steer carcasses (Table 3) was greater (P<.05) for steers fed 119 d or greater than steers fed 105 d. Overall maturity increased at a decreasing rate, producing both L (P<.01) and Q responses (P<.08). In contrast, Miller et al. (1987) detected no increase in lean, skeletal or overall maturity when cattle where fed up to 168 d. When overall carcass maturity was adjusted to a mean fat thickness, the effect of slaughter group became C (P<.04), this is difficult to explain. The percentage of condemned livers was not affected by slaughter group; which is similar to the findings of Hicks et al. (1987).

When time on feed is extended, marbling scores and percentage of U.S. Choice cattle generally increase (Dolezal et al., 1982; May et al., 1992; Miller et al., 1987). In this present study, marbling scores and the percentage of cattle grading choice (Table 4) increased (L, P<.01) across slaughter group, but the values were not highest for steers fed 147 d; consequently, we detected a Q (P<.02) response. Steers fed for 105 d were lower (P<.05) in both marbling score and percentage of cattle grading U.S. Choice than any other slaughter group. Inversely, the percentage of steers grading U.S. Select and Standard decreased (L; P<.01) with increased time on feed. Steers used by Hicks et al. 1987 were of similar breed-type, weight and slaughter dates; their results are very consistent ours for marbling score and percent U.S. Choice cattle. When marbling scores were adjusted to a mean fat thickness, marbling scores were the greatest (P<.05) for steers fed between 119 and 133 d. Steers in our population, similar to those of Hicks et al. (1987), continued to deposit fat subcutaneously, but did not deposit an increased amount of intramuscular fat after 133 days on feed. When marbling scores were adjusted for s.c. fat thickness, there was no advantage from feeding steers more than 119 d. Thus, steers in both studies may have reached their genetic potential to grade U.S. Choice between 119 and 133 d. This agrees with results of Williams et al. (1989), in which steers fed for 112 d had similar U.S. quality grades as those fed for 140 d.

*Chemical composition of longissimus muscle.* Table 5 illustrates the chemical composition of the LM. Cholesterol concentrations (mg/dL) in plasma increased (L; P<.01) across slaughter group, with steers fed for 105 d having the lowest (P<.05) concentration of any slaughter group. This was similar to the increase (L; P<.01) in the cholesterol concentration (mg/100 g wet tissue) within the LM. Steers being fed for 119 or less had lower (P<.05) amounts of cholesterol in the LM than steers fed for 133 d or more. Total lipid present within

the LM also increased (L; P<.01) across slaughter groups, with steers being fed for 105 d having less (P<.05) LM lipid than any other slaughter group. Regression equations using the percent fat deposited within the LM to determine the cholesterol content have been reported by Rhee et al. (1982) and Tue et al. (1967). Both of those equations predict cholesterol concentrations (mg/ 100g wet tissue) between 15 and 25% higher than we determined in this study. This could be due to decreased precision in the colorimetric assays used in the previous studies. Hoelscher et al. (1988) reported that approximately 90% of the total cholesterol found in adipose tissue was present in the storage fraction, leaving approximately 10% of the total cholesterol in the membrane fraction. Thus, increasing the amount of lipid found in the storage form, as would be the case with increased amounts of marbling, should increase the amounts of cholesterol present within the LM. When quality grade was regressed against cholesterol content of the LM, no significant quality grade by slaughter date interaction was detected. Thus, neither slaughter date or age was a factor in cholesterol deposition of the LM; instead increases in cholesterol content within the LM appear to be caused by a increase in fat content. This contrasts with work by Stromer et al. (1966) and Rhee et al. (1982), where no differences in LM cholesterol concentration were detected between carcasses of different quality grades.

With time on fed, LM moisture content decreased linearly (P<.01) while moisture and protein responded Q (P<.02). If fat replaced the moisture and protein within the LM as slaughter date increased, the amount of cholesterol within the LM would increase. When protein content of the LM was adjusted to a mean s.c. fat thickness, the Q effect of slaughter date disappeared. Hence, for our steers, feeding high concentrate diets for more than 119 d may be detrimental due to increased concentrations of cholesterol.

*Cooking properties of rib steaks.* Although LM area was not altered by slaughter group, raw weight and cooked weight of LM steaks increased (L; P<.01) with slaughter group (Table 6). Without precise control over thickness when cutting steaks, changes in raw and cooked weight of LM steaks may indicate merely that the thickness of steaks varied. However, the percent cooking shrink, a better indicator of relative changes, decreased (L; P<.02) at a decreasing rate (Q; P<.04) across slaughter date. The cooking time (minutes/100 g raw tissue) required to cook LM steaks to a medium degree of doneness also decreased (L; P<.01) with slaughter group.

Tenderness of Ribeye Steaks. As time on feed increases, tenderness increases (Dolezal et al., 1982; Miller et al., 187); however, this increase may be limited with a maximum at 139 d (Epley et al., 1968) or between 150 and 180 d (Zinn et al., 1970b); after this time, the effect of animal age may decrease tenderness. May et al. (1992) fed steers between 0 and 196 d and reported that the lowest shear force value was at 112 d; with shear force values at 28 and 196 d both were greater. Tenderness of the LM in this study, measured by Warner-Bratzler shear force, tended to increase (L; P<.07; Table 6) continuously with increasing days on feed. This contrasts with results of Matthews and Bennett (1962), Moody et al. (1970) and Dinius and Cross (1978) who reported that tenderness, as measured by taste panel or Warner-Bratzler shear force, did not change as time on feed increased. In our study, the percentage of steaks being considered very tender (< 3.86 kg) and tender (3.86 < tender <4.54 kg) where unaffected by time on feed. However, the percentage of steaks being considered tough (> 4.54 kg) decreased (L; P<.03) with increased time on feed; steers fed for 105 d had a higher (P<.05) percentage of tough steaks than steers fed for 147 d. Even though tenderness tended to increase with time on feed, no differences were detected between 119 and 147 days on feed for shear force, or

the percentage of steaks being considered tough. Thus, steers fed 119 d should have been fed long enough to be considered satisfactory in tenderness and palatability as was reported previously by Dolezal et al. (1982).

#### Implications

Many performance and carcass traits increased with increasing time on feed. Steers fed for 119 d had the highest ADG and feed efficiency (carcass adjusted basis) and acceptable levels of s.c. fat, KPH, yield grades and marbling. Concentrations of cholesterol in both plasma and LM increased with longer times on feed. This may be due to the increased amount of fat deposited within the LM. Tenderness of rib steaks also tended to increase with time on feed. With increased consumer demand for high quality but lean beef, feeding cattle more than 119 d may not be advantageous because s.c. fat and cholesterol deposited within the LM increased without improvements in quality grade or tenderness. Although the length of time cattle are kept on feed usually is determined by the feeders desire to maximize profit, effects on quality and acceptability of beef also need to be considered to maintain consumer appeal of beef products.

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	Diet Sequence						
Ingredient	1	2	3	4	Final		
Corn. rolled	40.20	50.20	(%) 60.20	70.20	82.20		
Alfalfa hay, pelleted Cottonseed hulls	25.00 25.00	20.00 20.00	15.00 15.00	10.00 10.00	4.00 4.00		
Molasses, cane Pelleted supplement <sup>a</sup>	4.00 5.80	4.00 5.80	4.00 5.80	4.00 5.80	4.00 5.80		

## Table 1. Composition of diets (dry matter basis)

Calculated Composition:

	Final Diet					
Nutrients	DM %	As Fed %				
Dry matter, % NEm, Mcal/kg NEg, Mcal/kg Crude protein, % K, % Ca, % P, %	100.00 2.09 1.33 11.90 .70 .54 .32	87.80 1.84 1.17 10.45 .62 .48 .28				

<sup>a</sup> Supplement composition: Cottonseed meal, 65.9%; calcium carbonate, 17.1%; urea, 9.49%; salt, 6.04%; dicalcium phosphate, 1.25%; vitamin A, D, E, .20%; Manganese Dioxide .02%.

	Days on feed					Observed Significance (P<)		
	105	119	133	147	SEM	Linear	Quadratic	Cubic
No. of Pens No. of Steers	8 61	8 63	8 64	8 64	-7			
Initial wt., kg Final wt., kg <sup>p</sup>	330 472 <sup>0</sup>	330 501d	329 516 <sup>e</sup>	329 533 <sup>f</sup>	.10 2.28	.01 .01	.44 .03	.77 .16
ADG, kg	1.36 <sup>c</sup>	1.44d	1.41cd	1.41	cd .01	.22	.07	.10
DIM, lb./day	9.92 <sup>C</sup>	10.0 <sup>cd</sup>	10.4d	10.3 <sup>CC</sup>	d.05	.03	.37	.24
Feed/Gain	7.30 <sup>d</sup>	6.97 <sup>C</sup>	7.38d	7.31	80. b	.26	.15	.01
Calc. energy in diet ME, Mcal/kg DM NEm, Mcal/kg DM NEg Mcal/kg DM	2.73 <sup>C</sup> 1.71 <sup>C</sup> 1.12 <sup>C</sup>	2.83d 1.80d 1.19d	2.76 <sup>ce</sup> 1.74 <sup>ce</sup> 1.15 <sup>ce</sup>	2.81 1.78 1.18	de .02 de .02 de .01	.08 .07 .08	.25 .29 .21	.01 .01 .01

#### Table 2. Effects of days on feed on performance of feedlot steers<sup>a</sup>

а

Least squares means; SEM n = 8. Calculated as hot carcass weight/.6495 (average dressing % for all steers). Means within a row with different superscripts differ (P<.05). b

cdef

	Days on Feed					Observed	d Significance	e Level (P<)	
	105	119	133	147	SEM	Linear	Quadratic	Cubic	
No. Pens No. Steers	8 61	8 63	8 64	8 64					
Carcass wt., kg. <sup>b</sup> Dressing, % <sup>C</sup> Dressing, % <sup>cd</sup> REA, cm <sup>2</sup> . S.C. Fat. Thick., cm KPH, % Maturity <sup>e</sup> USDA Yield Grade Percent YG4 Condemned liver, %	3089 65.1 68.7 82.6 1.48 <sup>h</sup> 1389 1389 2.12 <sup>i</sup> 1.56 <sup>h</sup> 12.72	325h 65.3 65.2 83.9 1,61h 145h 1469h 2.36hi 09 10.94	335 <sup>i</sup> 64.8 65.6 85.9 1.09gh 2,039 148h 148h 2.53gh 1.56h 9.38	347J 65.6 61.4 84.8 1.179 1.989 149h 1449h 2.759 9.38h 12.50	3.34 .25 1.37 .19 .02 .05 1.47 2.12 .09 2.76 5.39	.01 .85 .74 .01 .01 .01 .51 .01 .07 .93	.06 .30 .04 .36 .60 .10 .08 .38 .89 .12 .66	.24 .12 .47 .51 .89 .01 .61 .04 .75 .81 .86	

#### Table 3. Effects of slaughter group on carcass characteristics<sup>a</sup>

а

b

С

d

е

f

Least square means; SEM n = 8. Carcass weight adjusted for trimloss. Calculated by dividing final live weight by carcass weight. Adjusted for carcass weight as a covariate. Calculated by averaging lean and skeletal maturity. Adjusted for fat thickness as a covariate. Means within a row with different superscripts differ (P<.05). ghij

	Days on Feed					Observed Significance Level (P<)			
	105	119	133	147	SEM	Linear	Quadratic	Cubic	
No. Pens No. Steers	8 61	8 63	8 64	8 64					
Marbling Score <sup>b</sup> Marbling Score <sup>bc</sup>	377d 391d	433e 442e	452e 436e	446 <sup>e</sup> 410 <sup>d</sup>	11.48 8.16	.01 .21	.02 .37	.82 .73	
Prime, % Choice, % Select, % Standard, %	0 33.93d 52.46d 13.62 <sup>d</sup>	1.79 58.93e 39.29de 0e	0 68.75 <sup>e</sup> 31.25 <sup>e</sup> 0 <sup>e</sup>	0 68.75 <sup>e</sup> 29.69 <sup>e</sup> 1.56 <sup>e</sup>	.89 5.52 4.45 1.47	.67 .01 .01 .01	.34 .05 .22 .01	.21 .83 .95 .10	

#### Table 4. Effects of days on feed on USDA quality grades of steers<sup>a</sup>

a b

С

Least squares means. 300-399, slight; 400-499, small Adjusted for fat thickness as a covariate. Means within a row with different superscripts differ (P<.05). de

······	Days on Feed				Observed Significance Level (P<			
	105	119	133	147	SEM	Linear	Quadratic	Cubic
No. Pens No. Steers	8 61	8 63	8 64	8 64				
<u>Plasma:</u> Cholesterol, mg/dL	96.55 <sup>c</sup>	118.15 <sup>d</sup>	115.74 <sup>d</sup>	123.67d	4.05	.01	.13	.09
Proximate Analysis, % Total Lipid Protein Protein <sup>b</sup> Moisture Cholesterol, mg/100g wet tissue	3.01 <sup>C</sup> 22.70 <sup>C</sup> 22.78 <sup>Cd</sup> 73.64 <sup>C</sup> 47.26 <sup>C</sup>	3.66d 22.44cd 22.38cd 73.23de 47.00 <sup>c</sup>	3.75d 22.12d 22.19c 73.40cd 50.81d	4.00d 22.50 22.94d 72.97e 52.33d	.18 .12 .18 .12 .80	.01 .10 .19 .01	.28 .02 .72 .93 .32	.39 .18 .24 .06 .12

Table 5. Effects of slaughter group on plasma cholesterol and longissimus muscle composition<sup>a</sup>

a b

Least square means; SEM = 8. Adjusted for fat thickness as a covariate. Means within a row with different superscripts differ (P<.05). cde

	Days on Feed					Observed Significance Level (P<)		
	105	119	133	147	SEM	Linear	Quadratic	Cubic
No. Pens No. Steers	8 61	8 63	8 64	8 64		-	~	
Raw weight, g Cooked weight, g Cooking Shrink, % Cooking time, min.	293.78 <sup>e</sup> 206.16 <sup>e</sup> 29.86 <sup>e</sup> 7.63 <sup>e</sup>	294.96 <sup>e</sup> 211.27 <sup>ef</sup> 28.39 <sup>f</sup> 6.91 <sup>f</sup>	331.10 <sup>f</sup> 238.199 28.05 <sup>f</sup> 6.54 <sup>f</sup>	300.83 <sup>e</sup> 215.45 <sup>f</sup> 28.38 <sup>f</sup> 6.64 <sup>f</sup>	2.46 1.87 .37 .23	.01 .01 .02 .01	.01 .01 .04 .11	.01 .01 .79 .93
Shear Force, kg. Very Tender, % <sup>b</sup> Tender, % <sup>c</sup> Tough, % <sup>d</sup>	4.45 29.51 24.59 45.90 <sup>e</sup>	4.42 26.98 38.10 33.33ef	4.11 43.75 26.56 29.69 <sup>ef</sup>	4.16 39.06 32.81 28.13 <sup>f</sup>	.13 6.15 5.18 5.17	.07 .13 .58 .03	.73 .81 .50 .27	.34 .19 .10 .71

#### Table 6. Effects of slaughter group on cooking properties and shear force<sup>a</sup>

а

b c

Least square means; SEM n = 8. Very Tender < 3.86 kg. Tender = 3.86 < Tender < 4.54 kg.

d

Tough > 4.54 kg. Means within a row with different superscripts differ (P<.05). efg

2

#### MICHAEL THOMAS VAN KOEVERING

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#### Doctor of Philosophy

## Thesis: INFLUENCE OF β-HYDROXY-β-METHYL BUTYRATE ON PERFORMANCE, CARCASS QUALITY, LIPID DEPOSITION AND TENDERNESS OF LONGISSIMUS MUSCLES FROM SERIALLY SLAUGHTERED FEEDLOT STEERS

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