

THE EFFECT OF ZILPATEROL HYDROCHLORIDE ON ENERGY AND PROTEIN
METABOLISM AND EMPTY BODY COMPOSITION OF BEEF STEERS

By

LEE-ANNE JUDY WALTER

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ABSTRACT

Two trials (indirect calorimetry and comparative slaughter) were conducted to examine energy and protein metabolism and empty body composition of cattle supplemented zilpaterol hydrochloride (Z). The first trial utilized beef steers ($n=20$; 463 ± 14 kg) blocked ($n=5$) by body weight (BW) and source and adapted to maintenance intake for 21 d prior to Z (90 mg/hd/d) or control (C) treatment for 20 d (455 ± 14 kg at start of treatment). No differences in DMI, apparent nutrient digestibility, O_2 consumption or CH_4 production ($P \geq 0.12$) were detected between treatments but Z cattle had greater CO_2 production during the maintenance period ($P = 0.04$; 2,325 vs. 2,185 L/steer; 23.6 vs. 22.4 L/kg $BW^{0.75}$). Cattle treated with Z tended to have increased heat production ($P = 0.09$; 12.44 vs. 11.69 Mcal, respectively) but not on a $BW^{0.75}$ basis ($P = 0.12$; 0.126 vs. 0.120, respectively) with no treatment difference in fasting heat production ($P \geq 0.32$). Control cattle excreted more ($P = 0.05$) nitrogen in urine (39.8 vs. 32.4 g/d, respectively) whereas cattle fed Z tended to have increased nitrogen retention ($P = 0.07$; 22.14 vs. 14.12 g/d). Supplementation of Z did not improve HCW ($P = 0.12$) but did increase dressed carcass yield ($P = 0.02$; 62.12 vs. 60.65%, respectively) and LM area ($P = 0.02$; 77.81 vs. 70.90 cm^2) while tending to lower USDA calculated yield grade ($P = 0.06$; 1.8 vs. 2.2).

The second trial utilized single-sired, beef steers ($n=56$; initial BW = 590 ± 36 kg) blocked ($n=2$) by BW and terminal implant and further sorted into pairs ($n=14$ per block) by BW. Pairs were assigned to 0, 28 or 56 d of feeding, within 28 and 56 d to

maintenance (M) or ad libitum (A) intake, and within 56d of feeding, steers within a pair were randomly assigned to either 20d of Z supplementation (90 mg/hd/d) with a 4d withdrawal prior to harvest or no Z supplementation (C). Carcass sides were fabricated 48h post-harvest; samples of 9-10-11 rib primals were dissected into lean, fat, and bone with lean and fat ground and sampled for proximate analysis; all long bones were sliced horizontally and sampled for analysis. Treatment impacted ($P \leq 0.05$) live ADG; contrasts indicated A (1.33) was greater than M (0.14 kg/d), and Z (1.12) was greater than C (0.82 kg/d). Similarly, carcass ADG differences ($P < 0.01$) indicated A (1.04) was greater than M (0.36 kg/d), and Z (1.35) was greater than C (0.71 kg/d). Intake altered BW and empty body weight (EBW); M cattle had reduced BW and EBW ($P < 0.01$, 585 and 540 kg) than A cattle (647 and 597 kg). Cattle fed at M had less carcass and internal cavity mass ($P < 0.01$, 359 and 79.4 kg) than A cattle (394 and 93.5 kg). Moreover, mass of total splanchnic tissue was less ($P < 0.01$) for M cattle than A cattle (59.8 vs. 72.5 kg). Dressed carcass yield was greater ($P < 0.01$) for Z than C cattle (63.5 vs. 61.6 %). Cattle fed at M exhibited less 12th rib subcutaneous fat and lower U.S. yield grades than A cattle ($P < 0.01$; M-1.71 cm and 3.3, A-2.46 cm and 4.3) and lower Canadian yield grade ($P < 0.01$; 53.9 vs. 51.9 for M vs. A). Fat content of the EB (EBF) and carcass (CF) was impacted by treatment with 56d AC having greater EBF and CF ($P < 0.05$; 34.1 and 30.2%, respectively) than all other treatments except 28d A. In addition, M steers had less ($P < 0.05$) EBF (30.44 v. 32.30%) and CF (30.29 v. 32.23%) vs. A steers. Moisture of the EB (EBM) and carcass (CM) tended to be impacted by diet ($P < 0.10$) in an inverse fashion to EBF and CF; whereas Z increased ($P < 0.05$) EBF and CM. Protein of the empty body and carcass tended to be affected by treatment on an absolute basis with Z

steers exhibiting increased protein yield vs. C. Additionally, EB moisture, fat and protein daily gains were different ($P < 0.05$) with M decreasing gains of all components vs. A ($P < 0.01$) and Z tending to increase moisture vs. C ($P = 0.06$). Heat production of AZ was greater than AC steers ($P < 0.001$; 0.211 vs. 0.184 Mcal/EBW^{0.75}) whereas MZ did not differ from MC ($P > 0.05$). As a result, efficiency of metabolizable energy intake was 0.499 and 0.293 for C and Z steers respectively.

Results from the first trial indicate that Z treatment tended ($P \geq 0.07$) to increase nitrogen retention and modify heat production during maintenance by increasing CO₂ production. Results from the second trial indicated that days on feed, energy level intake and Z supplementation affected live performance, kill yields, carcass grading factors, empty body and carcass composition and efficiency of gain.

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LIST OF ABBREVIATIONS

ADG	Average daily gain
BW	Body weight
C	Control
CA	Carcass ash
CF	Carcass fat
CM	Carcass moisture
CP	Carcass protein
CR	Carbon retention
CH ₄	Methane
CO ₂	Carbon dioxide
d	Day
DE	Digestible energy
DM	Dry matter
DMI	Dry matter intake
EB	Empty body
EBA	Empty body ash
EBF	Empty body fat
EBM	Empty body moisture
EBP	Empty body protein
Feed efficiency	kg gain:kg feed

g	grams
GE	Gross energy
h	Hour
HCW	Hot carcass weight
IC	Internal cavity
LM area	<i>Longissimus dorsi</i> area
ME	Metabolizable energy
mL	milliliters
N	Nitrogen
NR	Nitrogen retention
NDF	Neutral detergent fiber
NE _m	Net energy maintenance
NE _g	Net energy gain
O ₂	Oxygen
<i>P</i>	Probability
RE	Retained energy
SAS	Statistical analysis system
SD	Standard deviation
SE	Standard error
SC	Subcutaneous
wt	Weight
Z	zilpaterol hydrochloride
%	Percentage

CHAPTER 1

INTRODUCTION

In beef cattle, tissue accretion of fat and protein differ as the animal's physiological age advances as well as with differing plane of nutrition (Koch et al., 1978). While maintenance energy requirements are a direct result of metabolic BW ($BW^{0.75}$; Fox and Black, 1984), gain requirements are a result of the composition of gain, and the proportion of fat and protein tissue accretion (Garrett, 1959). On high energy dense rations, fat accretion of both empty body and carcass occurs late in the growth phase at an accelerated rate (Simpfendorfer et al. 1974, Fox and Black, 1984; Bruns et al., 1992). The increase in fat accretion as a proportion of gain results in a higher energy requirement in the latter phases of the feeding period, resulting in reduced efficiency.

Zilpaterol hydrochloride is a β_2 -adrenergic agonist that binds with high affinity to the predominate number of β_2 -adrenergic agonist receptors present on bovine adipose and muscle tissue (Mersmann, 1998; Johnson et al. 2014). Zilpaterol treatment results in improved live BW, ADG and gain:feed (Montgomery et al., 2008; Elam et al., 2009) while also increasing HCW, LM area, dressed carcass yield, and carcass cutability (Elam et al., 2009; Hilton et al., 2010). Additionally, Z has been shown to promote significant carcass gains relative to live gain (Rathmann et al. 2012) and increase the transfer of non-carcass components to carcass components (McEvers et al. 2013). Due to the repartitioning of nutrients and increased carcass lean muscle mass, it has been speculated that Z has impacts on the energetics of maintenance and gain.

Previous research examining the energetics of wethers fed clenbuterol reported an increase in energy expenditure of 0.6 MJ/d over 20d (MacRae et al., 1987) while ewe lambs fed cimaterol did not increase maintenance energy expenditure after the first six days of treatment (Rikhardsson et al., 1991). Zilpaterol has been shown to increase carcass moisture and protein while decreasing empty body fat in Holstein steers (McEvers et al., 2014). Research with other β -adrenergic agonists including cimaterol and clenbuterol have reported increased protein deposition, decreased fat accretion and increased heat production in the first few days of administration (Ricks et al., 1984; Macrae et al., 1988; Reeds and Mersmann, 1990; Rikhardsson et al., 1991). Therefore, Z may potentially impact heat production and the efficiency of metabolizable energy used for gain by changing the composition of tissue growth. Thus, the objectives of the studies were to determine the impact of Z on maintenance energy requirements and protein turnover, non-carcass and carcass harvest yields, empty body composition and energetics in beef steers.

CHAPTER 2

REVIEW OF LITERATURE

2.1 History of Energetics

Lavoisier was the pioneer of energetics; first recognizing that the oxidization of a substance by the body produced heat. Lavoisier also demonstrated the relationship between oxygen (O₂) use, carbon dioxide (CO₂) and heat production in the late 1700's (Ferrell and Oltjen, 2008). Lavoisier and Laplace, using a guinea pig in a chamber surrounded by ice, determined that the amount of melted ice directly correlated to the exhalation of CO₂ (Brody, 1945). Discovery of thermodynamics and free energy changes in the late 1800's drove further energetics and nutrition knowledge (Ferrell and Oltjen, 2008). Brody (1945) summarized the laws of thermodynamics: *The first law of thermodynamics involves the conservation of energy. Under this law, the total amount of energy in a system remains constant (with all forms measured as heat), with loss in one area resulting in a gain in another area.* Simplistically stated, the energy released from the oxidation of nutrients must equal energy performed as work by the animal, maintenance energy of the animal and the heat increment of feeding (Brody, 1945). *The second law, the principle of conservation of matter states that matter and energy are inseparable and all forms of energy are quantitatively converted to heat.* Energy has the capacity to do work in various forms because reactions flow from high energy to low energy and the total energy of a system is made up of bound and free work (Brody, 1945).

With respect to nutrition, the first law of thermodynamics forms the foundation of energetics ($ME = RE + HE$).

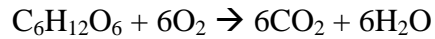
Where: ME = Metabolizable energy consumed by the animal that is not excreted in feces, urine or combustible gases (methane)

RE = retained energy, energy deposited in animal tissues or products

HE = heat energy, heat generated by the animal

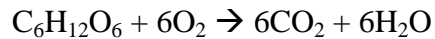
Living organisms employ a large amount of their energy for basal metabolism needs and energy is used in multiple forms within the body. An example of energy use in the body is breakdown of feed into its final productive precursors to be used as nutrients or for temporary storage (Brody, 1945). The second use of energy in the body is the work employed by utilizing precursor nutrients into living organisms or growth (Brody, 1945). In this sense, energy employed can be potential (ex. muscular contraction and relaxation) or energy can be lost as entropy (Brody, 1945). Under the first law of thermodynamics, so long as two of the above variables are measured, the third can be calculated by difference (Ferrell and Oltjen, 2008). The second law of thermodynamics (all forms of energy can be converted to heat) applies to the principle of a bomb calorimeter (Ferrell and Oltjen, 2008). Therefore a bomb calorimeter measures the change in heat of a substance. Under the second law of thermodynamics, the breakdown of carbon molecules under specific conditions results in consistent and measurable changes in energy based on composition of the molecule, bond length and bond angles of the molecule and degree of double bonding. Hess' Law states that bond energies holding the atoms of the molecule together are equal to the heat of dissociation of a molecule into its

atoms. For example, one mole of glucose oxidized by O₂ releases water and CO₂ producing 688 kcal of energy (Blaxter, 1962):



$$\Delta F^\circ = -688 \text{ kcal/mole (releases energy)}$$

Under standard biological conditions, the oxidation of glucose also releases entropy and therefore 673 kcal/mole is released:



$$\Delta H^\circ = -673 \text{ kcal/mole (releases energy)}$$

In the case of glucose, 15 kcal/mole is released as entropy. When examining biological systems, energy taken in by the animal cannot be defined in terms of the potential useful energy on complete oxidation but rather total heat produced on complete oxidation (ΔH):

$$\Delta H = \Delta F + T\Delta S \text{ where } T = \text{absolute temperature and } \Delta S = \text{entropy}$$

2.2 Direct Calorimetry

Early calorimeters directly measured the heat produced by an animal, employing the same general principle as a bomb calorimeter. In this sense, heat production increased the temperature of the surrounding medium of the chamber the animal was enclosed in and in the case of the original Lavoisier model, the amount of ice melted \times latent heat of ice was equal to the heat produced by the animal (Blaxter, 1962). The direct calorimetry system evolved to include animals housed in a double walled enclosure with inner copper walls acting to transfer heat produced by animals into water circulating in copper pipes within the surrounding area. The temperature of the outer wall had to be tightly controlled so that no heat escaped from the system and so that no ambient heat was placed on the system (Blaxter, 1962). All heat therefore was captured by the

circulated water pipes. The heat production by the animal inside the enclosure was calculated by the weight of the water circulating pipes per unit of time multiplied by the rise in the water temperature, which gave heat loss by the animal with respect to radiation and convection (Blaxter, 1962). Heat loss due to vaporization by the animal was accounted for by multiplying the weight of water vapor by the latent heat of vaporization (Blaxter, 1962). Early direct calorimetry chambers for animals employing this design were based off the Atwater-Rosa adiabatic calorimeter for man (Wesleyan University) and included Armsby and Fries in 1903 (Pennsylvania State University) and Capstick and Wood in 1920 (Cambridge University).

Gradient layer direct calorimetry chambers consisted of an inner shell that housed the animal surrounded by a narrow air space and were originally designed by Richet and Rubner in 1889. The inner air space was surrounded by an insulating layer that was separated from an outside layer by another insulating layer. The principle of the second generation calorimetry chambers was the average temperature gradient between the inner and outer surfaces of the standard thermal material separating the two surfaces is proportional to the total loss or gain of heat from a source within the cavity (Blaxter, 1962). All air spaces were hermetically sealed with differences in pressure between the inner and outer air spaces proportional to the gradient of temperature differences over the insulating layer (Blaxter, 1962). The system was slow to respond to any acute changes in heat production (Blaxter, 1962). Benzinger advanced the design of gradient layer calorimeters. In his system, heat is allowed to flow through the containing wall as walls with known thermal conductivity and uniform thickness (Blaxter, 1962). This method required numerous thermocouples (~6000/enclosure) laced together throughout the

system with minute by minute monitoring. In addition, incoming air had to be cooled to a precise temperature and saturated with water vapor to standardize temperature and pressure (Blaxter, 1962). Plate meters at the entrance and exit to the chamber measured air temperature and the difference in temperature resulted from heat production by the animal (Blaxter, 1962). A source of discontention with this method was the absorption of heat by feed and water buckets and the flooring material and thus incomplete capture of heat production by the animal.

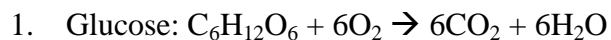
2.3 Indirect Calorimetry

Modern, indirect calorimetry chambers employ open circuits. Outdoor air is passed through the chamber of the instrument and changes in O₂, CO₂ and methane content between incoming outdoor air and outgoing chamber air are measured. Therefore, the amount of air which passes through the chamber and the incremental changes in gas concentrations are quantified (Blaxter, 1962). Specifically, total amount of CO₂ and methane produced and O₂ consumed are determined. Armsby et al. (1920) first recorded CO₂ production and the ration to direct heat production deriving a relationship of: $\{\text{Heat} / \text{CO}_2 = -(0.0226 * (\text{DMI} / \text{BW})) + 2.802\}$. As the open calorimetry circuit came into operation, potential technical issues came to light. The volume of air passing through the chamber must be measured very precisely and a very true sample of incoming and outgoing air also must be obtained and measured (Blaxter, 1962). Thus, a decrease in O₂ content and a subsequent increase in CO₂ and methane passing through the chamber must be precisely measured (Blaxter, 1962). To measure changes in CO₂, methane and O₂ content of air, ventilation is often adjusted so CO₂ content is ~1% of the air. At this level of CO₂ in the chamber, the accuracy of measuring

the composition of gases must be detectable to 0.007% (Blaxter, 1962). Therefore, less ventilation equates to greater changes in CO₂, O₂ and methane content of outgoing air.

Indirect calorimetry employs indirect estimation of heat from chemical bond energies. The primary assumption is that chemical bond energies equate to the heat of dissociation for a molecule of fat, protein or carbohydrate (Blaxter, 1962). Therefore, estimation from individual chemical bonds are used to calculate overall heat of formation and heat of combustion of the molecule in which the individual chemical bonds are contained (Blaxter, 1962). Unfortunately, this method does not take into account differences in physical chemistry (lengths and angles of chemical bonds), nor complex structures such as benzene rings, carboxyl groups or amide groups. Additionally, energy of the feed is based upon the content of fat, protein and carbohydrate percentage with palmitic acid, alanine and glucose bond energies used as the standard for fats, amino acids and carbohydrates to derive actual energy and therefore do not account for known differences between different amino acids, lipids and carbohydrates.

The body oxidizes three major classes of compounds: carbohydrates, fatty acids and proteins. Heat of combustion calculations are based on the amounts of O₂ consumed and the amounts of CO₂ and nitrogen excreted. Therefore, any of the three classes of compounds for energy in a dietary mixture can be oxidized for energy by the animal (Blaxter, 1962):

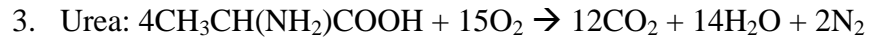


$$\Delta H^\circ = -673 \text{ kcal/mole (releases energy)}$$



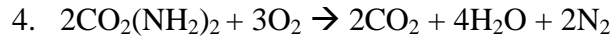
$$\Delta H^\circ = -2398.4 \text{ kcal/mole (releases energy)}$$

Amino acids increase in complexity because nitrogen is not completely oxidized but rather excreted as urea (Blaxter, 1962).



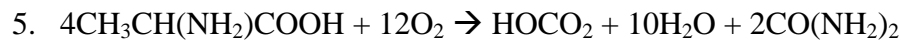
$$\Delta H^\circ = -387.7 \text{ kcal/mole (releases energy)}$$

Urea on complete oxidation:



$$\Delta H^\circ = -151.6 \text{ kcal/mole (releases energy)}$$

Subtraction derives heat of oxidation of alanine to CO_2 , water and urea



$$\Delta H^\circ/\text{mole alanine} = -311.9 \text{ kcal/mole (releases energy)}$$

The respiratory quotient (RQ) is the molar ratio of CO_2 produced to O_2 consumed and in the case of pure glucose $\text{RQ} = 6/6 = 1.0$ whereas palmitic acid has a $\text{RQ} = 16/23 = 0.7$ (Blaxter, 1962). The RQ is an indirect measure from the nutrient classification of feed rather than physical chemistry bond energies (Blaxter, 1962). A source of error is the wide variation of complex molecules with differing bonds and potential energy within the bond types and how to account for the wide variation with only O_2 consumption and CO_2 production (Blaxter, 1962). As an example, xylose has a determined heat of combustion of 561.5 kcal/mole using a bomb calorimeter whereas its calculated heat of combustion (using molar RQ ratio), is 560.8 kcal/mole. The dichotomy increases in the case of polysaccharides (glycogen, starch and cellulose) because calculated heat of combustion differs from determined heat of combustion (bomb) by 6% (Blaxter, 1962). The opposite is true in the case of short chain fatty acids where the calculated heat of combustion is overestimated by 2% relative to the actual heat of combustion (Blaxter,

1962). With respect to amino acids, the heat of combustion is again overestimated by 4% for alanine (Blaxter, 1962). A correction factor for nitrogen content of the compound is applied which also imposes a degree of error as an average nitrogen correction factor will overestimate heat from some amino acids and underestimate heat from other amino acids. An additional source of error arises due to incomplete oxidation of carbohydrates in ruminants resulting in methane production. Methane is produced during carbohydrate oxidation in ruminants and thus an energy correction factor of 0.5 kcal/L of methane produced is applied to derive ME. Ultimately, the physical chemistry of specific models is more complex than indirect calorimetry accounts for albeit, complex feed molecules often do not form major amounts of the diet. In order to minimize errors, the appropriate reference substance should be used to calculate the heat of combustion (Blaxter, 1962). In the case of measuring energy use in suckling calves, lactate needs to be used as the carbohydrate reference substance vs. glucose (Blaxter, 1962). Starch, cellulose, mixed fatty acids and intact proteins are commonly used as the reference material to derive the calculated heat of combustion in finishing cattle.

Multiple equations have been developed to estimate heat production via the respiratory quotient method and indirect calorimetry. Forbes et al. (1927) directly compared heat production of cattle with respiratory-quotient (RQ); they reported 0.707 as the RQ for fat or 4.686 calories per liter of CO₂ and the RQ for carbohydrates as 1.000 or 5.047 calories per liter of CO₂. The equation to estimate heat produced via indirect calorimetry proposed by Brouwer (1965) and has remained unchanged to date (Ferrell and Olten, 1998):

$$\text{Heat produced (kcal)} = 3.866 * \text{O}_2 \text{ consumed (L)} + 1.200 * \text{CO}_2 \text{ produced (L)} - 1.431 * \text{N excretion in urine (g)} - 0.518 * \text{CH}_4 \text{ produced (L)}$$

2.3.1 Measuring Heat Production

To measure fasting heat production (FHP), an acclimation period to a maintenance level of feed intake is given for least three weeks with animals trained to the calorimeter and kept in a thermoneutral environment (NRC, 2000). Measurements for FHP are made during the third and fourth day after feed withdrawal with methane values used as an index of post-absorptive state (Blaxter, 1962). Fasting values are adjusted for differences between fasted weight and live weight when fed and adjusted for activity allowance of 1kcal/kg of animal live weight (NRC, 2000). Measurements are repeated when cattle are fed maintenance and *ad libitum* intakes. Heat production at maintenance and *ad libitum* feed intake accounts for the heat of combustion of absorbed nutrients and the overall heat of the reaction (Blaxter, 1962). As a consequence, the heat of combustion from body constituents stored or lost can be estimated as the difference (ME – HP = RE; Blaxter, 1962) following the second law of thermodynamics. The heat produced above FHP results in a loss of energy known as the heat increment which increases as feed intake increases from maintenance to *ad libitum* (Blaxter, 1962).

2.3.2 Additional Benefits and Issues of Indirect Calorimetry

Indirect calorimetry methods are able to provide fasting heat, maintenance and retained energy estimations for the animal. Once the system is established, multiple animals can be put through the system with little additional cost for labor and system upkeep. Disadvantages of the indirect calorimetry system include a high initial capital investment as well as the novel lab environment with the animal not able to exercise,

withstand outside elements or to socialize, therefore not exhibiting normal behaviors nor exposed to dynamic production environments (NRC, 2000). This physiological stress resulting from exposure to novel environment can result in poor intake in the chambers and will also result in a lower fasting heat coefficient vs. animals in their natural environment (NRC, 2000). Other issues are that the number of animals used is very limited and therefore it is difficult to account for the wide variation of cohorts in contemporary production (Blaxter, 1962), while the measurements represent only a very acute time period (NRC, 2000). While indirect calorimetry methods also measure respiration coefficients when fed at maintenance and *ad libitum* intakes, getting the animal to eat at true *ad libitum* levels of intake requires highly trained, calm animals and a perfect scenario. Thus, increased numbers of statistical repetitions or a powerful experimental design such as a repeated latin square is required to overcome individual animal variation within treatment.

2.4 Comparative Slaughter

Comparative slaughter differs from indirect calorimetry because it measures metabolizable energy intake in and retained energy with heat production measured by difference (NRC, 2000). Retained energy is measured as the change in body energy after removal of digestive contents (Blaxter, 1962). In order to perform a comparative slaughter trial, animals need to be fed at two or more levels of intake, one level which approximates maintenance of body weight (NRC, 2000). The goal is to create a cluster of retained energy data around maintenance level of intake and a cluster of data around *ad libitum* level of data (Ferrell and Oltjen, 2008).

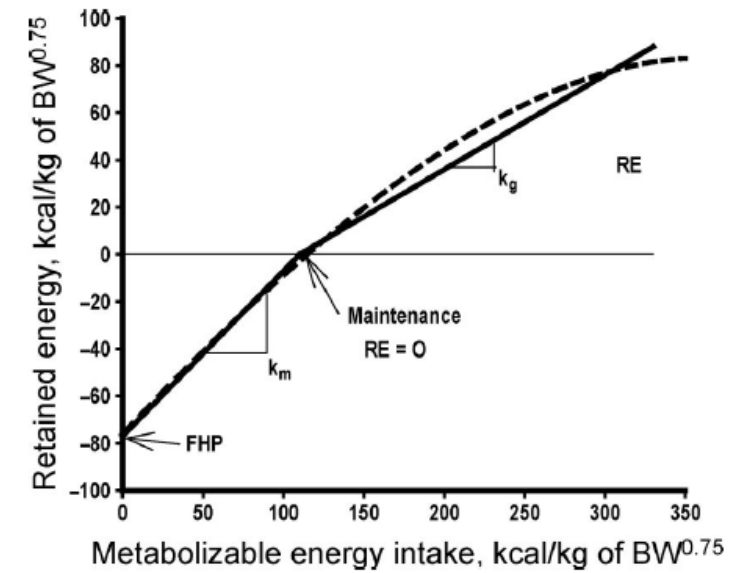


Figure 1.1 Representation of the relationship between retained energy (RE) and ME.

Source: Ferrell and Oltjen (2008)

As summarized by NRC (2000), feeding animals at two or more levels of intake and slaughtering a contemporary group at the start of trial allows for calculations of:

- Retained energy = net energy used for gain
 - Regressing retained energy on metabolizable intake provides a slope. The slope is an estimate of the efficiency of utilization of metabolizable energy for retained energy = k_g (Figure 1).
- The metabolizable intake at which retained energy is equal to zero provides an estimate of the metabolizable energy required for maintenance.
 - Intercept of the regression of logHE on metabolizable intake is used to calculate an estimate of fasting heat production which equates to net energy for maintenance NE_m

- Efficiency of utilization of metabolizable energy for maintenance (k_m)
(Figure 1) is calculated as the ratio of NE_m to ME_m (metabolizable intake at which retained energy is equal to zero).
- Therefore $NE_m = 0.077 * Mcal/EBW^{0.75}$

2.4.1 Additional Benefits & Issues of Comparative Slaughter

The comparative slaughter method requires a large number of animals and a great expense due to grinding of all empty body tissues. In the case of comparative slaughter, the fasting heat production is an extrapolation of the maintenance energy to the y-intercept (Lofgreen and Garrett, 1968). Cattle need to be fed at both maintenance and *ad libitum* levels of intake to discern efficiency of gain. In addition, the comparative slaughter method assumes cattle replicates are essentially homologous prior to the trial start. Body composition differences between cattle at the start can lead to major issues later in the trial that could be counted as treatment differences and lead to misleading results. The major benefit of comparative slaughter allows for separate estimations of energy used for maintenance and energy used for gain because the efficiency of metabolizable energy under these two systems is different. Although, a major issue of the comparative slaughter system is the assumption that efficiency for gain remains linear this relationship is likely curvilinear at higher intakes of metabolizable energy (Ferrell and Oltjen, 2008).

2.5 Feedstuff Evaluation

Feedstuff evaluation is based upon relative nutrient value of feeds. Relative nutrient value of feeds is vast due to a number of different assessments which derive nutrient composition and digestion coefficients (i.e., TDN system, Kellner's starch

equivalent system, Modified starch equivalent system, Estimated Net Energy system; Blaxter, 1956). Blaxter (1956) defined the nutritive value of a food as a biological measurement and thus “*a measure of its ability to promote or sustain some group of metabolic activities in the animal body*”. Therefore, merely deducing the nutritive value of a feed from its chemical composition is a gross overlook of the biological metabolism that may occur to the feed (Blaxter, 1956).

From an energetic standpoint, feed value is derived from the ability of the feed to promote energy retention or prevent energy loss. In order to measure retained measurements of body tissues, live gain has long been used as an indirect measure of energy retention whereas comparative slaughter can be used for a more direct measurement. For determination of nutritive value, BW maintenance is often chosen with feed consumed above maintenance recorded and retained weight gain established (Blaxter, 1956). In the 1950's, research was delving into estimating the changes in fat and protein content of animals indirectly in order to improve the accuracy of indirect energy retention associated with weight gain, etc. Blaxter (1956) summarized data showing energy retention when cattle were given supplements of digestible starch, fat or protein in 1 kg increments above maintenance. In cattle, 1 kg of digestible starch (4,185 kcal) resulted in 2,360 kcal of retained energy; 1 kg of digestible fat (9,500 kcal) resulted in 5,700 kcal of retained energy and 1 kg of digestible protein (5,710 kcal) resulted in 2,220 kcal of retained energy (Blaxter, 1956). This methodology does not account for associative effects of multiple nutrients or feedstuffs nor the curvilinear function of increased metabolizable intake and diminishing increasing energy retention (Blaxter, 1956). The current feedstuff system is still based on TDN equivalency and thus measures

the nutrient composition of the feed vs. observed animal performance (Blaxter, 1956).

As summarized by NRC (1984), conversion of 1 kg of TDN = 4.4 Mcal DE with the conversion of DE to ME = $0.82 \times \text{DE}$. Relationships to derive NE_m and NE_g of the feed are still those derived from Garrett (1980) where $\{\text{NE}_m = 1.37 \times \text{ME} - 0.138 \times \text{ME}^2 + 0.0105 \text{ME}^3 - 1.12\}$ and $\{\text{NE}_g = 1.42 \times \text{ME} - 0.174 \text{ME}^2 + 0.0122 \times \text{ME}^3 - 1.65\}$ (NRC, 2000).

2.6 Basal Energy Metabolism

Basal metabolism has been reported to be similar across animals and species when expressed on a body surface basis ($\text{BW}^{0.73}$; Brody, 1945) or body mass ($\text{BW}^{0.75}$; Kleiber, 1961). Armsby et al. (1918) demonstrated that cattle, horse and man at rest averaged 964, 948 and 935 average basal calorie expenditure at rest on a body surface basis. Brody (1945) estimated mean fasting heat production to be 70.5 kcal/kg $\text{BW}^{0.73}$ over a 24 h time period. Blaxter and Wainman (1966) fasted Ayshire steers for 122 h and reported HP on the 4th day to be 77 – 81.6% of HP on the first day across steers weighing 50 to 400 kg. Logarithmic regression on five Ayshire steers resulted in 24h fasting metabolisms of 72.9 to 106.1 kcal/kg $\text{BW}^{0.73}$ or 64.5 – 97.7 kcal/kg $\text{BW}^{0.75}$ (Blaxter and Wainman, 1966). When FHP was measured on eight Angus steers, the average FHP was 81.0 ± 1.5 kcal/kg $\text{BW}^{0.73}$ or 72.4 ± 1.5 kcal/kg $\text{BW}^{0.75}$ (Blaxter and Wainman, 1966). The currently accepted FHP of cattle is calculated from Lofgreen and Garrett (1968) where HP was calculated as the difference between metabolizable energy intake and retained energy using comparative slaughter data of 208 animals. Derived HP of cattle fed to maintenance and multiple of maintenance on a metabolic BW basis ($\text{BW}^{0.75}$) was then log transformed and regressed on daily metabolizable energy intake on $\text{BW}^{0.75}$ with FHP

calculated as the extension of the line to 0 intake resulting in FHP of 77 kcal/kg BW^{0.75} (Figure 2).

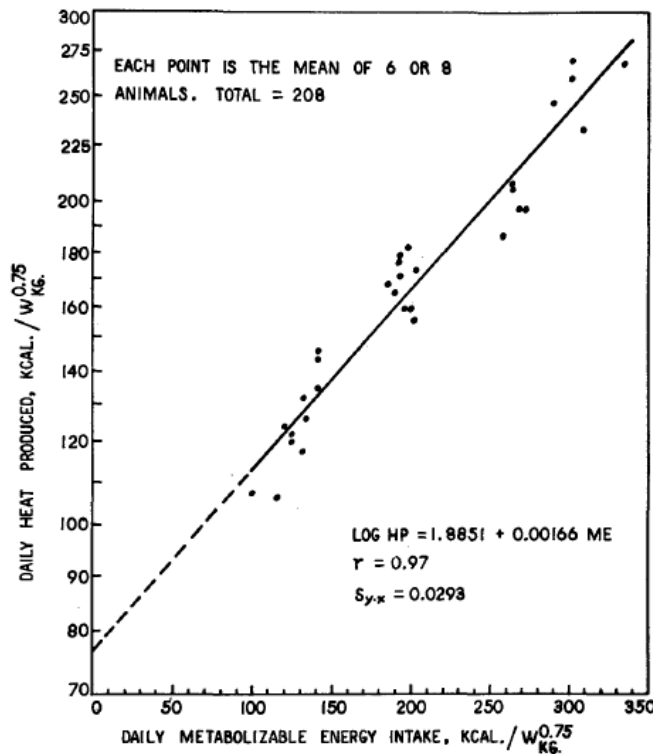


Figure 2. Determination of Fasting Heat Production with Comparative Slaughter.

Source: Lofgreen and Garrett, (1968).

2.6.1 Basal Energy Metabolism Differences Between Animals

Efficiency between animals is important not only from a production stand-point but also from a nutritional comparison stand-point. Trials involving energetics and nutritive feed value are the result of the chemical composition of the feed as well as the innate metabolic efficiency of each individual animal (Blaxter, 1956). Early work in nutrition and energetics focused on using only the most efficient animals or using a wide variety of animals in large numbers to deduce conclusions on the nutritive value of feed (Blaxter, 1956). It was also noted early on by Blaxter (1956) that the physiological

function that the nutrients are to be used for results in differences between feeds, resulting in higher feed energy values for growing vs. fattening.

Considerable variation in metabolizable energy (ME) used for maintenance exists within species. Maintenance energy expenditures vary with BW, breed, genotype, sex, age, season, temperature, physiological state and previous nutrition (NRC, 1984).

Assuming that all of these factors are constant there are a multitude of inherent factors that may cause inefficiencies although finite answers are still largely unknownst.

Splanchnic tissue and basal cellular metabolism are likely causes of the variation in ME required for maintenance while holding all other factors constant.

Contributors to ME expenditure include physical activity, thermoregulation and essential metabolic processes. Essential metabolic processes include sodium/potassium pump and ATP needed to drive the pump (Na^+/K^+ -ATPase, Figure 3) to maintain cellular concentration gradients, protein turnover (protein synthesis and degradation), urea synthesis and substrate cycles. Splanchnic tissue in ruminants is responsible for diet digestion, nutrient absorption and metabolism, maintenance of epithelial structure and immune functions, waste and toxin management and synthetic processes (Reynolds, 2002); therefore, the splanchnic tissue metabolizes a very large quantity of available nutrients (McBride and Kelly, 1990). The splanchnic tissues have been used to measure whole body O_2 use with total estimates of 22.8-40% (Huntington and Tyrrell, 1985; McBride and Kelly, 1990). Therefore, the splanchnic tissue uses disproportionately high fractions of whole body energy utilization.

2.6.1.1 Na⁺/K⁺ -ATPase Pump

The sodium/potassium pump has been estimated to cost 40% of metabolizable energy requirements for cell homeostasis. The action of the Na⁺/K⁺-ATPase results in the most costly energetic process of the gut (McBride and Kelly, 1990). With respect to the rumen, Na⁺/K⁺-ATPase pumps follow a diurnal daily pattern associated with feeding (McBride and Kelly, 1990) whereas Na⁺/K⁺-ATPase activity in the small intestine is associated with nutrient uptake as well as maintenance of ionic homeostasis (Milligan and McBride, 1985). McBride and Milligan (1985) tabulated that the energy cost for maintenance of Na⁺/K⁺-ATPase of the intestinal mucosa accounted for 28.5 to 61% of total gut O₂ consumption and 28.6 to 51.0% of hepatocyte O₂ consumption. Energy expenditure by the Na⁺/K⁺-ATPase pump in the rumen depends greatly upon intake level and energy concentration of the diet whereas substrate availability in vitro (i.e., glucose vs. acetate or level of crude protein) did not impact the energy expenditure of the Na⁺/K⁺-ATPase pump in the rumen nor the small intestine (McBride and Kelly, 1990). Therefore, substrate availability and concentration does not impact the amount of energy used by the Na⁺/K⁺ -ATPase pump in the rumen and small intestine.

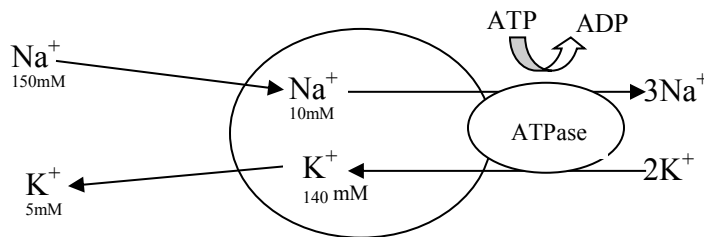


Figure 2.3. Sodium/Potassium pump detailing extracellular and intracellular concentration gradients

Interestingly, physiological state causes dramatic changes in visceral energy expenditure. Lactation has been shown to increase duodenal enzyme activity and small intestine weight (McBride and Milligan, 1984) whereas increases in liver, heart and intestine size was shown in lactating rats and explained 24% of the increase in energy expenditure (Canas et al. 1982). Thus, conversely a reduction in feed intake or an administration of a β -adrenergic agonist known to decrease gut mass would lead to a probable reduction in maintenance energy expenditure by the gut and subsequently the entire body. With respect to the liver Na^+/K^+ -ATPase activity increases with doses of growth hormone and hyperthyroidism status (McBride and Kelly, 1990).

2.6.1.2 Protein Turnover

Protein synthesis is also a very costly metabolic process. Protein synthesis requires 4 mmol of ATP per mmol of amino acid incorporated into the polypeptide chain and 1 mmol ATP per mmol amino acid transported into cell due to active transport by the sodium pump (Blaxter, 1962). Therefore, 5.4-7.2 MJ of energy is needed for every kg of protein synthesized; however, this depends upon type of protein to a certain degree. The ruminant gut has a fractional rate of protein synthesis of 10 to 30%/d which translates to 27.6 to 45.9% of whole body protein synthesis (Lobley et al., 1980). The small intestine represents the largest amount of energy needed for protein synthesis due to proliferation and secretory needs (Lobley et al., 1980); whereas, skeletal muscle which comprises 50% of total body protein, merely accounts for 25% of whole body protein synthesis (Reeds, 1987). Live protein synthesis accounts for 26.5% of total hepatic energy utilization (Reeds, 1987) and 4.9% of whole body ATP use (McBride and Kelly, 1990). Therefore splanchnic protein synthesis is a major encumbrance of maintenance energy.

Protein turnover is the summation of protein degradation and protein synthesis. Protein degradation requires 1 mmol ATP for every peptide bond that is degraded and accounts for 16.6% of gut energy utilization and 3.4% of hepatocyte energy utilization (McBride and Kelly, 1990). Therefore extensive energy is needed to drive daily protein synthesis and degradation needs with 4.9 to 5.5% of whole body energy expenditure required for gut protein turnover (McBride and Kelly, 1990).

Additionally, specific hormones can impact splanchnic protein synthesis. These hormones among others include insulin and insulin like growth factor-1 (IGF-1; McBride and Kelly, 1990). The small intestine has IGF-1 receptors particularly on the crypt cells (McBride and Kelly, 1990). This could indicate a plausible additional action of IGF-1 to stimulate intestinal villi growth and enhance nutrient uptake in animals actively secreting IGF-1 and in essence growing and incurring a positive energy balance.

2.6.1.3 Substrate and Urea Cycling

Futile substrate cycling in the liver is also a large user of ME. This includes the conversion of glucose to glucose 6-phosphate, acetate to acetyl-coA and fructose to fructose 1,6 bis-phosphate all at a net loss of ATP (McBride and Kelly, 1990). The purpose of substrate cycling may allow the animal to react quickly to stimuli. As a result, substrate cycling accounts for 18 to 30% of hepatic O₂ consumption and 3.6 to 6% of whole body O₂ consumption (McBride and Kelly, 1990).

The urea cycle also is a very costly energetic process in the body requiring large amounts of ATP. Urea synthesis in the liver requires four moles of ATP per mole of urea (McBride and Kelly, 1990). The amount of urea produced is highly dependent upon diet and the relative degradation of carbohydrates and protein in the rumen. Spillage of

ammonia will ultimately lead to a greater need for urea synthesis. Urea synthesis is estimated to cost 7.1% of liver O₂ consumption and 1.4% of whole body O₂ consumption (McBride and Kelly, 1990).

2.6.1.4 Overview of Energetic Efficiency

The gut and liver account for 40% of whole body O₂ consumption or 40% of energy needs. Within that, protein turnover (9-12%), ion Transport (Na⁺/K⁺-ATPase pump; 9%), futile substrate cycles (2-4%) and urea synthesis (1-2%) are the major draws to whole body O₂ consumption (McBride and Kelly, 1990). Dry matter intake has been the primary variable responsible for increased size of the splanchnic tissues and thus their energy demand (Reynolds, 2002). Dietary substrates absorbed by the gut and liver are exposed to the tissue's own catabolism and use and therefore future research should examine how to efficiently "feed the gut" (Reynolds, 2002). In addition, cell proliferation of the splanchnic tissues has been shown to be impacted by growth hormone and IGF-1 and therefore a modulation of these hormones could impact the energy expenditure of these tissues (McBride and Kelly, 1990). Therefore, any inherent ability to make these tissues more efficient could account for a major energetic difference between animals.

2.7 Animal Growth

Sex, breed, environment, growth promotant products, prior nutrition and energy density of the ration and age of the animal can all impact growth. Prior et al. (1977) reported that large-framed cattle (Charolais x Chianina) had increased carcass weights and increased estimated retail product and carcass protein when slaughtered at an equivalent age as small-framed cattle (Angus x Hereford). Old and Garrett (1987)

reported Charolais steers used ME less efficiently vs. Hereford steers when fed *ad libitum* and slaughtered at a constant time. The difference between breeds is likely the result of the composition of gain with Charolais steers reporting 43% and 14% fat and protein gain while Hereford steers had 46.2 and 12.3% fat and protein gain, respectively (Old and Garrett, 1987). With respect to the efficiency of gain, the composition of gain is very important because fat deposition has a high efficiency of 60 to 80% whereas protein deposition has a low efficiency of 10 to 40% (Garrett, 1980b). As described by Old and Garrett (1987), protein synthesis and protein accretion do not result in the same energetic efficiency because protein turnover is not accounted for in simple synthesis models. Garrett (1979) observed that increasing concentrate:roughage resulted in linear increases in retained energy from 3.51 Mcal/d for a 90:10 (roughage:concentrate) to 6.38 Mcal/d for a 10:90 (roughage:concentrate). Fox and Black (1984) proposed an equivalent frame size basis for comparing between different breeds to standardize gain requirements with an additional 12% adjustment for Holsteins for total NE requirements. Prior nutritional plane was accounted for by body condition score (scale of 1 to 9) with thin body condition increasing feed NE_m by 4.5% and feed NE_g by 10% (Fox and Black, 1984).

2.8 Improving Animal Efficiency with Growth Modifiers

2.8.1 Estradiol and Trenbolone Acetate

In commercial beef systems in the US, exogenous steroids are subcutaneously placed in the caudal aspect of the ear with a high concentration of compound in a lactose or cholesterol base. Payout of implants is a function of the absorption of the physical implant itself because the implant dissolves over time, yet results in a rapid increase of hormone 24-48 hours post administration with a slow decline in concentration over the

next 50-220 days, depending upon the implant type (Anderson and Johnson, 2003). Combination implants result in longer circulating estradiol concentrations and greater circulating trenbolone acetate concentrations (Trenkle, 1987). Once the hormone is circulated, receptors for estrogenic and androgenic hormones exist on multiple cell types but differ in proportions and binding affinities (Anderson and Johnson, 2003).

The cascade effect for direct action of steroid hormones involves the hormone receptor proteins acting as transcription factors. Lipophilic steroid hormones gain entry into cells by simple diffusion and subsequent binding to receptors (Anderson and Johnson, 2003). Estrogenic and androgenic receptors are located in the nuclear regions of the cell and following ligand binding the ligand-receptor complex binds to DNA sequences (hormone response elements) on target genes (Anderson and Johnson, 2003). At this point, binding to DNA sequences upregulates transcription or downregulates transcription (Anderson and Johnson, 2003). The binding of receptors increases transcription enhancer leading to the transcription of specific mRNA sequences and increases in translation leading to increases in protein synthesis (Anderson and Johnson, 2003). Unlike the standard genomic steroid action (gene transcription up/down regulation), non-genomic mechanisms of steroid hormones have been proposed, including the activation of secondary intracellular messenger such as cAMP (Anderson and Johnson, 2003).

Estrogens primarily act through indirect effects via modulation of the somatotrophic axis. Upon stimulation by estrogen, the pituitary gland releases somatotropin through an increase in the size and cell numbers of the pituitary, increased proportion of somatotropin producing cells and altered pattern of somatotropin secretion

(more frequent peaks and greater amplitude) (Anderson and Johnson, 2003). Changes that ensue are increased sensitivity of somatotropin receptors to somatotropin, decreased somatostatin, increased IGF-1 secretion and increased insulin activity (Frey et al., 1995; Kamanga-Sollo et al., 2004). Then, IGF-1 acts on receptors on muscle tissue where it is internalized leading to increases in cellular level transcription and translation and thus an increase in protein synthesis (Johnson et al., 1996; Kamanga-Sollo et al., 2004). This results in a net effect of increased muscle protein deposition via increased protein synthesis (Johnson et al., 1996). Ultimately, steers treated with combination implants exhibit increased ADG (20 – 25%); gain:feed (15 – 20%) and LM area (Johnson et al., 1996).

Somatotropin is a critical hormone responsible for growth of bone and muscle tissue while inhibiting adipose tissue growth (Beermann, 2003). As summarized by Beermann et al. (2003) somatotropin is very important in the stimulation of skeletal muscle cell growth while inhibiting adipocyte uptake of glucose and fatty acids, thereby decreasing triglyceride formation. Postnatal, somatotropin increases lipolysis and increases blood glucose via direct anti-insulin effects. Somatotropin also stimulates the release of IGF-1 which in turn stimulates chondrogenesis (bone cell formation) and satellite cell division and muscle cell hypertrophy resulting in increased muscle mass (Beermann, 2003). Growth-hormone releasing hormone stimulates somatotropin release through a G-protein-coupled receptor that increases cyclic adenosine monophosphate (cAMP) which in turn stimulates somatotropin release (Beermann, 2003).

With respect to androgenic hormones, trenbolone primarily works through direct actions on cells. Trenbolone acts directly on skeletal muscle cells, stimulating muscle

protein synthesis and deposition (Anderson and Johnson, 2003) through genomic modulation, as previously discussed. Briefly, once androgens are bound to target cell surface receptors they bind to specific nucleotide sequences on chromosomes, in turn promoting transcriptional activity in the cell nucleus and increased protein synthesis (Anderson and Johnson, 2003). Androgens also have indirect effects via glucocorticoid function which influences the balance of protein synthesis vs. degradation leading to a decrease in protein degradation and an overall increase in protein synthesis (Anderson and Johnson, 2003). Trenbolone acetate is a synthetic androgen steroid which is deacylated in blood to trenbolone. Trenbolone acetate results in increased protein synthesis and decreased protein catabolism as it hinders corticotropin-induced stimulation of cortisol production by competing for binding sites on cortisol binding proteins (Anderson and Johnson, 2003). This leads to a hypertrophic response in muscle cells. Research by Chung and Johnson (1998) reported that indirect effects of anabolic steroids in steers increases satellite cell proliferation and therefore provides additional nuclei to support muscle hypertrophy. Combinations of estrogen and androgen implants have the potential for additive effects. Ultimately they increase somatotropin release, decrease cortisol function, increase protein synthesis and decrease protein degradation resulting in a net gain in protein accretion.

2.8.1.1 Benefits and Issues of Growth Implants

Cattle administered a growth implant will deposit more lean tissue and less fat at any time during their growth phase relative to a negative control animal (Hutcheson et al., 1997). Growth implants can be administered at various times over the lifetime of the steer or heifer with implant type typically changing as the animal grows in size. A

typical implant administered in the finishing phase (last 100 to 200 days on feed) contains a ratio of trenbolone acetate to estradiol benzoate (140 mg trenbolone acetate and 14 mg estradiol benzoate). Growth implants account for a 21% improvement in ADG and an 11% improvement in feed efficiency (dry matter intake:ADG) in the finishing phase (Reinhardt, 2007). Furthermore, growth implants increase carcass weight and yield of saleable beef. Guiroy et al. (2002) estimated that growth implants contributed to a 44 kg increase in carcass weight over negative controls when adjusted to the same body fat percentage.

These increases in efficiencies result in considerable net returns to the producer, increasing the economic sustainability of beef operations. Because growth implants contribute to an increase in beef production with equal or less resources, growth implants also increase the environmental sustainability of beef operations. Growth promoting implants are particularly important in confined cattle feeding operations where high feed prices, increased importance on carcass weight and tight margins necessitate the importance of feeding efficiency and increased final body and carcass weights. Negative impacts on performance and profitability due to growth implants are increased sexual activity (e. g., bullers), increased masculinity and behavioral issues, increased incidences in dark cutters, increased bone ossification and reduction in marbling when fed to a constant time or weight end-point. Negative impacts can largely be overcome under proper management scenarios and the current implanting program for particular cattle and marketing conditions.

2.8.2 B-Adrenergic Agonists

Beta-adrenergic agonists (β - agonists) are phenetholamine compounds consisting of a phenyl ring attached to a catecholamine group (Beermann, 2003b). Epinephrine and norepinephrine are naturally occurring β - agonists produced and released by the adrenal medulla and as a sympathetic nervous system neurotransmitter for epinephrine and norepinephrine, respectively (Mersmann, 1998). Beta - adrenergic agonists were originally designed pharmacologically to relieve respiratory distress, control premature parturition and as a cardiostimulant to treat cardiac irregularities (Mersmann, 1998). There are multiple receptor tissue types and multiple β - agonists including cimaterol, clenbuterol, L-644,969, salbutamol, ractopamine and zilpaterol. Ractopamine is a β_1 -adrenergic agonist whereas zilpaterol is a β_2 -adrenergic agonist; both compounds are orally active and fed in finishing diets at the end of the feeding period. Beta-adrenergic agonists act as repartitioning agents, partitioning dietary energy away from fat deposition to muscle deposition (Anderson and Johnson, 2004), which results in significant improvement to beef carcass leanness (Johnson et al., 2014).

Beta-adrenergic agonists act directly on cells via β -adrenergic receptors (BAR) on the cellular surface (Mersmann, 1998). There are three receptor sub-types (Table 1): β_1 -AR, β_2 -AR and β_3 -AR which in turn bind to G_s proteins (Mersmann, 1998). There is considerable homology among receptor types with receptors expressing seven hydrophobic transmembrane domains anchoring the receptor in the plasma membrane (Mersmann, 1998). Once the receptor and the G_s protein couple together, cyclic AMP becomes activated acting as an intracellular messenger (Mersmann, 1998). The intracellular messenger cyclic AMP binds to protein kinase A which subsequently

induces a chain of reactions when stimulated (Figure 3; Mersmann, 1998). The activation of protein kinase A phosphorylates a number of intracellular proteins including hormone sensitive lipase (Mersmann, 1998). With respect to adipose tissue, hormone sensitive lipase is activated (Anderson and Johnson, 2003), stimulating lipolysis. Additionally, intracellular lipogenesis is inhibited by the activation of BAR on adipose tissue cell membranes (Anderson and Johnson, 2003). Therefore, triglyceride substrates are repartitioned to other tissues in the body. Relatively, Z a β_2 -agonist has a larger impact on lipolysis in cattle relative to ractopamine a β_1 -agonist but lesser than clenbuterol (Anderson and Johnson, 2003). Further examination of BAA in skeletal muscle has shown that β_2 -agonists increase protein synthesis while decreasing protein degradation (Anderson and Johnson, 2003). Conversely, β_1 -agonists have only been reported to increase protein synthesis in bovine muscle cells (Anderson and Johnson, 2003). Therefore β_2 -agonists will result in more profound carcass leanness relative to β_1 -agonists. The difference in receptor sub-type with cattle having predominantly β_2 -receptor subtypes likely explains the difference in protein accretion and fat lipolysis between zilpaterol and ractopamine.

Table 2.1 Relative Abundance of BAA receptor sub-type by species and tissue

Species	Tissue	Abundance
Pig	Lung	67% β_1
	Adipose Tissue	73% β_1 , 20% β_2 and 7% β_3
Human	Lung	27% β_1
	Adipose Tissue	35% β_1 , 65% β_2
Cattle	Skeletal Muscle	>99% β_2
	Adipose Tissue	>90% β_2

Sources: Mersmann, 1998, Johnson et al. 2014

Beta-adrenergic agonist compounds do not act through the GH/IGF-1 axis and therefore act independently of steroid hormones. Steroidal implants result in increased DNA content in skeletal muscle fibers whereas β - agonists increase ratio of protein to

DNA (Johnson et al., 2014). Adipose tissue has been shown to have estrogenic receptors in low concentration but not androgenic receptors. The impact of steroidal hormones on adipose tissue is mainly through indirect effects from increased somatotropin (Anderson and Johnson, 2003). Carcasses from growth implanted cattle are traditionally leaner at harvest, this is primarily due to the increase of muscle mass and not the catabolism of fat (Anderson and Johnson, 2003). In contrast, β - agonists impact both adipose and skeletal muscle tissue leading to an increase in lipolysis and a net increase in protein accretion.

2.8.2.1 Benefits and Issues of β -Adrenergic Agonists

Elanco Animal Health (Greenfield, IN) manufactures Optaflexx, a commercial ractopamine product (β_1 -adrenergic agonist) and recently conducted a meta-analysis of the effect of Optaflexx in steers ($n = 26,483$). Relative to control steers, Optaflexx increased live BW (10.2 kg) and ADG (0.28 lb/d) in steers receiving increased doses of ractopamine up to 300 mg/hd/d (Elanco, 2012). Feed efficiency was also improved in a linear fashion with increasing doses of Optaflexx. Hot carcass weight was also improved by 9.1 kg at 300 mg/hd/d while percentage of Choice carcasses decreased 4.8 percentage points and percentage of Select carcasses increased 4.2 percentage points (Elanco, 2012). In the meta-analysis of factors, there was also a significant linear effect in the improvement of yield grade with 300 mg/hd/d resulting in the highest percentage of yield grade (YG) 1 and YG 2 carcasses (Elanco, 2012). Merck Animal Health (Summit, NJ) manufactures a commercial Z product called Zilmax (β_2 -adrenergic agonists), that has been tested on 26,606 steers in which a 15 kg HCW advantage was found over the controls (Merck Animal Health, 2013). Zilmax was very efficient at improving yield grade and dressing percent with the number of YG 1 and YG 2 carcasses increasing 9 and

6%, respectively and subsequently reducing YG 4 and 5 carcasses (Merck Animal Health, 2013). Zilpaterol also decreased quality grade with the percentage of choice carcasses decreasing by 7 percentage points (Merck Animal Health, 2013).

Zilpaterol also greatly improves carcass cutability with increases in fabrication yields (Hilton et al., 2009). This results in an increased net return to the packer and increased value through the beef production system. Zilpaterol has been shown to have a negative impact on tenderness (Strydom et al., 2009; Scramlin et al., 2010) and recent work on Optaflexx at above FDA-approved doses (400 mg/hd/d) has also shown a negative impact on tenderness (Arp et al. 2014). However, consistent ageing times greater than 21 d resulted in comparable tenderness to steaks from cattle not administered β - agonists (Brooks et al. 2009). Decreases in quality grade can be managed by extending days on feed (Vasconcelos et al. 2008 and Rathmann et al. 2012). Therefore considerable technology exists to improve lean muscle protein deposition in growing and finishing cattle. Management scenarios are likely to change with adoption of new technologies in order to capture the greatest economic advantage and decrease risks associated with the technologies.

2.9 Summary

Energetics is rooted in physics and chemistry and has been studied for centuries. Advancements in technology led to the creation of indirect calorimetry chambers and the use of RQ to estimate heat production in animals (Blaxter 1962; Brouwer 1965). Multiple studies sought to estimate individual animal HP as well as to explore the energetic value of feedstuffs (Armsby et al., 1918; Armsby et al., 1920; Blaxter, 1956; Blaxter and Wainman, 1966). With the advent of comparative slaughter techniques to

estimate energetics (Lofgreen and Garrett, 1968), chamber utilization decreased as North America adopted the California Net Energy System to estimate maintenance and gain requirements of cattle (NRC, 2000). Comparative slaughter techniques measure retained energy over a period of time and indirectly measure fasting heat production or basal energy metabolism of cattle (NRC, 2000).

Individual animal to animal variation of energetic efficiency is greatly dependent upon basal cell metabolic processes and in ruminants specifically the cellular metabolic processes that occur in the gut (McBride and Kelly, 1990; Reynolds 2002). Ultimately, metabolic requirements are a function of the surface area of the animal (Brody, 1945). In contrast, the utilization efficiency of gain is dependent upon the composition of gain (Garrett, 1980b; Old and Garrett, 1987). Therefore, equivalent empty body composition at harvest is critical for studying between breeds and across technologies for ultimate efficiency of gain (Fox and Black, 1984). While multiple studies have investigated empty body and carcass composition, the vast majority of studies (Lofgreen and Garrett, 1968; Old and Garrett, 1987; Guirouy et al., 2001) utilize predictions of empty body composition from specific gravity of the carcass (Garrett and Hinman, 1959) or 9-10-11 rib dissection (Hankins and Howe, 1946). Additionally, indirect calorimetry has been used to investigate the energy metabolism of ruminants treated with the β -agonists cimaterol and clenbuterol (Rikhardsson et al., 1991 and MacRae et al. 1988) but to date, no research has investigated the impact of Z, a FDA licensed β -agonist, on the energetics of beef cattle.

Thus, there is a need to re-evaluate the energy metabolism of beef steers using both comparative slaughter and indirect calorimetry as both genetics of cattle and

available growth promoting technologies have evolved. Therefore the objectives of the studies were to determine the impact of Z on energy and protein metabolism, non-carcass and carcass harvest yields, empty body composition and prediction of carcass and empty body composition.

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CHAPTER 3
THE EFFECT OF ZILPATEROL HYDROCHLORIDE SUPPLEMENTATION
ON ENERGY METABOLISM AND NITROGEN AND CARBON
RETENTION OF STEERS FED AT MAINTENANCE
AND FASTING INTAKE LEVELS

3.1 Abstract

An indirect calorimetry trial examined energy metabolism, apparent nutrient digestibility, carbon retention (CR) and nitrogen retention (NR) of cattle supplemented with zilpaterol hydrochloride (Z). Beef steers (n=20; 463 ± 14 kg) blocked (n=5) by weight and source were individually fed and adapted to maintenance energy intake for 21 d prior to Z (90 mg/hd/d) or C treatment for 20 d (455 ± 14 kg at start of treatment). Respiration chambers (n=4) were used to quantify heat production (HP) during maintenance (M; d 12 to 16 of Z period) and fasting heat production (FHP; d 19 to 20 of Z period, total 4 d fast). Steers were harvested after a 6 d Z withdrawal and carcasses graded 24 h post-harvest. Control cattle lost more BW ($P < 0.01$; 9 vs. 2 kg, respectively) during M while BW loss of Z steers was greater ($P < 0.01$; 9 vs. 4 kg, respectively) during FHP although no differences ($P \geq 0.76$) were detected for gain:feed, ADG and end of Z BW. No differences in DMI, apparent nutrient digestibility, O₂ consumption or CH₄ production ($P \geq 0.12$) were detected between treatments, however Z cattle had greater CO₂ production during M ($P = 0.04$; 2325 vs. 2185 L/steer; 23.6 vs. 22.4 L/kg BW^{0.75}).

Digestible energy and ME did not differ ($P \geq 0.19$) between treatments, however urinary energy was increased ($P = 0.05$; 0.091 vs. 0.074 Mcal) in C cattle. Steers treated with Z tended to have greater HP ($P = 0.09$; 12.44 vs. 11.69 Mcal, respectively) but the effect was reduced on a $BW^{0.75}$ basis ($P = 0.12$; 0.126 vs. 0.120 Mcal/kg $BW^{0.75}$, respectively). No treatment difference in FHP was observed ($P \geq 0.32$) although CO_2 production (L/steer) increased with Z treatment ($P = 0.04$; 1423 vs. 1338 L/steer). Control cattle excreted more ($P = 0.05$) N in urine (39.8 vs. 32.4 g/d, respectively), thus NR ($P = 0.07$; 22.14 vs. 14.12 g/d) tended to be greater for Z fed steers. Steers treated with Z lost more carbon via CO_2 ($P = 0.04$; 1,036.9 vs. 974.3 g, respectively) although total CR did not differ between treatments ($P \geq 0.23$). Empty body weight (EBW), HCW and harvest yields (g/kg EBW) were not different between treatments ($P \geq 0.13$). Supplementation of Z increased dressed carcass yield ($P = 0.02$; 62.12 vs. 60.65%, respectively) and LM area ($P = 0.02$; 77.81 vs. 70.90 cm²) while tending to lower USDA calculated yield grade ($P = 0.06$; 1.8 vs. 2.2). Results from this trial indicate that Z treatment tends ($P \geq 0.07$) to increase NR as well as modify HP during maintenance by increasing CO_2 production.

Keywords: beef, energy metabolism, nutrient digestibility, zilpaterol hydrochloride

3.2 Introduction

Zilpaterol hydrochloride (Z; Zilmax[®]) is a β_2 -adrenergic agonist that binds with high affinity to β_2 -adrenergic agonist receptors present on bovine adipose and muscle tissue (Mersmann, 1998; Johnson et al. 2014). The binding of β -adrenergic agonist receptors results in intracellular messaging that alters repartitioning of nutrients to lean muscle growth at the expense of adipose tissue accretion (Mersmann, 2002). As a result, Z causes pronounced live and carcass characteristic effects including improved ADG,

gain:feed, HCW, LM area, dressed carcass yield and yield grade while decreasing marbling score and estimated empty body fat percentage (Elam et al. 2009; Montgomery et al. 2009a,b; Rathmann et al. 2012). Additionally, Z has been shown to promote significant carcass gains relative to live gain (Rathmann et al. 2012) and increase the transfer of non-carcass components to carcass components (McEvers et al. 2013).

Due to the repartitioning of nutrients and increased carcass lean muscle mass, previous research has speculated that Z has impacts on the energetics of maintenance and gain. Previous research on β_2 -adrenergic agonists (L-644,490, cimaterol, Z and clenbuterol) have illustrated decreased protein degradation (Wheeler and Koohmaraie, 1992), increased nitrogen retention (NR; MacRae et al., 1988; Hovell et al., 1991; Rickardsson et al., 1991; Brake et al., 2011) and increased protein synthesis without dietary protein for 12 d (Inkster et al. 1989). Decreased protein degradation may be indicative of changing maintenance energy requirements (Reeds and Mersmann, 1991) or altered calpain:calpastatin ratios (Strydom et al. 2009). Previous research examining the energetics of wethers fed clenbuterol reported an increase in energy expenditure of 0.6 MJ/d over 20d (MacRae et al., 1987) whereas ewe lambs fed cimaterol did not increase maintenance energy expenditure after the first six days of treatment (Ricardsson et al., 1991). Currently, no research has been conducted with respect to the effect of Z on maintenance energy efficiency. Thus, the objectives of this study were to determine the impact of Z on maintenance energy requirements and carbon and nitrogen retention.

3.3 Materials and Methods

All experimental procedures involving live animals were approved by the Animal Care and Use Committee at West Texas A&M University and adhered to the regulations

in the Guide for the Care and Use of Agricultural Animals in Agricultural Research and Teaching (Federation of Animal Science Societies, 2010, Savoy, IL).

3.3.1 Live Cattle Procedures

Steers (n=20) were acquired from two ranch sources and blocked (n=5) by weight and source. Upon acquisition, cattle were treated for internal and external parasites with ivermectin (Ivomec, Merial, Duluth, GA). Cattle had previously been vaccinated against bovine rhinotracheitis virus, bovine viral diarrhea virus type 1 and 2, bovine parainfluenza-3 virus, bovine respiratory syncytial virus and *Mannheimia haemolytica* leukotoxin (Pyramid 5 + Presponse SQ, Boehringer Ingelheim Vetmedica, Inc., St. Joseph, MO; or Bovi-Shield Gold One Shot, Zoetis, Kalamazoo, MI, depending upon source) while both sources of cattle were vaccinated against *Clostridium chauvoei*, *septicum*, *novyi*, *sordellii* and *perfringens* Types C&D (Vision 7, Merck Animal Health, Summit, NJ) before purchase. Steers were not implanted during the handling and acclimation phase nor the trial and collection phase so as not to invoke a days on implant effect. Steers from one source (n=8) were administered a low dose growth implant (Ralgro, Merck Animal Health, Summit, NJ) 132 d prior to purchase while cattle from the other source (n=12) were never implanted. All cattle ears were palpated at purchase for remaining implants with no implants found.

3.3.2 Experimental Design and Treatment Application

The trial was conducted as a randomized complete block design (n=5 blocks) with individual steer as the experimental unit (n=4 per block). Steers (BW at randomization = 389 ± 11 kg) were assigned to blocks based on initial BW and assigned randomly to treatment within block (Z or C). Chambers were randomly assigned to steers for Block 1;

subsequent chamber assignments for Blocks 2 through 5 were conducted by rotating treatments by one chamber each block to prevent confounding chamber with treatment. Calorimetry data during maintenance intake were determined on d12 through d16 of Z or C treatment. Fasting began on d17 when steers were removed from the chambers and placed in a soil-surfaced pen. On d18 through d 20 steers were moved into the respiration chambers again with fasting heat production (FHP) determinations collected on d 19 and d 20. Heat production (HP) was calculated according to Brouwer et al. (1965).

Prior to maintenance adaptation, steers were halter trained, handled daily and acclimated to harness style fecal bags and digestion stalls for 6, 12 and 24 h increments followed by acclimation to respiration chambers (outside dimension of 244 cm length × 115 cm width × 210 cm height) while wearing fecal bags for 24, 48 and 72 h increments. Steers were fed an intermediate ration (73% concentrate, *ad libitum*) prior to a 12 d adaptation to a 90% concentrate finishing ration (Table 1) at 90% of previous *ad libitum* intake. Blocks were adapted 14 d apart to ensure the same number of transition days to a 90% concentrate diet (12 d) and the same number of days on maintenance adaptation (21 d). Cattle were placed on a maintenance level of intake $\{((BW \times 0.891)^{0.75} \times 0.077) / \text{diet NE}_m\}$, mean BW on d -1 and 1 of maintenance adaptation} for 21 d before being fed 90 mg/d zilpaterol hydrochloride (1.875 g/d Zilmax; Merck Animal Health, Summit, NJ) or C (C; no β -adrenergic agonist) treatment. During the 21d maintenance period, intakes were adjusted based on BW (d -1, 1 vs. 4, 5, 9, 10, 14, 15, 19 and 20) after 10h water and feed withdrawal (no feed remained for any animals at start of withdrawal periods). All cattle weights were recorded from an electronic scale (Trojan Livestock Equipment,

Weatherford, TX) with ± 0.45 kg accuracy; the scale was validated with 499 kg of certified weights before every use. Feed intakes were not adjusted during the 20 d treatment period.

Dosing of Z during fast was initially accomplished with 0.11 kg of rice hulls mixed with 1.875 g of Z and 45 mL of water (45 mL of water was also added to the C rice hulls). In the first period, one Z treated steer refused to eat rice hulls and subsequently all daily doses of Z were administered with an oral drench at 0845 and rice hulls were still offered as 0.11 kg per steer for the duration of the fast. Following the fast, steers were allowed a 6 d recovery period. On the first two days of recovery, steers were given long stem, wheat grass hay (2.5 kg) and intermediate ration (2.5 kg), the following four days, steers were given their pre-determined maintenance level of intake of the 90% concentrate finishing ration. Steers were not fed the morning of harvest. Respiration and digestion collections for Block 1 began 105 d after training and handling began on all steers. Total trial duration (start of Block 1 maintenance adaptations to the harvest of Block 5 steers) was 104 d.

3.3.3 Diets and Feeding

Cattle were individually fed using the Calan Broadbent Feeding System (American Calan, Northwood, NH, USA). Diets were mixed weekly in 454 kg batches; each batch of feed and ingredients were sampled weekly and dried at 55 °C, to a constant weight, in a forced-air convection oven (LBB 2-12, Despatch Industries, Minneapolis, MN) for determination of DM. Rations were stored in 200 L plastic drums in a refrigerated walk-in cooler to prevent feed spoilage and moisture loss. For each maintenance period, the same batch of feed was used and feed was weighed daily using

an electronic platform scale (Ohaus SD75, Ohaus Corporation, Parsippany, NJ) to \pm 0.045 kg accuracy, validated daily. An additional 0.10 kg of feed was weighed with each ration offering, then a 0.10 kg sub-sample was removed as the feed sample for each animal. After the maintenance collection period, the daily samples were composited by animal and dried at 55 °C for further analysis. Orts remained for one steer in Block 3 on Z treatment on two consecutive days of maintenance intake. Orts were composited (0.22 kg total), analyzed and nutrient content and energy subtracted from daily intake. Vitamins, minerals, monensin sodium and tylosin were added to the diet with an automated micro-ingredient machine (MicroBeef Technology, Amarillo, TX). Cattle when not on collections were fed once daily at 0730. Cattle on collections were fed after urine and fecal collections and after system validation at 0845

3.3.4 Indirect Respiration Calorimetry System

A four chamber, indirect respiration calorimetry system was used to measure gaseous exchange. The details of the chambers have been previously reported (Hales et al., 2012); with total chamber capacity of 6000 L. Each chamber was plumbed from the outgoing pipe to an individual flow kit (Flowkit-2000; Sable Systems International, Las Vegas NV) with the system creating a vacuum. Exterior, outside air flowed into the rear of each chamber via a 7.6 cm diameter polyvinylchloride pipe (PVC pipe) sealed to the roof, while inside chamber air was evacuated from the front of the chamber with a 5.1 cm diameter PVC pipe sealed to the roof. Air was systematically sampled using a multiplexer (RM8 Intelligent Multiplexer; Sable Systems International) in order of exterior air, chamber 1, chamber 2, exterior air, chamber 3 and chamber 4 during each 60

minute period. A constant aliquot of 2.5 mL of air was subsampled every second for 10 min using the multiplexer sequence of sampling.

Flow rate for each chamber was set at 600 L/min; ethanol recoveries were 100.22% for O₂ (SD ± 4.68) and 99.73% for CO₂ (SD ± 5.03). Each chamber was equipped with its own air conditioning system (Fredrich Company, San Antonio, TX) to maintain steady temperature (18.9 °C) and humidity. Air conditioning units were serviced and temperature validated prior to study start date. Relative humidity, temperature and barometric pressure across chambers and outside air were measured with a RH-300 Analyzer (Sable Systems International) after which air was dried (ND-2 Gas Dryer, Sable Systems International). Oxygen was measured with a paramagnetic analyzer (PA-10 Oxygen Analyzer, Sable Systems International) whereas carbon dioxide and methane were measured with infrared analyzers (CA-10A Carbon Dioxide Analyzer and MA-10A Methane Analyzer, respectively, Sable Systems International). Gas analyzers were calibrated daily using prepared gas standards (carbon dioxide, methane and oxygen PraxAir, Amarillo, TX).

Due to low intake levels and a contemporary high concentrate finishing diet with ionophores, a back-up continuous monitoring system was designed for methane readings. The system was plumbed with a separate 6.35 mm diameter Nalgene plastic tubing into the top of the chambers, 10 cm from the outgoing main air line. The plastic tubing from each chamber as well as a separate identical tubing for exterior air were fed into a heavy duty sealed trunk container (Vac-U-Chamber, SKC, Eighty Four, PA) kept under negative pressure by a continuous pump (Universal PCXR8, SKC, Eighty Four, PA). The negative pressure was designed to pull a steady quantity of air (5.9 mL/min) into

individual 10L FlexFoil PLUS air sample bags (SKC, Eighty Four, PA) filled to 80% of capacity within the 22.5 h collection period. After analyzers were calibrated each morning, back-up bags of exterior air and each chamber were run on the system for 5 minutes each before the system daily start-up.

3.3.5 Digestion Collections

System shut down started at 0730, with urine and feces collection following the opening of the sealed chambers. Cattle were fed immediately prior to system start up at 0845 with chambers sealed for 22.5 hours per day. Urine collection was performed via a high density polyethylene pan (66 cm length x 51 cm width x 15 cm height) placed underneath the cattle and level with the elevated floor boards. Urine pans were covered with a heavy duty, plastic coated steel grate. Prior to collections, the underside of the steel grate was covered with a double layer of cheesecloth to prevent any debris from entering the urine pan and 24 hour validation of urine pans was performed prior to start of trial and at the end of trial by placing 20 L of water in each pan and ensuring full recovery. To prevent the volatilization of N as ammonia, 300 mL of a 25% vol/vol (3 N HCl acid solution) was added to each pan to ensure the pH of the urine was <4. Urine was aspirated daily from each pan using an indwelling hose and a vacuum into a 20 L carboy; urine pH was checked daily (Oakton pH 11 meter, Oakton Instruments, Vernon Hills, IL). Feces were collected into harness style fecal bags lined with disposable kitchen garbage bags, which were changed daily and weighed. Feces and urine was weighed using a scale with an accuracy of ± 0.02 kg (Sartorius Signum[®], Hamburg, Germany). Ten percent of urine and feces were sub-sampled daily, kept separate by day, and refrigerated at 4 °C. Following collections, feces and urine was composited,

respectively with half of the feces dried in a forced air convection oven at 55 °C and half frozen at -20 °C in whirl-Pak bags. Urine was composited and frozen at -20 °C in 4 x 15 mL centrifuge tubes (for subsequent analysis of creatinine, 3-n-methylhistidine, energy, and N) as well as a bulk urine sample contained in 1 L polyethylene bottles.

3.3.6 Sample Analysis

After drying of feces, diet and orts to a constant weight at 55°C, in a forced-air convection oven, (LBB 2-12, Despatch Industries, Minneapolis, MN), all samples were ground through a 1-mm screen using a Wiley Mill. Feed, feces and orts were analyzed for gross energy using an Automatic Isoperibol Calorimeter (Parr Instrument Company, Moline, IL). For determination of urine energy, urine was thawed at 4°C, filtered through Whatman-450 filter paper and lyophilized in small plastic pouches (Parr Instrument Company, Moline, IL) prior to bomb calorimeter analysis (Automatic Isoperibol Calorimeter, Parr Instrument Company, Moline, IL); the energy value of the plastic pouch (n = 10 control samples) was subtracted from the total urine energy value. Total C of feed and feces was determined using a C-N analyzer (Elementar Vario MAX CN, Elementar Americas Inc., Mt Laurel NJ) whereas C in the urine was analyzed with a Shimadzu TOC-L (carbon) analyzer with an ASI-L autosampler (Shimadzu, Columbia, MD) after filtering urine through Whatman-450 filter paper. Dried and ground feed, feces and orts was analyzed by a commercial lab (ServiTech Labs, Amarillo TX) for DM (AOAC #934.01), N (AOAC #990.03), starch (Xiong et al. 1990), ADF (Ankom Technology Method 5), NDF (Ankom Technology Method 6) and ether extract (AOAC #2003.06); feed was also analyzed for Ca and P (AOAC #968.08).

3.3.7 Harvest and Carcass Grading

All steers were harvested at the WTAMU Meat Lab (USDA Est. 7124) following a 12 h lairage with access to water. At harvest, stun weight, exsanguinated weight and hot carcass weight were recorded in addition to the weights of all internal viscera. Omental fat was trimmed away from the stomach compartments and weighed separately; gastrointestinal tracts were cleaned, flushed of contents and weighed empty. Kidney-pelvic-heart fat was trimmed from the carcass and weighed. Twenty-four hours post-harvest, a detailed carcass evaluation was conducted on chilled (4 °C) carcasses which included marbling score, lean and skeletal maturity, 12th rib s.c. fat depth (cm), and LM area (cm²). A final quality grade and calculated yield grade was determined (USDA, 1997). Also, percentage of empty body fat was estimated for each animal (Guiroy et al., 2001).

3.3.8 Carcass Fabrication and Protein Turnover Analysis

At 48 h post-harvest, left carcass sides were cut into primals and weighed to determine the cold carcass side weight. Carcass primals were separated into bone, fat and lean. All soft tissue was weighed and ground through a meat grinder (346SS, Biro, Marblehead, OH) with a 9.5 mm plate. All ground meat was then thoroughly mixed and re-ground through a fine grind plate (3.2 mm); grind was sampled three times and the three samples thoroughly mixed and subsampled into three homogenous Whirl-Pak bags. Samples were immediately vacuum sealed and frozen at -20 °C until further analysis. Dry matter was determined by lypophilization method and by drying for 3 h at 105 °C (NFTA #2.1.4). After lypophilization, samples were ground in a Waring Blender prior to determination of N (AOAC #978.10; SDK Labs, Hutchinson, KS). Total N content of the skeletal muscle mass was used to determine total skeletal 3-n-methyl histidine pool

using the methods of Nishizawa et al. (1979). Urine samples for creatinine analysis were thawed at 4 °C, centrifuged at $2,860 \times g$ at 4 °C for 15 min and analyzed using a creatinine colorimetric 96-well plate kit (MAK080-1KT, Sigma-Aldrich Co., St. Louis, MO). Urine 3-*n*-methylhistidine was determined by HPLC analysis (AminoAcids, St. Paul, MN) using the methods of Wheeler and Koohmairie (1992) and was used to estimate protein degradation calculations.

3.3.9 Statistical Analysis

Data was analyzed as a randomized complete block using the Mixed procedure of SAS (SAS 9.3, SAS Institute Inc. Cary, NC) with the fixed effect of treatment and the random effect of block and chamber. Significant effects were declared at $P \leq 0.05$ and trends discussed if a resulting *P*-value was between 0.05 and 0.10.

3.4 Results and Discussion

3.4.1 Animal Performance and Nutrient Digestibility

Cattle weight at the start of maintenance and 21 d later at the start of Z supplementation did not differ ($P = 0.64$) between treatments (Table 2). While no treatment differences ($P = 0.79$) occurred for the start of maintenance chamber BW, C cattle lost more ($P = 0.01$) BW (9 vs. 2 kg) during maintenance collections (d 12 through d 16) while Z cattle lost more BW ($P = 0.01$; 9 vs. 4 kg, respectively) during FHP collections. Weight at the end of Z withdrawal period (end of trial weight) did not differ ($P = 0.76$) and in concurrence ADG ($P = 0.78$) and gain:feed ($P = 0.81$) did not differ between treatments. Results from the current trial do not reflect outcomes reported in previous literature on performance during Z supplementation in which Z has improved BW, ADG and gain:feed (Vasconcelos et al., 2008; Elam et al., 2009 and Montgomery et

al., 2009). In this trial, Z supplemented cattle appeared to maintain weight while in chambers for maintenance whereas C cattle did not; this is likely the result of Z continuing to exert a positive gain impetus while on maintenance. Conversely, during FHP, Z treated cattle lost more weight than C cattle; this loss may be indicative of tissue shrink in Z cattle that did not previously occur during the maintenance collection period. Research by Rikhardsson et al. (1991) reported that cimaterol fed for 70 d resulted in increased ADG and gain:feed in ewe lambs housed in respiration chambers for collection periods. Differences may be due to the type and duration of β -adrenergic agonist feeding as well as lack of fast across all lambs in the trial by Rikhardsson et al. (1991).

Absolute DMI during maintenance did not differ (Table 3, $P = 0.90$) between treatments. Intake had to be increased throughout the 21 d maintenance adaptation period by 6.1% relative to the initial prediction derived from the NRC equation for maintenance energy (NRC, 2000). Intake expressed as $\text{g/kg BW}^{0.75}$ was not different between treatments ($P = 0.56$; 35.49 vs. 35.23 for C and Z, respectively). Fecal output and urine output did not differ between treatments ($P \geq 0.15$). Previous research in ewe lambs reported that cimaterol treatment increased urinary output vs. C but resulted in less energy per volume of urine (Rikhardsson et al. 1991). Intake, excretion and apparent digestibility of NDF, starch and ether extract also did not differ between treatments ($P \geq 0.17$). Rikhardsson et al. (1991) also noted no difference in DM, CP or DE between cimaterol treated and C lamb ewes and Brake et al. (2011) did not observe a difference in DM, organic matter or N total tract digestion in steers treated with Z or C steers. Previous research (Walker and Drouillard, 2010) suggested that β -adrenergic agonists modify in vitro DM digestibility and decrease rumen ammonia concentrations however

results from this study do not support the hypothesis that β -adrenergic agonists modify total tract digestibility of nutrients.

3.4.2 Energy Losses

Clenbuterol has been reported to increase heat production (Macrae et al., 1988; Lobley et al., 1985) whereas cimaterol has increased heat production in the first four days after administration (Rikhardsson et al., 1991). Results of the current study (Table 4) indicate that absolute O_2 consumption ($P = 0.12$) or O_2 consumption per unit of metabolic BW ($P = 0.17$) did not differ between treatments during maintenance energy intake. However, carbon dioxide production was increased ($P = 0.04$) by Z supplementation (2325 vs. 2185 L/d and 23.63 vs. 22.37 L/kg BW^{0.75}, respectively) and RQ remained unchanged (0.93; $P = 0.87$) by treatment. Methane production did not differ between treatments ($P \geq 0.19$) when expressed as L/kg of DMI or as Mcal/kgBW^{0.75}.

At FHP (Table 4), O_2 consumption did not differ ($P \geq 0.46$) between treatments as L/steer nor L/kg BW^{0.75}. Carbon dioxide production during fasting followed in parallel fashion to carbon dioxide production at maintenance intakes; cattle supplemented with Z had greater ($P = 0.04$) carbon dioxide production (1423 vs. 1338 L/steer, respectively). However, when carbon dioxide production was expressed as L/kg BW^{0.75} no treatment differences existed ($P = 0.12$). Previous research on cimaterol and clenbuterol fed to lambs reported differences early in the feeding of β -adrenergic agonists and thus, more stimulus may occur earlier on when the β -adrenergic agonist is initially fed vs. results from the current trial after feeding Z for 12 to 16 d and 19 to 20 d for maintenance and FHP, respectively.

Results from the current study (Table 5) did not detect treatment differences for GE intake ($P \geq 0.28$) or fecal energy loss ($P \geq 0.23$) resulting in no differences to digestible energy (DE) intake ($P = 0.61$; 0.135 and 0.136 Mcal/kg BW^{0.75} for C and Z treatments, respectively), as expected. Urinary energy was greater ($P = 0.05$) in C than Z steers (0.091 vs. 0.074 Mcal/d, respectively). When expressed as Mcal/kg urine, C cattle had greater urine energy losses ($P = 0.02$; 0.026 Mcal/kg urine for C vs. Z treated cattle at 0.018 Mcal/kg urine). Methane energy losses were not different between treatments when expressed as a % of DE ($P = 0.27$; 2.50 and 3.16 % of DE for C and Z, respectively). However, overall metabolizable energy (ME) intake did not differ between treatments ($P = 0.68$; 12.80 and 12.92 Mcal/d for C and Z treated cattle, respectively); ME intake expressed on a metabolic BW basis was 0.131 Mcal/kg BW^{0.75} ($P = 0.93$) for both treatments. Digestible energy intake and methane losses from this study are similar to previous results from Rikhardsson et al. (1991) in that no significant differences were observed between DE intake and energy lost as methane in C or cimaterol treated lamb ewes. Results of the current study differ from Rikhardsson et al. (1991) as percentage of energy lost in the urine was not different between cimaterol and C lamb ewes in the previous study. As noted by Rikhardsson et al. (1991), a high level of digestible protein was fed and may have contributed to larger than normal energy losses in urine overall and thus undetectable differences between C and cimaterol treatments.

Heat production at maintenance intake tended to be different ($P = 0.09$) between treatments with 11.69 and 12.44 Mcal for C and Z cattle, respectively. Heat production at maintenance on a metabolic BW basis was not different ($P = 0.12$) between treatments and was 0.120 Mcal/kg BW^{0.75} for C and 0.126 Mcal/kg BW^{0.75} for Z treated cattle.

Fasting heat production was not different ($P = 0.32$) between treatments nor was it different ($P = 0.45$) when expressed on a metabolic BW basis ($0.093 \text{ Mcal/kg BW}^{0.75}$ and $0.095 \text{ Mcal/kg BW}^{0.75}$ for C and Z, respectively). Previous research has reported shifts in heat production early in the administration of β -adrenergic agonist feeding with the effects dissipating over days on treatment. Rikhardsson et al. (1991) reported cimaterol increased heat production from d 1 through 4 of feeding but did not detect differences after 10 d of cimaterol feeding whereas MacRae et al. (1986) reported clenbuterol increased maintenance heat production throughout 20 d of treatment but was highest during the first 5 d. Rikhardsson et al. (1991) reported no differences in fasting heat losses between C and cimaterol treated ewe lambs (80.1 and $84.4 \text{ kcal/kg BW}^{0.75}$ for C and treated ewe lambs, respectively).

3.4.3 Carbon Balance

Carbon balance has not previously been elucidated in β -adrenergic agonist work. Carbon intake, apparent carbon digestion and urine and feces excretion did not differ between treatments ($P > 0.29$, Table 6), however carbon lost as CO_2 was increased ($P = 0.04$) in Z treated cattle (1037 g/d) relative to C cattle (974 g/d). This may be indicative of adipose tissue breakdown and release of carbon, expired as carbon dioxide through the respiration system. Hales et al. (2014) noted an increase in the slope of respiration rate as days on feed increased but were unable to discern differences between C and Z fed cattle. Overall retained carbon did not differ ($P = 0.43$) between the two treatments in this study. Previous carbon retention research investigated steam-flaked corn-based diets with increasing levels of wet distillers grains with solubles (WDGS) (Hales et al., 2013). The authors reported a decreasing linear trend for carbon retention (% of C intake) as WDGS

increased in the ration; primarily due to more carbon excreted in the feces (Hales et al. 2013). In contrast, Hales et al. (2012) reported that cattle fed steam-flaked corn had increased carbon retention vs. those fed dry-rolled corn primarily due to lower fecal and methane carbon losses.

3.4.4 Nitrogen Balance and Estimated Protein Turnover

Nitrogen intake, fecal N excretion and apparent N digested did not differ ($P \geq 0.75$) between treatments whereas urinary N excretion was lower ($P = 0.05$, Table 7) in Z cattle (32.38 g/d) vs. C (39.77 g/d). Total N excretion tended to be 11.2% higher ($P = 0.07$) in C (62.0 vs. 55.1 g/d, respectively) and accordingly, N retention tended to be increased by 56.8% ($P = 0.07$) in Z (22.1 g/d) vs. C (14.1 g/d). The increased N retention may indicate an impetus for lean protein accretion and/or reduced muscle turnover with Z supplementation resulting in reduced urinary N excretion observed in the current study. Rikhardsson et al. (1991) reported 11.7% lower urinary N excretion and 18.5% greater N retention in lamb ewes during weeks 3 and 6 of cimaterol feeding. The authors noted the largest improvement in N retention (27%) occurred during the third week of cimaterol feeding. Brake et al. (2011) reported that Z increased nitrogen retention 62.8% across diets in *ad libitum* fed rations.

With respect to the skeletal muscle protein pool, Z steers had an increased skeletal muscle protein pool than C steers ($P = 0.03$; 41.2 vs. 38.4 kg, respectively). Further analysis for 3-*n*-methylhistidine is warranted to explore the impact of Z on fractional protein breakdown. Williams et al. (1987) reported increased daily urinary creatinine excretion and reduced urinary 3-*n*-methylhistidine excretion in veal calves treated with clenbuterol. Fractional protein breakdown illustrated clenbuterol calves were 66% that of

the C (Williams et al., 1987). Wheeler and Koohmairie (1992) reported $L_{644,490}$ decreased fractional degradation rate during week 3 and increased fractional accretion rate during week 1, 3, 5 and 6 relative to C steers. With respect to energetics, the increase in skeletal muscle protein associated with β -adrenergic agonist feeding would typically be met with an increase in energy demand required to support the accretion and continual degradation and re-synthesis of the protein (Rikhardsson et al. 1991). However, if protein degradation is decreased with β -adrenergic agonist supplementation, increasing protein accretion may not require additional energy. In previous research, protein deposition energy requirement was reduced by 4.1 kcal/g of protein deposition (14.4 kcal/g for control vs. 9.3 kcal/g for cimaterol) in lambs treated with cimaterol (Caine and Mathison, 1988 as reported by Rikhardsson et al. 1991). Therefore, the impact of β -adrenergic agonists on reducing protein degradation while increasing skeletal muscle protein may not result in substantial changes in energy demands.

3.4.5 Harvest Yields and Carcass Composition

In this trial, feeding Z did not ($P \geq 0.18$) affect EBW, or harvest yields (Table 8). Hot carcass weight was not different ($P = 0.12$) between treatments whereas dressed carcass yield was improved ($P = 0.002$) with Z treatment (62.12 vs. 60.65%; Table 9). Cattle treated with Z also had greater ($P = 0.02$) LM area than to C cattle (77.8 vs. 70.9 cm², respectively) and a tendency ($P = 0.06$) for improved calculated yield grade (1.8 vs. 2.2, respectively). Neither marbling score ($P = 0.15$) nor calculated empty body fat ($P = 0.69$) differed between treatments. Increase in LM area and subsequent improvements in yield grade have also been reported by Avendano-Reyes et al. (2006) and Montgomery et al. (2009a) with an 8.4 cm² and an 8.0 cm² increase in *l. dorsi* area, respectively similar to

the current observation of 6.9 cm². While HCW was numerically greater in the current study for Z (269 vs. 263 kg), previous research has reported larger differences in HCW of 13 kg (Montgomery et al. 2009a) and 17.2 kg (Vasconcelos et al. 2008). In contrast, increased dressed carcass yield, as a result of Z treatment, in the current study is similar to previous research results of 1.2, 1.3 and 1.7% (Montgomery et al., 2009ab and Vasconcelos et al., 2008, respectively). The diminished carcass gains in the current study are likely due to the maintenance level of intake followed by a 4 d fasting period and a 6 d recovery. It is important to note that total live BW change from the start of Z feeding to the end of Z feeding plus a 3 d withdrawal was -3 kg for C and -4 kg for Z and thus carcass gain in lieu of no additional BW gain is evidence that Z repartitioned energy stores from gastrointestinal or adipose tissue to skeletal muscle growth as evidenced by the additional 2.78 kg of skeletal muscle protein (Table 6). Marbling scores and calculated EBF did not differ in the current study unlike previous reports of decreased marbling scores and calculated EBF (Montgomery et al. 2009a). Cattle used in this study were lighter than cattle used in most finishing trials and once on a maintenance plane of intake, neither C nor Z cattle were likely to have deposited additional fat which may have resulted in the lack of fat-associated differences between Z and C cattle in this study. Similar to the current study, Wheeler and Koohmaraie (1992) reported the β -AA L_{644,490} to improve HCW, LM area and yield grades while having no significant impact on marbling scores, KPH % or 12th rib s.c. fat thickness in an intensive steer metabolism trial.

Results from the current study indicate tendencies for protein and energy metabolism to shift in cattle treated with Z at maintenance intake levels. Supplementation

of Z resulted in increased carbon dioxide production at maintenance and tended to increase HP at maintenance whereas FHP remained unchanged. Supplementation of Z tended to increase N retention by reducing urinary N output. In addition, Z treatment increased skeletal muscle protein, dressed carcass yield and LM area indicating changes to protein metabolism. This data would suggest that Z increases protein deposition and results in tendencies for HP to increase at maintenance intake levels. Further research is warranted to examine the efficiencies of gain and maintenance and, in addition, the efficiencies of fat and protein deposition in cattle supplemented with Z to elucidate the repartitioning hypothesis of β -agonist supplementation.

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Table 3.1. Dietary ingredients (% DM basis), and nutrient basis (DM basis) of finishing rations fed to steers.

Ingredients, % DM basis	
Steam Flaked corn	71.91
Corn Gluten Feed	9.65
Wheat Hay	9.57
Molasses Blend	1.21
Fat	2.37
Supplement	5.29
Nutrient Analysis (\pm std deviation)	
DM, %	78.79 ± 2.235
Crude Protein, % DM	13.84 ± 0.636
Starch	53.29 ± 4.450
NDF, % DM	15.65 ± 1.215
ADF, % DM	7.70 ± 0.314
Ether extract, % DM	6.32 ± 0.888
Ash, % DM	4.43 ± 0.380
Calcium, % DM	0.49 ± 0.107
Phosphorus, % DM	0.34 ± 0.019

Table 3.2. Weights and gain efficiency for control and zilpaterol-fed beef steers at maintenance and fast

Item	Treatment		SEM	P-value
	Control	Zilpaterol		
n	10	10		
Initial wt at Start of Maintenance adaptation., kg	461	464	6.8	0.64
Initial wt at Start of ZH, kg	453	456	6.6	0.64
Start of Maintenance Chambers, kg	454	456	5.4	0.79
Mid-pt Maintenance Chambers, kg ¹	450	455	5.6	0.38
BW loss Maintenance Chambers, kg	-9.0	-2.2	2.41	0.01
Start of Fasting Heat Production Chambers, kg	445	454	6.1	0.20
Mid-pt Fasting Heat Production Chambers, kg ¹	444	449	6.2	0.37
BW loss Fasting Heat Production Chambers, kg	-3.8	-9.3	1.50	0.01
End of trial wt, kg ²	450	452	5.4	0.76
ADG	-0.12	-0.16	0.162	0.78
Gain:Feed	-0.034	-0.04	0.047	0.81

¹ Mid-point weights calculated as the average between the start and end of chamber Maintenance and FHP periods

² Weight after Zilpaterol and 3 day withdrawal

Table 3.3 Dry matter intake (DMI), and apparent digestibility of steers fed control or zilpaterol treated diets at maintenance intake

Item	Treatment		SEM	P-value
	Control	Zilpaterol		
DMI, kg	3.47	3.47	0.058	0.90
DMI, g/kgBW ⁷⁵	35.49	35.23	0.265	0.56
Fecal output, kg	0.83	0.77	0.068	0.35
Urine output, kg	3.72	4.78	0.690	0.15
<i>Nutrients</i>				
NDF				
Intake, g/d	549.9	533.6	11.73	0.34
Fecal excretion, g/d	376.6	331.6	31.00	0.17
Apparent digested, g/d	173.3	204.4	29.73	0.31
Apparent digestibility, % of intake	31.55	38.08	5.49	0.25
Starch				
Intake, g/d	1849.9	1848.3	64.79	0.98
Fecal excretion, g/d	5.5	6.2	1.11	0.55
Apparent digested, g/d	1844.4	1842.2	65.06	0.97
Apparent digestibility, % of intake	99.7	99.7	0.06	0.55
Ether Extract				
Intake, g/d	218.3	219.2	3.97	0.84
Fecal excretion, g/d	14.3	13.9	0.77	0.58
Apparent digested, g/d	204.0	205.3	3.87	0.75
Apparent digestibility, % of intake	93.4	93.6	0.35	0.65

Table 3.4. Daily O₂ consumption and CH₄ and CO₂ emissions of control and zilpaterol-fed beef steers at maintenance and fasting

Item	Treatment		SEM	P-value
	Control	Zilpaterol		
O ₂ consumption at maintenance				
L/steer	2366.2	2513.8	87.32	0.12
L/kgBW ^{0.75}	24.25	25.52	0.853	0.17
CO ₂ production at maintenance				
L/steer	2184.8	2325.2	59.82	0.04
L/kgBW ^{0.75}	22.37	23.63	0.541	0.04
RQ	0.93	0.93	0.019	0.87
CH ₄ production at maintenance				
L/steer	34.6	46.0	8.15	0.19
L/kg of DMI	10.07	13.26	2.436	0.22
CO ₂ to CH ₄ ratio	71.98	75.03	13.56	0.83
O ₂ consumption at fast				
L/steer	1940.9	1996.4	72.88	0.46
L/kgBW ^{0.75}	20.11	20.47	0.760	0.64
CO ₂ production at fast				
L/steer	1337.9	1422.6	37.13	0.04
L/kgBW ^{0.75}	13.87	14.60	0.428	0.12

Table 3.5. Daily energy losses for control and zilpaterol-fed beef steers

Item	Treatment		SEM	P-value
	Control	Zilpaterol		
GE, Mcal/kg ^z	4.81	4.80	0.014	0.28
GE, Mcal	16.68	16.65	0.287	0.93
GE, Mcal/kgBW ^{0.75}	0.171	0.169	0.002	0.42
Fecal Energy, Mcal	3.48	3.21	0.247	0.28
Fecal Energy, % GE	20.92	19.25	1.331	0.23
DE, Mcal/kg ^z	3.81	3.87	0.063	0.31
DE, Mcal	13.20	13.44	0.286	0.42
DE, % of GE	79.08	80.75	1.332	0.23
DE, Mcal/kgBW ^{0.75}	0.135	0.136	0.0032	0.61
Urinary Energy, Mcal	0.091	0.074	0.008	0.05
Urinary Energy, % GE	0.55	0.45	0.049	0.07
Urinary Energy, Mcal/kg Urine	0.026	0.018	0.003	0.02
CH ₄ Energy, Mcal	0.32	0.43	0.076	0.19
CH ₄ Energy, Mcal % of GE	1.95	2.58	0.472	0.21
CH ₄ Energy, Mcal % of DE	2.50	3.16	0.568	0.27
CH ₄ Energy, Mcal/kgBW ^{0.75}	0.0033	0.0044	0.00077	0.20
ME, Mcal/kg ^z	3.69	3.72	0.063	0.58
ME, Mcal	12.80	12.92	0.287	0.68
ME, % of GE	76.66	77.64	1.356	0.48
ME, Mcal/kgBW ^{0.75}	0.131	0.131	0.0025	0.93
Heat Production, Mcal	11.69	12.44	0.396	0.09
Heat Production, % GE	70.24	74.83	1.861	0.03
HP, Mcal/kgBW ^{0.75}	0.120	0.126	0.0038	0.12
Retained Energy, Mcal	1.03	0.56	0.471	0.34
Retained Energy, Mcal/kgBW ^{0.75}	0.010	0.006	0.005	0.32
FHP, Mcal	9.06	9.39	0.315	0.32
FHP, Mcal/kgBW ^{0.75}	0.093	0.095	0.0032	0.45

Table 3.6. Daily N balance of control and zilpaterol-fed beef steers
at maintenance level of intake

Item	Treatment		SEM	P-value
	Control	Zilpaterol		
N intake, g/d	76.41	77.04	1.937	0.75
N excretion, g/d				
Urine	39.77	32.38	3.461	0.05
Feces	22.56	22.47	2.031	0.96
Total	62.05	55.13	3.499	0.07
N excretion, % of total N excretion				
Urine	63.47	58.15	4.070	0.21
Feces	36.53	41.85	4.070	0.21
N excretion, % of total N Intake				
Urine	52.20	42.42	4.731	0.06
Feces	29.55	29.20	2.578	0.88
Apparent N Digested				
g/d	53.90	54.52	2.389	0.80
% of N intake	70.45	70.83	2.578	0.88
N retained				
g/d	14.12	22.14	4.021	0.07
% of N intake	18.25	28.41	4.966	0.06
% of digested N	25.49	39.73	6.804	0.06
Skeletal Muscle Protein, kg	38.43	41.21	1.01	0.03

Table 3.7. Daily C balance of control and zilpaterol-fed beef steers
at maintenance level of intake

Item	Treatment		SEM	P-value
	Control	Zilpaterol		
C intake, g/d	1512	1514	25.9	0.94
C loss, g/d				
Urine	51.3	49.6	4.95	0.74
Feces	309.3	285.0	22.49	0.29
CO ₂ -C	974.3	1036.9	26.67	0.04
CH ₄ -C	15.4	20.5	3.64	0.19
Total	1352.4	1389.9	42.80	0.40
C excretion, % of total C loss				
Urine	3.8	3.6	0.36	0.54
Feces	22.7	20.6	1.31	0.12
CO ₂ -C	72.2	74.4	1.09	0.07
CH ₄ -C	1.1	1.5	0.27	0.25
C excretion, % of total C Intake				
Urine	3.4	3.3	0.34	0.73
Feces	20.4	18.8	1.34	0.23
CO ₂ -C	64.5	68.7	1.33	0.01
CH ₄ -C	1.0	1.4	0.25	0.21
Apparent C Digested				
g/d	1203.7	1228.5	27.53	0.38
% of C intake	79.6	81.2	1.34	0.23
C retained				
g/d	157.4	126.8	37.62	0.43
% of C intake	10.3	8.13	2.47	0.39
% of C digested	13.0	9.8	2.81	0.29

Table 3.8. Harvest yields of control and zilpaterol-fed beef steers after maintenance and fasting

Item	Treatment		SEM	P-value
	Control	Zilpaterol		
Final BW, kg	434	435	5.6	0.82
Exsanguinated BW, kg	418	422	5.8	0.55
Empty BW	402	406	4.8	0.51
Empty BW/Final BW, %	92.8	93.3	0.59	0.46
Kill Yields, g/kg EBW				
Hide	92.78	91.90	3.646	0.81
Penis	2.85	2.73	0.390	0.76
Bladder	0.65	1.49	0.602	0.18
Trachea	2.70	3.04	0.251	0.19
Heart	4.86	4.69	0.191	0.37
Lungs	6.36	6.48	0.365	0.75
Pluck Trim	4.48	4.19	0.511	0.59
Spleen	1.92	1.92	0.100	0.93
Liver	9.88	9.77	0.254	0.68
Gall Bladder	0.34	0.31	0.098	0.77
Pancreas	0.99	0.93	0.119	0.59
Weasand	0.83	0.90	0.053	0.23
Stomach & Intestines Empty	73.82	76.27	2.330	0.31
GIT Trimmable Fat	13.02	11.41	1.34	0.25
KPH Fat	9.24	10.23	1.128	0.40
HCW, g/kg EBW	648	661	8.0	0.13

Table 3.9. Carcass grading and skeletal muscle protein of control and zilpaterol-fed beef steers after maintenance and fasting

Item	Treatment		SEM	P-value
	Control	Zilpaterol		
Hot carcass weight, kg	263	269	3.614	0.12
Dressed carcass yield, %	60.65	62.12	0.374	0.002
12 th rib fat thickness, cm	0.72	0.64	0.111	0.47
LM area, cm ²	70.90	77.81	2.520	0.02
Kidney-Pelvic-Heart fat, %	1.42	1.55	0.24	0.44
Calculated Yield Grade ¹	2.2	1.8	0.17	0.06
Marbling Score ²	31	33	0.98	0.15
Calculated empty body fat ³ , %	24.86	24.25	1.21	0.69

¹USDA calculated yield grade = $2.5 + (2.5 \times \text{FT}) + (0.2 \times \text{KPH}) + (0.0038 \times \text{HCW}) - (0.32 \times \text{REA})$, where FT = 12th rib fat depth in cm, KPH = percentage of kidney, pelvic, and heart fat, HCW = hot carcass weight in kg, and REA = longissimus muscle area in cm²

² Marbling Scores: 20=Traces; 30=Slight; 40=Small; 50=Modest

³Empty body fat, % = $17.76207 + (4.68142 \times \text{s.c. fat depth, cm}) + (0.01945 \times \text{HCW, kg}) + (0.81855 \times \text{quality grade}) - (0.06754 \times \text{LM area, cm}^2)$. Numerical quality grade values were assigned based on the marbling score derived quality grade such that Standard = 3 to 4; Select = 4 to 5; low Choice = 5 to 6; average Choice = 6 to 7; high Choice = 7 to 8; low Prime = 8 to 9; and average Prime = 9 to 10; Guioy et al. (2001).

CHAPTER 4
EFFECT OF DIETARY ENERGY LEVEL AND ZILPATEROL
HYDROCHLORIDE ON THE LIVE PERFORMANCE,
GRADING PERFORMANCE AND HARVEST YIELDS
OF BEEF STEERS

4.1 Abstract

A trial was conducted to examine live growth efficiency, harvest yields, and carcass grading performance of steers fed at maintenance (M) or at *ad libitum* (A) level of intake during zilpaterol hydrochloride (Z) supplementation. Single-sired, beef steers (n=56; start of trial BW 590 ± 36 kg) blocked (n=2) by weight and terminal implant were further sorted into pairs (n=14 per block) by weight. Pairs were assigned to 0, 28 or 56 d of feeding, within 28 and 56 d to M or A intake, and within 56 d of feeding, steers within a pair were randomly assigned to either 20 d of Z supplementation (90 mg/hd/d) with a 4d withdrawal period prior to slaughter or to no ZH supplementation (C). Steers were housed and fed in individual pens. Weights of all non-carcass and carcass components were recorded at slaughter; carcasses were graded 24h post-mortem. Data was analyzed via a mixed model; the fixed effect was treatment combination with random effects of block and pair. Live growth data used days on feed as the repeated measure and animal as the subject. Single df contrasts were constructed for d0 vs. d28, d0 vs. d56, d28 vs. d56, M vs. A and C vs. Z. Treatment impacted ($P \leq 0.05$) live ADG; contrasts indicated A (1.33) was greater than M (0.14 kg), and Z (1.12) was greater than C (0.82 kg). Similarly, carcass ADG differences ($P < 0.01$) indicated A (1.04) was greater than M

(0.36 kg), and Z (1.35) was greater than C (0.71 kg). Intake level altered BW and empty body weight (EBW); M cattle had lower BW and EBW ($P < 0.01$, 585 and 540 kg) than A cattle (647 and 597 kg). Cattle fed at M had less carcass and internal cavity mass ($P < 0.01$, 359 and 79.4 kg) than A cattle (394 and 93.5 kg). Liver mass was lowered by M feeding ($P < 0.01$; M-5.03, A-6.69 kg) and Z treatment ($P < 0.01$; Z-5.64, C-6.06 kg). Moreover, mass of total splanchnic tissue was less ($P < 0.01$) for M cattle than A cattle (59.8 vs. 72.5 kg). Dressed carcass yield was greater ($P < 0.01$) for Z than C cattle (63.5 vs. 61.6 %). Cattle fed at M had less 12th rib subcutaneous fat, lower U.S. yield grades ($P < 0.01$; M-1.71 cm and 3.3, A-2.46 cm and 4.3) and greater Canadian lean yield percentage ($P < 0.01$; 53.9 vs. 51.9% for M and A, respectively) than A cattle. Results indicate that days on feed, energy intake level and Z supplementation effect live performance, harvest yields and carcass grading factors.

4.2 Introduction

Zilpaterol hydrochloride (Z) is a FDA licensed β_2 -adrenergic agonist labeled for increased rate of weight gain, improved feed efficiency, and increased carcass leanness for cattle fed in confinement for slaughter during the last 20 – 40 days on feed (Merck Animal Health, 2006). Zilpaterol treatment results in improved live BW, ADG and gain:feed (Montgomery et al., 2008; Elam et al., 2009) while also increasing HCW, LM area, dressed carcass yield, and carcass cutability (Elam et al., 2009; Hilton et al., 2010). In addition, Z treatment results in increased rate of carcass weight gain compared to live weight gain thus dramatically improving carcass transfer (carcass ADG / live ADG) and ultimately maximizing economically important tissues at the end of the finishing period (Vasconcelos et al., 2008; Rathmann et al., 2012). Montgomery et al. (2008) and Holland

et al. (2010) hypothesized that non-car cass components might be catabolized in response to Z treatment to provide a pool of nutrients whereas muscle fibers are stimulated for growth. McEvers et al. (2013) discovered that in serially harvested Holstein steers supplemented with Z; non-car cass yields were reduced while car cass yields increased resulting in improved car cass transfer whereas Holland et al. (2010) did not detect differences in offal mass in beef steers supplemented with Z. Further examination of Z treatment is necessary to elucidate differential tissue response in non-car cass vs. car cass components.

Splanchnic tissues utilize a disproportionate amount of oxygen and dietary amino acids for their own metabolism relative to splanchnic tissue weight as a percentage of EBW (Reynolds et al., 1991; Loble y, 2003 and Baldwin et al., 2004). Furthermore, different dietary energy densities have resulted in differing visceral mass and subsequent changes to live performance and kill yields in the finishing period (McCurdy et al., 2010; Sharman et al., 2013). To date, no research has investigated the impact of Z treatment for cattle at maintenance on live performance, tissue yields, and car cass characteristics. Therefore, the objectives of this trial were to quantify live growth performance, non-car cass and car cass harvest yields, and car cass grading attributes of cattle fed differing energy levels and fed Z.

4.3 Materials and Methods

All experimental procedures involving live animals were approved by the Animal Care and Use Committee at West Texas A&M University and adhered to the regulations in the Guide for the Care and Use of Agricultural Animals in Agricultural Research and Teaching (Federation of Animal Science Societies, 2010, Savoy, IL).

4.3.1 Live Cattle Procedures

Single-sired steers (Hereford sire and Angus x Hereford dams) were sourced from one owner (two separate ranches), with all steers from first parity cows. Calves were born over a 21 d calving period and were weaned at 203 to 268 d of age. At weaning all calves were transported from the two ranches to Grandview, ID (205 and 160 km) and were vaccinated against bovine rhinotracheitis virus, bovine viral diarrhea virus type 1 and 2, bovine parainfluenza-3 virus and bovine respiratory syncytial virus (Pyramid 5, Boehringer Ingelheim Vetmedica, Inc., St. Joseph, MO), *Clostridium*' *chauvoei*, *septicum*, *hemolyticum*, *novyi*, *sordellii* and *perfringens* Types C&D (Vision 8, Merck Animal Health, Summit, NJ), treated for internal and external parasites with an injectable anthelmintic (Dectomax, Zoetis, Florham Park, NJ), administered a metaphylaxis treatment (Duramycin, Durvet Inc., Blue Springs, MO) and growth implant (Revalor G, Merck Animal Health). Steers were then backgrounded on a high roughage diet at a controlled rate of gain for 42 to 86 d. At the end of the backgrounding phase steers were weighed, administered a second growth implant (Revalor IS, Merck Animal Health) and adapted to a high concentrate finishing diet over 14 d. At the start of the finishing diet, steers ranged in age from 294 to 303 d. Steers were administered a terminal growth implant 90 d after the start of concentrate adaptation of either Revalor IS or Revalor XS (Merck Animal Health) depending upon estimated days to market. At terminal implant, DNA samples were collected from individual steers to ensure all candidate steers were of the same sire.

Steers (n=60; BW 574±36 kg) were selected and transported 1,944 km to a private research facility in Canyon, TX. Upon arrival, cattle were weighed immediately off the

truck and on the consecutive day prior to morning feeding. Cattle were placed on an intermediate level concentrate ration (73% concentrate) for 3 d prior to being transitioned back to a finishing diet (Table 1). Cattle were penned in two pens (n=30) by block; sort weights were recorded on d -5 and -6 (Block 1, initial BW = 589±25 kg and Block 2, initial BW = 614±39 kg). Steers were sorted into individual pens on d -4; Block 2 started the trial 28 days after Block 1 and remained penned as a group for the 28 d with parallel sorting and weighing protocols starting 28 d after Block 1. Within each block, two steers with weights furthest from the mean (heaviest and lightest steer in Block 1 and two lightest steers in Block 2) were penned in individual pens as alternates for the duration of the trial.

4.3.2 Experimental Design

The trial was constructed as a multi-factorial treatment design with three harvest dates, two dietary energy levels and Z supplementation or control (C). Cattle were blocked by terminal implant status and randomized to pairs on the sort days (d -5 and d -6) by weight. Pairs were randomized to harvest day (0, 28 or 56 d) and maintenance (M) or *ad libitum* (A) dietary energy level within harvest date. Within d 56 harvest date, one animal within each pair for both M and A level of feeding was randomly assigned to Z (90 mg per head per day for 20 d; Merck Animal Health) or control (no Z).

Maintenance adjustments were based on the NRC (1996) $\{((BW \times 0.891)^{0.75} \times 0.077)/\text{diet NE}_m\}$, BW d-1 and 1} with a positive 6.3% initial DM adjustment based on prior experience (Walter et al. 2015. *The effect of zilpaterol hydrochloride supplementation on energy metabolism, nitrogen and carbon retention of steers at maintenance and fasting intake levels*. Chapter I). Steers were weighed individually (d -

1, 11, 21, 27, 28, 55, 56) after 9h water and feed withdrawal. All cattle weights were recorded individually using four load cells mounted under a chute (Trojan Livestock Equipment, Weatherford, TX) and a GSE 350 digital weight indicator (Avery Weigh-Tronix Group, Fairmont, MN) with ± 0.45 kg accuracy; the scale was validated with 738 kg of weights before each use and checked with 270 kg of weights after weighing every 20 animals. Dry matter intake adjustments were made for maintenance steers using BW changes on d 11, 21, 27 and 28 with further as-fed intake for maintenance steers adjusted on d 1, 12, 22, 28, 40 and 50 based on daily DM 5 d averages (DM from d 5 to 9 for adjustment starting on d 12; DM from d 15 to 19 for adjustments starting on d 22; DM from d 22 to 25 for adjustments starting on d 28; DM from d 33 to 37 for adjustments starting on d 40 and DM from d 43 to 47 for adjustments starting on d 50). Steers given *ad libitum* access to feed were adjusted daily depending upon feed refusal and morning bunk calls at 0600. Orts were weighed and sampled on d 0, 27, 28, 55 and 56 in addition to daily weighbacks if feed refusal was visually estimated to be >5% of the previous days offering.

Diets were mixed in 900 kg batches with a mixer accuracy for ingredient loading to 4.5 kg. Feed samples were collected daily and sub-sampled for daily DM determination to a constant weight at 55°C, in a forced-air convection oven, (LBB 2-12, Despatch Industries, Minneapolis, MN). Daily sub-samples were composited (as-is) by week for further determination of nutrients. Cattle were fed once daily at 0800 with ration amounts weighed individually per pen on a scale with ± 0.045 kg accuracy, validated daily. Orts were dried at 55°C for 48 h and analyzed for nutrient composition. Vitamins, minerals, tylosin (9.9 mg/kg) and monensin sodium (33.1 mg/kg) were added

to the diet with an automated micro-ingredient machine (MicroBeef Technology, Amarillo, TX).

4.3.3 Harvest and Carcass Grading

Prior to harvest, steers were held in a separate pen without feed and water for 9 h. All steers transported 52 km and harvested at a commercial packing company (Est. ID 675; Hereford, TX). At harvest, weights of all harvest components (non-carcass and carcass) were recorded; samples of blood and hide (100 cm²) were retained for further analysis. Internal offal was collected in 208 L capacity drums, drum weights were tared and gross internal cavity component weights were captured at the plant (± 0.45 kg) prior to transportation to the WTAMU meat lab (Canyon, TX) for further processing. At the WTAMU meat lab, viscera was weighed ± 0.01 kg (ABM-60, Universal Weight Enterprise Company, Xindian City, Tawiiwan) individually (pancreas, liver, kidneys, gallbladder, thymus, kidney-pelvic-heart fat (KPH), gallbladder, trachea, heart, lungs and pluck trim). Omental fat was trimmed from the stomach compartments and weighed; the gastrointestinal tracts were cleaned, flushed of contents with pressurized tap water and weighed empty.

Twenty-four hours after harvest, a detailed carcass evaluation was conducted which included marbling score, lean and skeletal maturity, 12th rib subcutaneous fat depth (cm) and LM area (cm²). A final yield grade was calculated (USDA, 1997). In addition, Canadian grading factors were calculated for muscle score (maximum length and width) and grade fat (fat class) to calculate the Canadian lean yield percentage (Canadian Beef Grading Agency, 2001).

4.3.4 Sample Analysis

After drying of diet and orts to a constant weight at 55°C, in a forced-air convection oven, (LBB 2-12, Despatch Industries, Minneapolis, MN), all samples were ground through a 1-mm screen using a Wiley Mill. Feed and orts were analyzed for energy (Automatic Isoperibol Calorimeter, Parr Instrument Company, Moline, IL). A commercial laboratory (Servi-tech Labs, Amarillo, TX) analyzed dried and ground feed and ort samples for DM (AOAC #934.01), nitrogen (AOAC #990.03), starch (Xiong et al. 1990), ADF (Ankom Technology Method 5), NDF (Ankom Technology Method 6) and ether extract (AOAC #2003.06) while feed samples were also analyzed for Ca and P (AOAC #968.08).

4.3.5 Statistical Analysis

The mixed procedure of SAS (SAS Institute, Inc. Cary, NC) was used for model analysis; fixed effects were treatment combinations with random effects of block and pair. Single df contrasts of d0 v. d28, d0 v. d56, d28 v. d56, M v. A and C v. Z were constructed to examine differences between days on feed, dietary energy intake and Z supplementation. Performance data was analyzed as a repeated measures design with DOF as the repeated measure and individual animal as the experimental unit. Several variance-covariance structures were evaluated for each variable analyzed (ie. unstructured, simple, compound symmetry, heterogenous compound symmetry, first-order antedependence, first order auto-regressive, first order heterogenous auto-regressive and first-order auto-regressive moving average). Variance-covariance structures were chosen depending upon Akaike information criterion and Bayesian information criterion being closest to zero. The KENWARDROGER option was used to generate new

denominator degrees of freedom. An LSMEANS statement generated means and a PDIF statement was used to assess differences ($\alpha = 0.05$) and trends ($\alpha = 0.10$) due to DOF or ZH supplementation.

4.4 Results

4.4.1 Performance

Live growth performance differed across days on feed, dietary energy level and Z supplementation ($P < 0.05$; Tables 2 and 3). Start of trial BW did not differ across treatments ($P = 0.99$) but end of trial BW was greater in A than M cattle ($P < 0.001$; 647 vs. 585 kg). Additionally, cattle harvested on d 56 had a greater BW ($P = 0.04$) than cattle harvested on d 28 (628 vs. 593 kg, respectively) and tended to have a higher BW ($P = 0.07$) than cattle harvested on d 0 (586 kg).

Dry matter intake differed ($P < 0.01$) between diets as designed; A fed cattle consumed 55% more feed than M from d 1 to 28 and 53% more feed from d 29 to 56. Cattle on A intake consumed 1.46% of mid-point BW in the first 28 d. This is likely due to stress resulting from steers being penned individually as well as time off feed for weigh days. Body weight gain was affected by DOF, diet and Z supplementation (Table 3, $P < 0.01$). Gain was greater ($P < 0.01$) during d 29 to 56 vs. d 1 to 28 and in A vs. M fed cattle. Additionally, Z cattle had an increase in BW gain ($P = 0.05$) vs. control even though end of trial and start of trial BW was not different between ZH treatments. There was a weight difference at the beginning of d 29 to 56 period in which AC cattle had greater BW than AZ cattle (625 vs. 612 kg) and subsequently AZ cattle gained more BW during d 29 to 56 resulting in no difference to final BW (Table 2). The reduced performance in A fed cattle between d 1 to 28 and d 29 to 56 may be a result of isolation

stress of being penned separately or stress of multiple weigh days and therefore a difference in BW at the start of d 29 to 56 between AC and AZ cattle was not expected. Maintenance cattle from d 29 to 56 exhibited positive gains ($P < 0.01$) vs. weight loss expressed by M cattle d 1 to 28; this outcome is primarily due to weight loss in maintenance cattle during the first 28 days and a final BW in maintenance cattle similar to the start of trial weights.

Average daily gain increased ($P < 0.01$) over days on feed from 0.27 kg/d for d 1 to 28 to 0.97 kg/d during d 29 to 56 across all treatments and as expected was greater ($P < 0.01$) for A fed cattle (1.33 kg/d) vs. M fed cattle (0.14 kg/d) over the length of the trial. Average daily gain was also impacted by Z treatment ($P = 0.05$); d 29 to 56 AZ cattle had the greatest ADG (1.69 kg/d), though not different ($P > 0.05$) from d 29 to 56 AC cattle (1.26 kg/d) but greater than all other treatment groups. Maintenance cattle initially (d 1 to 28) had a negative ADG (-0.50 kg) but conversely a positive ADG during d 29 to 56 with (0.55 kg/d) or without (0.37 kg/d) ZH supplementation. Feed conversion efficiency (gain:feed) values were similarly influenced ($P < 0.01$) by days on feed and dietary intake level. Day 1 to 28 M cattle exhibited the lowest ($P = 0.05$) gain:feed (-0.125). Day 29 to 56 AZ cattle had the highest gain:feed (0.182) but were not different than d 29 to 56 AC (0.153) or d 29 to 56 MZ (0.126) treatments. The gain:feed of d 29 to 56 MZ cattle (0.126) did not differ ($P > 0.05$) from any A fed cattle whereas d 29 to 56 MC cattle had reduced ($P < 0.05$) gain:feed as compared to d 29 to 56 AC or AZ steers.

Carcass performance was also impacted by treatment ($P < 0.01$). Carcass gain increased ($P < 0.001$) over days on feed (7.2 vs. 20.3 kg, for d 1 to 28 and 29 to 56, respectively), with increased dietary energy intake (7.8 vs. 24.0 kg, for M vs. A,

respectively) and with Z treatment (29.5 vs. 12.6 kg, for Z vs. C, respectively). Carcass ADG differed across DOF, Z treatment and dietary energy level ($P < 0.01$); d 29 to 56AZ cattle exhibited the greatest (1.25 kg/d; $P < 0.05$) carcass ADG followed by d 1 – 28 A (0.76 kg/d) and d 29 to 56MZ cattle (0.75 kg/d). In addition, d 29 to 56MC and d 29 to 56AC cattle had similar carcass ADG of 0.33 and 0.57 kg/d, respectively. The lowest carcass ADG was evidenced in d 1 to 28 maintenance cattle (-0.24 kg/d). Carcass gain:feed was also affected by days on feed, dietary intake level and ZH treatment ($P < 0.01$). Carcass gain:feed of d 29 to 56 MZ (0.17) cattle was higher than all other treatments ($P < 0.05$) except d 29 to 56 AZ (0.14). Carcass transfer was greater ($P < 0.05$) in d 29 to 56 MZ steers (136.6%) than all other treatments and Z treated steers had increased carcass transfer vs. control steers (105.6% vs. 51%, respectively).

Biometric growth of steers was impacted by treatment ($P < 0.05$; Tables 2 and 3). Starting hip height tended ($P = 0.09$) to be less in d 0 steers than d 56 while end of trial hip height was lower in d 0 steers vs. all other treatments ($P < 0.05$). Additionally, M fed steers had reduced hip height vs. A steers ($P = 0.01$; 131 vs. 134 cm, respectively). Although, end of trial hip height differed between treatments, hip height gain (Table 3) did not differ by treatment ($P = 0.20$) but did range from 0.11 (d 1 to 28M) to 0.59 (d 29 to 56AC) mm/d. Start of trial shoulder height did not differ ($P = 0.33$) across treatments whereas end of trial shoulder height tended to be impacted by treatment ($P = 0.06$). Day 56 steers had taller shoulders than d 0 steers (128 vs. 121 cm, respectively) and tended to have taller shoulders than d 28 steers (125 mm). Shoulder height gain was greater ($P < 0.01$) for A (0.8 mm/d) than M (0.2 mm/d) steers.

4.4.2 Empty Body Weight and Tissue Component Yields

Final BW, empty body weight (EBW) and tissue component yields were impacted by treatment ($P < 0.05$; Table 4). Final BW increased ($P = 0.05$) from d 28 to d 56 (593 to 627 kg) whereas EBW tended ($P = 0.09$) to increase during the same time period (551 to 577 kg). Differences in final BW and EBW also occurred between M and A fed cattle ($P < 0.01$); as expected, A fed cattle had heavier final BW and EBW (62 and 56 kg difference, respectively) vs. M steers. In addition, day 56 AZ cattle had the heaviest final BW and EBW of 666 and 615 kg, respectively, but were not different ($P > 0.05$) from the BW and EBW of d56 AC cattle (658 and 602 kg, respectively). Day 56 AC cattle were not different from d 28 A (573 kg), with respect to EBW, but were different from all other treatments ($P < 0.05$). The ratio of EBW/BW tended ($P = 0.08$) to differ across treatments; d28 was 0.86% greater ($P = 0.05$; 92.1 vs. 93.0%, respectively) than d56, and Z was 1.2% greater ($P = 0.01$; 92.7 vs. 91.5%, respectively) than C. Conversely, fill followed the opposite trend and ranged from 7.0 to 8.5% of BW.

Tissue component yields of HCW and non-carcass differed across treatments ($P < 0.01$, Table 4). Hot carcass weight increased between ($P = 0.03$) d 0 and 56 by 30 kg (354 vs. 384 kg, respectively) and between d 28 and 56 by 23 kg (361 and 384, respectively). In addition, A fed cattle exhibited a 35 kg heavier carcass than maintenance cattle ($P < 0.01$; 394 vs. 359 kg, respectively) and cattle supplemented with Z had a tendency ($P = 0.07$) for a heavier HCW (17 kg) vs. C cattle (393 vs. 376 kg, respectively). The 56 d AZ cattle exhibited a heavier ($P < 0.05$) HCW compared to both 56 d MZ and 56 d MC whereas the 56 d AC did not have a heavier ($P > 0.05$) HCW vs. the 56 d MZ cattle and the 28 d A cattle. Zilpaterol hydrochloride supplementation

resulted in increased carcass tissue yield ($P < 0.01$) vs. C cattle (673 vs. 658 g/kg EBW). Day 56 AZ and day 56 MZ cattle had greater ($P < 0.05$) HCW (670 and 677 g/kg EBW, respectively) than all other treatments except 56 d MC (661 g/kg EBW). Additionally, d 56 cattle had greater ($P < 0.001$) carcass yield vs. d 0 (665 vs. 651 g/kg EBW, respectively) and d 28 (665 vs. 655 g/kg EBW, respectively). Blood and hide did not differ across treatments ($P \geq 0.16$). In contrast, mass of non-carcass bone (hooves, metacarpals, metatarsals and oxtail) was impacted ($P = 0.02$) and head mass tended to be impacted ($P = 0.06$) by days on feed; d 56 cattle had heavier head and non-carcass bone than d0 or d28 cattle ($P \leq 0.05$). When non-carcass bone and head weights were expressed as g/kg EBW both variables were decreased by increased dietary intake level ($P < 0.01$). Furthermore, Z supplementation decreased ($P = 0.02$) non-carcass bone weight (g/kg EBW) as compared to C cattle. Harvest trim included cod fat, additional fat trim from carcass (around hide entry points), spinal cord and penis, and was impacted by intake level. Cattle fed at M intake had reduced ($P < 0.01$) trim relative to A fed cattle (13.1 vs. 15.1 kg, respectively). More specifically, M cattle at d 56 had reduced ($P < 0.05$) trim compared to A fed cattle at d 56 irrespective of Z supplementation (12.84 vs. 15.44, respectively), likely due to a decrease in cod fat.

Internal cavity as a whole weight was impacted ($P < 0.01$) by dietary intake level; M cattle exhibited reduced ($P < 0.01$) internal cavity mass compared to A fed cattle (79.4 vs. 93.5 kg). Internal cavity mass as a fraction of EBW was impacted by dietary intake level ($P < 0.01$; M-147, A-155 g/kg EBW) and Z supplementation ($P < 0.01$; C-152, Z-145 g/kg EBW). Moreover, at d 56, cattle had less internal cavity g/kg EBW when compared to d 28 ($P = 0.02$; 149.48 vs. 156.43). Total non-carcass weight was impacted

by treatment ($P < 0.001$) when expressed as an absolute mass, g/kg of EBW, or g/kg of BW. On an absolute weight basis, M level of intake reduced ($P < 0.01$) total non-carcass components compared to A feeding (181.35 vs. 202.76 kg, respectively). In addition 28 and 56 d M cattle had reduced ($P < 0.05$) non-carcass components relative to 56 d A intake cattle. When expressed on a relative basis to EBW and BW, non-carcass component weight was decreased over days on feed from d0 to d56 ($P < 0.01$) and d28 to d56 ($P < 0.01$); Z supplementation also decreased non-carcass weights expressed as g/kg of EBW and BW ($P < 0.01$).

4.4.3 Individual Harvest Yields

Harvest yields of kidneys, stomachs (reticulorumen, omasum and abomasum), intestines (small and large intestines and cecum), spleen, liver, pancreas, omental fat and kidney-pelvic-heart fat (KPH) as well as total gastrointestinal tract (GIT, sum of stomach and intestines) and total splanchnic tissue (sum of GIT, liver, spleen, gallbladder and pancreas) with and without KPH and omental fat were impacted by treatment on an absolute basis (TST and TST_{FAT}) ($P < 0.05$, Table 5). Kidney weight was impacted by dietary intake level ($P < 0.01$) with A cattle having larger kidneys than M fed cattle (1.11 vs. 0.97 kg, respectively). Stomach, intestines and GIT were increased ($P < 0.01$) in A relative to M fed cattle (A-43.87, M-37.49 kg). Additionally, d 56 cattle tended ($P = 0.07$) to have less total GIT vs. d 28 cattle (39.74 vs. 42.57 kg, respectively). Spleen weights were impacted by treatment with d28 and d56 cattle having heavier ($P < 0.05$) spleens than d0 cattle. In addition, M cattle tended ($P = 0.08$) to have a lighter spleen than A fed cattle. The liver was greatly impacted by treatment; d 28 and d56 cattle had lighter ($P < 0.01$) livers than d 0 cattle; primarily driven by the large decrease in liver

weight in cattle fed M intake levels ($P < 0.001$; M-5.03, A-6.69). Liver weight was also impacted by Z supplementation ($P = 0.04$) with Z supplemented cattle having a reduced liver weight to C cattle (5.64 vs. 6.05 kg, respectively). Pancreas weight was impacted by days on feed with d 28 and d 56 cattle having an increased pancreas weight vs. d0 cattle ($P < 0.01$). Moreover, Z supplementation tended ($P = 0.10$) to decrease pancreas weight; AZ cattle had reduced ($P < 0.05$; 0.45 vs. 0.57 kg) pancreas weight compared to AC cattle. As a result, TST weight (not including fat) was decreased ($P < 0.001$) by dietary intake level with M fed cattle having decreased TST weight compared to A fed cattle (44.35 vs. 52.66 kg, respectively). Omental and KPH weights were affected ($P < 0.01$) by dietary intake level; A cattle had greater internal fat than M fed cattle (19.87 vs. 15.46 kg, respectively). Weight of KPH also tended to be decreased by Z supplementation ($P = 0.08$); Z supplemented cattle had less KPH than C cattle (7.72 vs. 8.73 kg, respectively). Total splanchnic tissue including omental fat was decreased ($P < 0.001$) in M as compared to A fed cattle (M-52.69, A-63.73 kg).

Comparing individual viscera and offal yields on a g per kg of EBW basis yielded similar trends to the absolute weight for most components (Table 4). Trachea and heart were impacted ($P < 0.05$) by treatment on a g/kg EBW basis whereas actual weights were not different between treatments. Neither Spleen nor KPH exhibited differences ($P \geq 0.11$) when expressed as g/kg EBW. Trachea weight per unit of EBW was not impacted by a specific treatment but was lower ($P = 0.05$) in 56 d A cattle vs. 28 d M and 56 d MZ cattle. Heart weight was affected by treatment ($P < 0.01$) with M cattle expressing greater weight as g/kg EBW compared to A fed cattle ($P < 0.01$, 4.6 vs. 4.2 g/kg EBW, respectively). Kidney was impacted by days on feed, when expressed as g/kg EBW ($P <$

0.05); d 56 cattle exhibited a reduced kidney weight on an EBW basis vs. d 28 and d0 cattle. Additionally, M and Z cattle had smaller kidney weights as g/kg EBW compared to A and C cattle, respectively ($P < 0.05$). Stomachs, intestines and total GIT as g/kg of EBW was affected ($P < 0.01$) by treatment; stomach weight was reduced in d 0 cattle and d 56 cattle compared to d 28 cattle. Similarly, M cattle had less stomach per unit of EBW than A cattle ($P = 0.02$) and Z supplemented cattle had less than C cattle ($P = 0.03$). Intestinal weight was reduced in a similar fashion to stomach; d 56 cattle exhibited reduced intestinal weight as g/kg EBW vs. d0 and d28 cattle and Z supplemented cattle had decreased ($P = 0.04$) intestinal weight compared to C cattle. A tendency ($P = 0.09$) was detected between M and A cattle for reduced intestinal mass as g/kg EBW. Total GIT weight as g/kg EBW was impacted by dietary intake level ($P = 0.01$) and Z supplementation ($P = 0.01$); M and Z supplemented cattle had reduced GIT mass as g/kg EBW. Additionally, total GIT weight was reduced in d 56 cattle compared to d0 and d28 cattle ($P < 0.01$). Liver was decreased ($P < 0.01$) by increasing DOF ($P < 0.01$) as well as by M dietary intake level ($P < 0.01$) and Z supplementation ($P < 0.01$). Pancreas weight was reduced in d0 compared to d 28 and d56 cattle ($P < 0.05$) and tended ($P = 0.06$) to be reduced in Z supplemented vs. C cattle. Omental fat and total omental + KPH fat was reduced ($P < 0.01$) in M vs. A fed cattle. Total splanchnic tissue with and without omental fat was impacted by treatment ($P < 0.001$) when expressed on g/kg EBW basis with d 56 cattle having a reduced TST mass compared to d 0 and d 28 ($P < 0.05$) as well as M intake and Z supplementation causing decreased TST ($P < 0.01$).

4.4.4 Carcass Traits

Carcass traits utilizing both USA and Canadian grading factors were impacted by treatment ($P < 0.05$, Table 6). Hot carcass weight was affected ($P < 0.05$) by days on feed, dietary energy level and zilpaterol hydrochloride (Table 5) in a parallel fashion as HCW without KPH (Table 4). Dressed carcass yield was also impacted by treatment ($P < 0.01$); carcass yield tended to increase from d0 to d56 ($P = 0.08$; 61.7 vs. 62.6%, respectively). Zilpaterol supplementation also increased carcass yield as compared to C cattle ($P < 0.01$, 63.7 vs. 61.6, respectively). Carcass yield when calculated with a 4% pencil shrink ($BW \times 0.96$) applied across live BW concurrent with industry standards resulted in 64.3, 65.0, 64.5, 65.8, 64.0, 66.8 and 64.3% for d0, d28 A, d28 M, d56 AZ, d56 AC, d56 MZ and d56 MC cattle, respectively.

Individual carcass traits (adjusted 12th rib subcutaneous fat thickness, USDA calculated yield grade, Canadian fat class and Canadian calculated lean yield percentage) differed across treatments ($P < 0.05$, Table 5) while KPH % tended ($P < 0.10$, Table 5) to differ across treatments. Adjusted 12th rib s.c. fat thickness was reduced ($P < 0.05$) in M compared to A steers (7.0 vs. 8.6, respectively) and between d0 vs. d56 (6.9 vs. 8.0, respectively) whereas LM area was not impacted by treatment ($P = 0.53$). The percentage of kidney pelvic heart fat (KPH) was decreased ($P = 0.02$) in Z supplemented cattle (1.93%) compared to C cattle (2.26%) and was less ($P = 0.05$) for M than A fed cattle (1.94 vs. 2.19%, respectively). As a result, calculated USDA yield grade was increased from d 0 to d 56 ($P = 0.05$; 3.3 vs. 3.9, respectively) and by increased dietary intake ($P < 0.01$; 3.3 vs. 4.2 for M and A fed steers, respectively), similar to 12th rib subcutaneous fat thickness.

With respect to individual Canadian grading traits, fat class and yield grade were affected by treatment ($P < 0.01$). Canadian fat class was impacted in a similar fashion as adjusted 12th rib fat thickness as cattle fed M had reduced fat class vs. A fed steers (7.0 vs. 8.6, respectively) while d0 steers also had reduced fat class vs. d56 (6.9 vs. 8.0, respectively). Canadian calculated lean yield percentage (CANYG) was impacted by treatment ($P = 0.03$) with A steers having reduced lean yield vs. M ($P < 0.01$; 51.9 vs. 53.9%, respectively). Marbling scores, skeletal maturity and color scores were not impacted by treatment ($P \geq 0.38$).

4.5 Discussion

Dry matter intake in this trial across A intake level averaged 8.88 kg whereas M cattle consumed 4.12 kg. Intakes for A cattle were 1.46, 1.40 and 1.38% of mid-point BW (unshrunk) for d 1 – 28, d 29 – 56 ZH and d 29 – 56 control, respectively. Previous research conducted on long term feeding trials in Angus steers fed *ad libitum* (70% concentrate rations) resulted in 1.28% of mid-point BW from 615 to 734 kg BW (Bond et al., 1982). Other trials with comparable DMI during the end of the finishing trial in steers include Vasconcelos et al. (2008) in cattle fed a finishing diet for 177 and 198 d that reported DMI of 1.48 (8.60 kg) and 1.47% (9.03 kg), respectively of mid-pt BW during the last 43 days and Montgomery et al. (2008) that reported an average DMI of 8.76 kg across both Z and C cattle during the last 35 days. Maintenance DMI were increased (in addition to the positive 6.3% initial adjustment) during the first 28 d by $4.76\% \pm 3.25$ for Block 1 and $2.74\% \pm 2.73$ for Block 2 from the initial calculated value based on weight loss. Maintenance steers were fed 45% of A intake during d 1 to 28 and 47% of A intake during d 29 to 56. With respect to performance, A cattle gained 1.03

kg/d during d 1 to 28 and an average of 1.48 kg/d during d 29 to 56 whereas M fed cattle lost 0.50 kg in the first 28 days after which M steers incurred a slightly positive BW during d 29 to 56. Vasconcelos et al. (2008) in extended feeding of steers for 177 and 198 to a final BW of 610 and 644 d reported an ADG of 1.31 and 1.40 for the last 43 days on feed, similar to the results of this trial during d 29 to 56. The lower ADG of 1.03 kg/d during d 1 to 28 is likely due to a loss of fill at the start of trial with multiple weigh days off feed and water as well as the stress of being penned individually.

With respect to Z supplementation effects on live performance, results from this trial are similar to previous research for improvements in live BW and ADG. Steers fed A intakes had 13 kg higher end BW, 34 % improvement in ADG and, whilst not significant, a 19.0% improvement in gain:feed, compared to AC fed steers. Additionally, steers fed M and supplemented Z had 6 kg more end of trial BW, a 48% improvement in ADG and a 37.0% improvement in gain:feed vs. MC steers. Although gain:feed was not different for Z vs. C ($P > 0.10$) this may be due to limited animal numbers ($n = 8$ per treatment) in the current study. Previous studies reported similar improvements to final BW of 8 kg and 10.6 kg when Z was fed for 20 d (Elam et al., 2009; Vasconcelos et al., 2008). Previous studies reported similar improvements of 33% and 26% to ADG and gain:feed when Z was fed at 6 mg/kg for 40 d (Plascencia et al., 2008) and 44% and 47% improvement in ADG and gain:feed, respectively when Z was fed at 8.3 mg/kg for 20 d (Montgomery et al. 2009). When Z was fed for 20 d, Vasconcelos et al. (2008) and Elam et al. (2009) reported improvements of 11 and 16% for ADG and 10 and 16% for gain:feed, respectively. The lesser response in the latter two trials (Vasconcelos et al. 2008; Elam et al. 2009) may be due to performance data including the last 43 and 50 days

of feeding performance, respectively, thus diluting the 20 d Z period improvement with additional time.

Carcass performance in the current study reflected changes to carcass ADG and gain:feed over days on feed, dietary plane of intake and Z supplementation. Carcass ADG and carcass gain:feed was increased in cattle supplemented with Z, regardless of plane of intake. Cattle fed A intake levels and supplemented with Z had a 119 and 50% improvement in carcass ADG and gain:feed, respectively vs. AC steers whereas M fed steers supplemented with Z exhibited a 127 and 89% improvement in carcass ADG and gain:feed, respectively vs. MC steers. Therefore, Z exerted a large impetus for increased skeletal muscle accretion regardless of plane of intake, perhaps due to catabolism of non-carcass components in favor of nutrients for carcass components (Holland et al., 2010; McEvers et al., 2013). Rathmann et al. (2012) reported improvements of carcass ADG and gain:feed due to Z in heifers of 34 and 36%, respectively, similar to A fed cattle during d 1 to 28 in the current study. Carcass transfer in the current study (carcass ADG / live ADG) was highest in d 29 to 56 MZ treated cattle (137%) whereas d 29 to 56 MC cattle had a carcass transfer of 57%; some of the increase in transfer was due to tissue weight loss during d 1 to 28 (d 1 to 28 M carcass transfer of 44%) and an increase in efficiency over time resulting in a positive energy balance during d 29 to 56 in M cattle. Carcass transfer of d 29 to 56 AZ cattle was 75% whereas in d 29 to 56 AC cattle, carcass transfer was 45%. Similar to A fed cattle in the current study, Rathmann et al. (2012) reported a carcass transfer of 73% for control fed heifers and 89% for Z supplemented heifers during the treatment period. The low carcass value of d 28 to 56 cattle may have

been due to large variation in dressing percentage as d 1 to 28 A had a high carcass transfer of 86%.

Biometric data of hip height and withers height were collected in this trial. Nkrumah et al. (2004) reported an average hip height of 127.5 cm in the last 70 days of feeding for cattle with a final body weight of 515 kg and reported no difference between hip heights or hip height gain in steers with different residual feed intake classifications. Bergen et al. (2006) reported weak correlations of hip height to HCW ($r = 0.16$) when measured in yearling bulls. The current study details changes between start and end of period hip height over days on feed, although hip height gain was not different, potentially as a result of large variation between steers in a limited (56 d) time span. Dietary intake level impacted shoulder height gain. This may be indicative of limited calories and negative feedback on continued frame growth. Bond et al. (1982) reported Angus steers fed A level of intake slaughtered at 18 months had a shoulder height of 115.0, 3.2 cm greater than cattle fed M intake and slaughtered at the same age. The steers used in the current trial vs. those used by Bond et al. (1982) are likely to be of different genetic capacity for growth, even though both studies utilized British based breeds. Bond et al. (1982) recorded a BW at 18 months of 305 kg and a shoulder height of 115.0 cm in steers fed a 70% concentrate ration at A intake levels. Thus, cattle in the current study harvested at a similar age were 300 kg heavier and 10 cm taller at the shoulder than cattle over three decades ago. Regardless of treatment, both hip and shoulder height increased with longer days on feed, indicating the ability of the animal on high plane of nutrition diet to increase in frame size.

In the current study, as days on feed increased, carcass weight increased as a fraction of EBW and subsequently, non-carcass weight decreased as a fraction of EBW. Carcass weight increased as a fraction of EBW with Z supplementation and exhibited a tendency to increase with reduced dietary intake. Holland et al. (2010) did not report an impact of Z supplementation on carcass weight as a fraction of EBW nor an effect on total non-carcass components. Discrepancy due to Z supplementation is likely due to the reduced head count used by Holland et al. (2010) of three steers per pen and pen used as the experimental unit. Other research has reported growth of internal cavity and non-carcass components becomes non-linear with age and carcass components steadily increase as a proportion of empty body weight (Carstens et al. 1991). In the current study, tissue component weights of hide and blood remained constant over days, diet and Z supplementation, conversely non-carcass bone increased over days, indicative of a growing animal. Internal cavity also increased as a fraction of EBW over days whereas M intake level and Z supplementation reduced internal cavity weight as a fraction of EBW. Similar to the current study, Hutcheson et al. (1997) reported total organ mass as a percentage of EBW decreased in estrogen and combination implanted steers.

Total non-carcass weights were not changed by increasing days as carcass weights were but rather were reduced by M intake level. When represented as a fraction of EBW, total non-carcass decreased over days and also due to reduced dietary intake level and Z supplementation. During the first 28 days, A cattle produced an additional 22 kg of carcass but only an additional 7.59 kg of non-carcass weight. Total non-carcass components remained similar to C cattle harvested at the start of the trial. Control, A cattle had heavier total non-carcass components than d0 cattle but as a fraction of EBW,

no difference was detected. Thus, A cattle not supplemented with Z appear to remain constant in depositing the same fraction of carcass and non-carcass components, regardless of days on feed whereas cattle supplemented with Z and placed on a M plane of intake had reduced non-carcass components and increased carcass components as a fraction of EBW over time.

Visceral organ and tissues were changed with treatment including kidneys, stomachs, intestines, spleen, liver, pancreas and internal fat whereas further changes (trachea and heart) occurred when represented as weight per kg of EBW. In the current study, a reduction of dietary energy intake to M level resulted in changes to stomachs, intestines, total gastrointestinal tract, liver, omental fat, kidney pelvic heart fat and total splanchnic tissue with and without fat as well as tendencies for reduction in spleen weight. Researchers including Reynolds et al. (1991) and Lobley (2003) have stated that the visceral organ mass and largely the splanchnic tissues consume a drastic amount of energy in the animal with metabolic inefficiencies resulting from large GIT mass, poorly digestible diets and more complex items including individual amino acid and volatile fatty acid uptake of tissues for the tissues own energy needs. Furthermore, the splanchnic tissues have proven to be incredibly dynamic, with changes in size and efficiency over time due to level of dry matter intake and plane of nutrition (Reynolds et al. 1991). Total splanchnic tissue mass, without omental fat, reduced 2.14 kg from d 1 to 28 on M intake whereas cattle fed A intake increased weight by 5.55 kg during the same 28 d period. During the following 28 d period, TST without fat decreased an additional 2.02 kg in MC cattle and an additional 4.46 kg in MZ cattle. In A cattle TST without fat stayed constant (-0.9 kg) whereas in AZ fed cattle TST without fat decreased by 3.73 kg. When TST

included omental fat (TST_{fat}), M plane of intake reduced TST_{fat} by 3.18 kg in the first 28 d period while increasing TST_{fat} by 6.18 kg in A cattle. During d 28 – 56, TST_{fat} decreased 2.17 kg in MC steers and 4.31 kg in MZ steers from 28 d M steers and also decreased 3.29 kg in AZ steers, and increased 0.86 kg in AC steers from 28 d A steers. As a result of dietary intake reductions and Z supplementation, differences in visceral tissues on a g/kg EBW basis occurred over days on feed with d 56 cattle having reduced kidney, stomach, intestines, GIT, liver and TST vs. d 28 and with d 56 cattle having reduced kidney, intestines, GIT, liver, pancreas and TST vs. d 0 cattle. Prior research analyzing non-carcass components due to different repartitioning effects have reported a reduction in internal fat in lambs fed cimaterol (Hanrahan et al., 1987). Hutcheson et al. (1997) reported decreased gastrointestinal tract mass as a percentage of EBW resulting from estrogen or combination estrogen/trenbolone acetate implants. Conversely, Hutcheson et al. (1997) reported increased liver percentage with implants vs. unimplanted steers. Differences in liver weights with implants may potentially be a result of estrogenic implants stimulating insulin-like growth factor which acts on the liver (Hannon et al., 1991) vs. beta-adrenergic agonists used in the current study which has a direct, cell mediated effect (Johnson et al., 2014).

The impact of Z on visceral tissue mass appears to be in addition to the impact of M plane of intake on reducing visceral tissue mass. The reduction in tissues associated with reduced dietary energy level has been hypothesized to be a result of decreased passage time thereby improving rumen microbial energy efficiency, increased total tract digestibility and therefore improved digestible energy of the ration and improved clearance of nutrients from the portal drained viscera. The additional decrease in visceral

tissue mass with Z was not due to a reduction in dry matter intake because that was not experienced in the current study nor is it due to an improvement in total tract digestibility at A or M levels (Brake et al., 2011 and Walter et al., 2015. *The effect of zilpaterol hydrochloride supplementation on energy metabolism, nitrogen and carbon retention of steers at maintenance and fasting intake levels*. Chapter I). It is plausible that Z exerts a direct stimulus on TST resulting in improvements to protein turnover and efficiency and therefore a reduced mass necessary to support the tissues own needs as well as the need to metabolize nutrients and support additional maintenance and gain functions. The ability of Z to stimulate similar improvements in HCW and dressing percentage with concurrent reductions in visceral tissue mass at M level of intake is likely indicative of adjustments to cattle maintenance and gain requirements.

Carcass traits of HCW (with KPH), dressed carcass yield, adjusted 12th rib subcutaneous fat thickness and USDA yield grade increased over time on feed. Hot carcass weight and dressed carcass yield increased over days on feed with 56 d AZ cattle exhibiting the heaviest HCW (420 kg) and 56 d MZ treatment resulting in the highest dressing percentage (64.1%). Previous research using Z (20 d) in steers resulted in an average improvement of 15.7 kg and a 1.45% to HCW and dressing percentage, respectively (Vasconcelos et al., 2008; Elam et al., 2009; Montgomery et al., 2009; Parr et al., 2011). While only a tendency existed for Z to increase HCW in the current study (16 kg, regardless of dietary intake level), results are very similar to prior research. Dressed carcass yield in the current study improved 1.8% and 2.4% with Z supplementation between 56 d A or M fed cattle, respectively. The additional HCW in

the 56 d M cattle is indicative of energy being repartitioned from other sources or a reduction in maintenance energy requirements due to Z supplementation.

With respect to rib fat thickness, AC cattle accreted 0.52 and 0.57 cm of 12th rib s.c. fat (adjusted) from d 1 – 28 and d 29 – 56, respectively. Cattle fed A intake level and supplemented with Z accreted 0.37 cm of 12th rib subcutaneous fat (adjusted) during the last 28 days on feed. While no difference was found for adjusted 12th rib subcutaneous fat thickness in cattle supplemented with Z, dietary intake level was significant with M cattle at 28 or 56 d not different from d 0 cattle. Canadian fat class was impacted in a similar fashion as adjusted 12th rib s.c. fat thickness with A cattle having increased Canadian fat class vs. M cattle as well as fat class to in 56 d than d0 cattle. Both 12th rib s.c. fat thickness and Canadian fat class are measures of 12th rib s.c. fat depth.

Subcutaneous fat deposition likely dissipated in M fed cattle because fat deposition requires a positive plane of energy. Canadian ribeye length and width factors nor LM area were impacted by treatments, regardless USDA and Canadian calculated yield grades were higher in A vs. M cattle primarily due to the increased backfat thickness in A cattle. Marbling score did not differ in M fed cattle regardless of days on feed or Z supplementation. Ultimately, cattle fed M did not appear to accrue nor catabolize subcutaneous or intramuscular fat depots as an energy source even though HCW increased in M cattle fed Z.

Results of the current study illustrate prominent effects of dietary energy intake level to live and carcass performance and kill yields. The effect of Z on carcass performance and numerical reduction in TST mass occurred irrespective of dietary energy intake. The effect of Z on non-carcass and carcass components at both energy

intake levels may be indicative of shifting maintenance or gain energy requirements.

Further research is needed to elucidate the efficiency of maintenance and gain in cattle supplemented with Z. Additionally, further research needs to delve into the prediction of empty body and carcass physical and chemical composition utilizing live and carcass grading factors.

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Table 4.1. Dietary ingredients (% DM basis), and nutrient basis (DM basis) of finishing rations fed to steers.

Ingredients, % DM basis	
Steam Flaked corn	72.32
Corn Gluten Feed	9.38
Wheat Hay	9.59
Molasses Blend	1.20
Fat	2.35
Supplement	5.26
Nutrient Analysis (\pm std deviation)	
DM, %	79.92 ± 0.011
Crude Protein, % DM	13.28 ± 0.659
Starch	51.25 ± 2.153
NDF, % DM	16.38 ± 0.774
ADF, % DM	7.63 ± 0.405
Ether extract, % DM	5.72 ± 0.298
Calcium, % DM	0.52 ± 0.042
Phosphorus, % DM	0.34 ± 0.016

Table 4.2. Live weights and biometric data of steers fed to 0, 28 or 56 days on feed (0, 28 and 56) and given maintenance (M) or ad libitum (A) intake and control (C) or zilpaterol hydrochloride (Z) treatment.

Item	0	28 A	28 M	56 AZ	56 AC	56 MZ	56 MC	SEM	P-value	0 v. 28	0 v. 56	28 v. 56	M v. A	C v. Z
<i>N</i>	8	8	8	8	8	8	8							
Start of Trial BW ¹ , kg	586	586	582	597	589	595	594	16.1	0.99					
End of Trial BW ¹ , kg	586 ^b	617 ^{ab}	569 ^b	667 ^a	658 ^a	595 ^b	591 ^b	17.5	<0.01	0.80	0.07	0.04	<0.001	0.68
<i>Biometric Data</i>														
Start Hip Height, cm	127	132	128	131	132	130	133	1.2	0.09	0.16	0.02	0.21	0.26	0.13
End Hip Height, cm	127 ^c	133 ^{ab}	130 ^{bc}	133 ^{ab}	136 ^a	131 ^b	133 ^{ab}	1.4	<0.01	0.01	<0.001	0.10	0.01	0.07
Start Shoulder Height, cm	121	125	121	124	125	123	127	1.7	0.33					
End Shoulder Height, cm	121	124	126	128	129	127	127	1.7	0.06	0.11	<0.01	0.06	0.65	0.61

^{a-c} Least squares means within a row with differing superscripts differ ($P \leq 0.05$)

¹ BW is unshrunk because animals were off feed and water 9 h

Table 4.3. Live performance and biometric gain of steers fed for 0 – 28, or 28 – 56 days and given maintenance (M) or ad libitum (A) intake and control (C) or zilpaterol hydrochloride (Z) treatment.

Item	Treatments						SEM	P - value	Contrast P - values		
	1-28 A	1-28 M	29-56 AZ	29-56 AC	29-56 MZ	29-56 MC			DOF	M vs A	C vs Z
<i>N</i>	24	24	8	8	8	8					
<i>Live Performance</i>											
DMI, kg	8.85 ^a	4.01 ^b	8.96 ^a	8.83 ^a	4.22 ^b	4.13 ^b	0.227	<0.01	0.28	<0.01	0.57
BW gain, kg	28.9 ^b	-14.1 ^d	47.4 ^a	35.2 ^b	15.5 ^c	10.3 ^c	3.56	<0.01	<0.01	<0.01	0.05
ADG, kg	1.03 ^b	-0.50 ^d	1.69 ^a	1.26 ^{ab}	0.55 ^c	0.37 ^c	0.128	<0.01	<0.01	<0.01	0.05
Gain:Feed	0.113 ^{bc}	-0.125 ^d	0.182 ^a	0.153 ^{ab}	0.126 ^{abc}	0.092 ^c	0.021	<0.01	<0.01	<0.01	0.19
<i>Carcass Performance</i>											
Carcass gain, kg	21.2 ^b	-6.79 ^d	34.9 ^a	15.9 ^{bc}	21.0 ^b	9.2 ^c	2.65	<0.01	<0.01	<0.01	<0.01
Carcass ADG, kg	0.76 ^b	-0.24 ^d	1.25 ^a	0.57 ^{bc}	0.75 ^b	0.33 ^c	0.110	<0.01	<0.01	<0.01	<0.01
Carcass gain:feed	0.08 ^c	-0.06 ^d	0.14 ^{ab}	0.07 ^c	0.17 ^a	0.09 ^{bc}	0.017	<0.01	<0.01	0.02	<0.01
Carcass transfer ¹ , %	85.6 ^b	44.4 ^d	74.6 ^{bc}	44.9 ^{cd}	136.6 ^a	57.1 ^{bcd}	11.06	<0.01	0.08	0.22	<0.01
<i>Biometric Data</i>											
Hip height gain (mm/d)	0.48	0.11	0.33	0.59	0.22	0.18	0.205	0.20			
Shoulder height gain (mm/d)	0.13 ^b	0.46 ^{ab}	1.13 ^a	1.13 ^a	0.33 ^{ab}	-0.13 ^b	0.306	0.05	0.28	<0.01	0.38

^{a-d} Least squares means within a row with differing superscripts differ ($P \leq 0.05$)

¹ Carcass transfer = (HCW ADG / Live ADG) * 100

Table 4.4 Empty body and tissue yields of steers fed for 0 – 28, or 28 – 56 days and given maintenance (M) or ad libitum (A) intake and control (C) or zilpaterol hydrochloride (Z) treatment.

Item	0	28 A	28 M	56 AZ	56 AC	56 MZ	56 MC	SEM	P-value	0 v. 28	0 v. 56	28 v. 56	M v. A	C v. Z
<i>N</i>	8	8	8	8	8	8	8							
Final BW, kg	586 ^b	615 ^{ab}	570 ^b	666 ^a	658 ^a	593 ^b	591 ^b	18.8	<0.001	0.80	0.08	0.05	<0.001	0.74
Empty BW, kg	543 ^b	573 ^{ab}	529 ^b	615 ^a	602 ^a	552 ^b	540 ^b	15.89	<0.01	0.74	0.10	0.09	<0.001	0.36
EBW/Final BW, %	92.7	93.2	92.8	92.5	91.5	93.0	91.5	0.48	0.08	0.68	0.26	0.05	0.89	0.01
Fill, %	7.3	6.8	7.2	7.5	8.5	7.0	8.5	0.46	0.08	0.68	0.26	0.05	0.89	0.01
Carcass ¹														
Kg	354 ^c	376 ^{bc}	346 ^c	412 ^a	394 ^{ab}	373 ^{bc}	357 ^c	10.8	<0.001	0.65	0.03	0.03	<0.001	0.07
g/kg of EBW	651 ^d	656 ^{cd}	655 ^{cd}	670 ^{ab}	654 ^{cd}	677 ^a	661 ^{bc}	4.3	<0.001	0.26	<0.001	<0.001	0.09	<0.001
g/kg of BW	604 ^c	611 ^{bc}	607 ^{bc}	619 ^{ab}	599 ^c	629 ^a	605 ^c	5.7	<0.001	0.34	0.07	0.33	0.26	<0.001
Blood														
Kg	14.65	14.64	14.18	14.75	16.28	13.74	14.21	0.809	0.30					
g/kg of EBW	27.0	25.7	26.8	24.1	27.1	24.9	26.4	1.49	0.62					
Hide ²														
Kg	53.18	52.42	50.54	56.12	54.43	50.23	51.64	1.935	0.26					
g/kg of EBW	97.9	91.7	95.7	91.1	90.3	91.2	95.6	2.16	0.16					

^{a-d} Least squares means within a row with differing superscripts differ ($P \leq 0.05$)

¹ Carcass weight does not include kidney, pelvic heart fat

² Muzzle, ears and tail included with hide weight

Table 4.4 Continued Empty body and tissue yields of steers fed for 0 – 28, or 28 – 56 days and given maintenance (M) or ad libitum (A) intake and control (C) or zilpaterol hydrochloride (Z) treatment.

Item	0	28 A	28 M	56 AZ	56 AC	56 MZ	56 MC	SEM	P-value	0 v. 28	0 v. 56	28 v. 56	M v. A	C v. Z
<i>N</i>	8	8	8	8	8	8	8							
Empty BW, kg	543 ^b	573 ^{ab}	529 ^b	615 ^a	602 ^a	552 ^b	540 ^b	15.89	<0.01	0.74	0.10	0.09	<0.001	0.36
Non-carcass bone excluding head ³														
Kg	10.77 ^c	11.37 ^{abc}	10.91 ^c	11.74 ^{ab}	11.72 ^{ab}	11.08 ^{bc}	11.84 ^a	0.265	0.02	0.27	<0.01	0.05	0.13	0.17
g/kg of EBW	19.8 ^{bc}	19.9 ^{bc}	20.7 ^{ab}	19.1 ^c	19.5 ^{bc}	20.2 ^{bc}	21.9 ^a	0.46	<0.01	0.43	0.51	0.79	<0.001	0.02
Head ⁴														
Kg	12.40	12.83	12.45	13.36	12.98	13.12	12.82	0.224	0.06	0.45	0.02	0.05	0.16	0.10
g/kg of EBW	22.8 ^{ab}	22.5 ^{ab}	23.6 ^a	21.8 ^b	21.6 ^b	23.9 ^a	23.8 ^a	0.59	<0.01	0.81	0.92	0.61	<0.001	0.79
Trim from Harvest ⁵														
Kg	13.56 ^{bc}	14.38 ^{abc}	13.46 ^{bc}	15.28 ^{ab}	15.61 ^a	12.79 ^c	12.90 ^c	0.710	0.02	0.69	0.47	0.72	<0.001	0.74
g/kg of EBW	25.0	25.1	23.3	25.0	25.9	23.2	24.0	1.00	0.54					
Internal Cavity ⁶														
Kg	85.17 ^{bc}	91.67 ^{ab}	81.10 ^c	91.91 ^{ab}	96.78 ^a	77.56 ^c	79.47 ^c	4.834	<0.001	0.78	0.74	0.99	<0.001	0.24
g/kg of EBW	156.6 ^{ab}	159.6 ^a	153.3 ^{ab}	149.6 ^b	156.9 ^a	140.1 ^c	147.2 ^{bc}	5.68	<0.001	0.96	0.06	0.02	<0.001	<0.01
Total Non-Carcass														
Kg	189.73 ^{bcd}	197.32 ^{abc}	182.64 ^{cd}	203.16 ^{ab}	207.8 ^a	178.52 ^d	182.88 ^{cd}	6.470	<0.001	0.97	0.61	0.53	<0.001	0.32
g/kg of EBW	349.1 ^a	344.4 ^{ab}	345.4 ^{ab}	330.5 ^{cd}	345.6 ^{ab}	323.4 ^d	338.8 ^{bc}	4.34	<0.001	0.26	<0.001	<0.001	0.09	<0.001
g/kg of BW	323.7 ^a	320.8 ^a	320.5 ^a	305.6 ^{cd}	316.1 ^{ab}	300.8 ^d	310.0 ^{bc}	3.90	<0.001	0.41	<0.001	<0.001	0.13	<0.01

^{a-d} Least squares means within a row with differing superscripts differ ($P \leq 0.05$)

³ Includes front and hind hooves, metacarpals, metatarsals and oxtail

⁴ Skinned head; tongue and tongue trim removed prior to weighing

⁵ Includes cod fat, fat trim from carcass, spinal cord and penis

⁶ Includes all internal viscera and offal as well as tongue and tongue trim and kidney pelvic heart fat weights

Table 4.5. Internal cavity yields of steers fed for 0 – 28, or 28 – 56 days and given maintenance (M) or ad libitum (A) intake and control (C) or zilpaterol hydrochloride (Z) treatment.

Item	0	28 A	28 M	56 AZ	56 AC	56 MZ	56 MC	SEM	P-value	0 v. 28	0 v. 56	28 v. 56	M v. A	C v. Z
<i>n</i>	8	8	8	8	8	8	8							
Internal Viscera and Offal Yields														
Tongue and Tongue Trim														
kg	5.51	5.27	5.12	5.66	5.43	5.29	5.04	0.175	0.12					
g/kg of EBW	10.2	9.2	9.7	9.2	9.1	9.6	9.4	0.42	0.28					
Bladder														
kg	0.74	0.46	0.58	0.67	0.77	0.55	0.80	0.171	0.59					
g/kg of EBW	1.4	0.8	1.1	1.1	1.3	1.0	1.5	0.29	0.55					
Thymus														
kg	1.69	1.83	1.94	2.16	2.36	2.01	1.83	0.186	0.24					
g/kg of EBW	3.1	3.2	3.6	3.5	3.9	3.7	3.4	0.29	0.56					
Trachea														
kg	0.96	0.98	1.00	1.00	0.99	1.02	0.95	0.996	0.86					
g/kg of EBW	1.8 ^{ab}	1.7 ^{ab}	1.9 ^a	1.6 ^b	1.7 ^b	1.9 ^a	1.8 ^{ab}	0.09	0.05	0.92	0.84	0.91	0.67	0.16
Heart														
kg	2.42	2.50	2.45	2.53	2.46	2.45	2.50	0.080	0.97					
g/kg of EBW	4.5 ^{ab}	4.4 ^{ab}	4.6 ^a	4.1 ^b	4.1 ^b	4.5 ^{ab}	4.6 ^a	0.17	0.03	0.79	0.39	0.14	<0.01	0.55
Lungs														
kg	2.62	2.68	2.78	2.88	2.86	2.79	2.91	0.170	0.70					
g/kg of EBW	4.8	4.7	5.3	4.7	4.8	5.1	5.4	0.28	0.17					
Pluck Trim														
kg	2.63	2.38	2.25	2.40	2.76	2.23	2.18	0.334	0.48					
g/kg of EBW	4.8	4.1	4.2	3.9	4.6	4.0	4.0	0.52	0.50					
Esophagus														
kg	0.36	0.37	0.34	0.41	0.36	0.38	0.39	0.178	0.09	0.91	0.16	0.05	0.43	0.21
g/kg of EBW	0.7	0.6	0.6	0.7	0.6	0.7	0.7	0.03	0.09	0.69	0.60	0.22	0.06	0.47
Kidneys														
kg	1.06 ^{ab}	1.11 ^a	1.06 ^{ab}	1.06 ^{ab}	1.15 ^{ab}	0.90 ^c	0.96 ^{bc}	0.050	0.02	0.64	0.52	0.13	<0.01	0.12
g/kg of EBW	1.9 ^{ab}	2.0 ^{ab}	2.0 ^a	1.7 ^{cd}	1.9 ^{abc}	1.6 ^d	1.8 ^{bcd}	0.07	<0.01	0.73	0.02	<0.01	0.35	0.02
Kidney-Pelvic-Heart Fat														
kg	7.80 ^b	7.92 ^b	6.94 ^b	8.29 ^b	10.17 ^a	7.15 ^b	7.28 ^b	0.895	0.01	0.64	0.55	0.15	<0.01	0.08
g/kg of EBW	14.34	13.80	13.09	13.56	16.92	12.88	13.49	1.389	0.11					

^{a-e}Least squares means within a row with differing superscripts differ ($P \leq 0.05$)

Table 4.5 Continued. Internal cavity yields of steers fed for 0 – 28, or 28 – 56 days and given maintenance (M) or ad libitum (A) intake and control (C) or zilpaterol hydrochloride (Z) treatment.

Item	0	28 A	28 M	56 AZ	56 AC	56 MZ	56 MC	SEM	P-value	0 v. 28	0 v. 56	28 v. 56	M v. A	C v. Z
<i>n</i>	8	8	8	8	8	8	8							
Internal Viscera and Offal Yields														
Stomachs ¹														
kg	19.18 ^{bcd}	22.87 ^a	19.42 ^{bcd}	21.02 ^{abc}	21.35 ^{ab}	16.98 ^d	18.72 ^{cd}	0.989	<0.01	0.12	0.75	0.06	<0.001	0.23
g/kg of EBW	35.3 ^b	39.8 ^a	36.7 ^{ab}	34.2 ^b	35.4 ^b	30.7 ^c	34.8 ^b	1.23	<0.001	0.05	0.28	<0.001	0.02	0.03
Intestines ²														
kg	21.26 ^{ab}	22.56 ^a	20.29 ^{ab}	20.83 ^{ab}	22.99 ^a	18.55 ^b	18.50 ^b	1.145	<0.01	0.90	0.39	0.19	<0.001	0.25
g/kg of EBW	39.1 ^a	39.3 ^a	38.4 ^a	33.8 ^b	38.2 ^a	33.5 ^b	34.2 ^b	1.38	<0.01	0.87	<0.01	<0.01	0.09	0.04
Total GIT ³														
kg	40.44 ^{abc}	45.43 ^a	39.71 ^{bcd}	41.86 ^{ab}	44.34 ^{ab}	35.53 ^d	37.22 ^{cd}	1.998	<0.001	0.35	0.72	0.07	<0.001	0.17
g/kg of EBW	74.4 ^b	79.1 ^a	75.1 ^{ab}	68.0 ^{cd}	73.7 ^b	64.2 ^d	69.0 ^c	2.04	<0.01	0.22	<0.01	<0.01	0.01	0.01
Spleen														
kg	0.98 ^c	1.24 ^a	1.09 ^{abc}	1.27 ^a	1.20 ^{ab}	1.03 ^{bc}	1.26 ^a	0.071	0.02	0.04	<0.01	0.71	0.08	0.27
g/kg of EBW	1.8	2.2	2.1	2.1	2.0	1.9	2.3	0.14	0.17					
Liver														
kg	6.62 ^a	6.65 ^a	5.12 ^b	6.56 ^a	6.86 ^a	4.72 ^b	5.25 ^b	0.218	<0.001	0.02	<0.01	0.85	<0.001	0.04
g/kg of EBW	12.2 ^a	11.6 ^{ab}	9.7 ^d	10.7 ^c	11.4 ^b	8.6 ^e	9.7 ^d	0.26	<0.001	<0.001	<0.001	<0.01	<0.001	<0.001
Gall Bladder														
kg	0.29	0.33	0.26	0.39	0.34	0.28	0.27	0.429	0.40					
g/kg of EBW	0.5	0.6	0.5	0.6	0.6	0.5	0.5	0.07	0.89					
Pancreas														
kg	0.37 ^c	0.57 ^a	0.48 ^{abc}	0.45 ^{bc}	0.57 ^a	0.49 ^{ab}	0.49 ^{ab}	0.512	0.01	<0.01	<0.01	0.62	0.14	0.10
g/kg of EBW	0.7 ^c	1.0 ^a	0.9 ^{abc}	0.7 ^{bc}	1.0 ^a	0.9 ^{abc}	0.9 ^{ab}	0.59	0.04	0.01	0.03	0.30	0.92	0.06

^{a-e} Least squares means within a row with differing superscripts differ ($P \leq 0.05$)

¹ Stomachs includes reticulorumen, omasum and abomasum

² Intestines includes small intestine, large intestine and cecum

³ Total gastrointestinal tract (GIT) includes reticulorumen, omasum, abomasum, small intestine, large intestine and cecum

Table 4.5 Continued. Internal cavity yields of steers fed for 0 – 28, or 28 – 56 days and given maintenance (M) or ad libitum (A) intake and control (C) or zilpaterol hydrochloride (Z) treatment.

Item	0	28 A	28 M	56 AZ	56 AC	56 MZ	56 MC	SEM	P-value	0 v. 28	0 v. 56	28 v. 56	M v. A	C v. Z
<i>n</i>	8	8	8	8	8	8	8							
Internal Viscera and Offal Yields														
TST ⁴ , no omental or kidney pelvic heart fat														
kg	48.65 ^{abc}	54.20 ^a	46.51 ^{bcd}	50.47 ^{ab}	53.30 ^a	42.05 ^d	44.49 ^{cd}	1.999	<0.001	0.49	0.62	0.10	<0.001	0.12
g/kg of EBW	89.5 ^{ab}	94.4 ^a	88.0 ^{bc}	82.0 ^d	88.6 ^b	76.0 ^e	82.5 ^{cd}	2.16	<0.001	0.48	<0.01	<0.001	<0.001	<0.01
Omental Fat														
kg	9.38 ^{ab}	10.01 ^{ab}	8.34 ^b	11.45 ^a	11.77 ^a	8.49 ^b	8.19 ^b	1.392	0.01	0.85	0.54	0.28	<0.001	0.99
g/kg of EBW	17.2	17.3	15.8	18.7	19.6	15.2	15.1	2.18	0.10	0.67	0.96	0.58	<0.01	0.76
Omental and Kindney-Pelvic-Heart Fat														
kg	17.18 ^{bc}	17.93 ^{bc}	15.27 ^c	19.75 ^{ab}	21.94 ^a	15.64 ^c	15.47 ^c	2.285	<0.01	0.76	0.53	0.20	<0.001	0.40
g/kg of EBW	31.6	31.1	28.9	32.2	36.5	28.1	28.6	3.55	0.07	0.58	0.94	0.47	<0.001	0.22
TST ⁴ , including omental fat														
kg	58.03 ^{bc}	64.21 ^{ab}	54.85 ^{cd}	61.92 ^{ab}	65.07 ^a	50.54 ^d	52.68 ^{cd}	3.256	<0.001	0.63	0.86	0.35	<0.001	0.23
g/kg of EBW	106.7 ^{abc}	111.7 ^a	103.7 ^{bcd}	100.7 ^{cd}	108.2 ^{ab}	91.2 ^e	97.6 ^{de}	3.90	<0.001	0.75	0.02	<0.001	<0.001	0.01

^{a-e} Least squares means within a row with differing superscripts differ ($P \leq 0.05$)

⁴ Total splanchnic tissue mass (TST) includes reticulorumen, omasum, abomasum, small intestine, large intestine and cecum, spleen, liver, gall bladder and pancreas

Table 4.6. Hot Carcass Weight and Carcass Characteristics of beef steers fed for 0 – 28, or 28 – 56 days and given maintenance (M) or ad libitum (A) intake and control (C) or zilpaterol hydrochloride (Z) treatment.

Item	0	28 A	28 M	56 AZ	56 AC	56 MZ	56 MC	SEM	P-value	0 v. 28	0 v. 56	28 v. 56	M v. A	C v. Z
HCW, kg ¹	361 ^c	383 ^{bc}	353 ^c	420 ^a	404 ^{ab}	380 ^{bc}	364 ^c	11.4	<0.01	0.67	0.03	0.03	<0.01	0.10
Dressed carcass yield ² , %	61.7 ^c	62.4 ^{bc}	62.0 ^{bc}	63.2 ^{ab}	61.4 ^c	64.1 ^a	61.7 ^c	0.52	<0.01	0.41	0.08	0.27	0.47	<0.01
Adj. 12 th rib fat thickness, cm	1.63 ^{cd}	2.15 ^{bc}	1.68 ^{cd}	2.52 ^{ab}	2.72 ^a	1.92 ^{cd}	1.52 ^d	0.19	<0.01	0.22	0.01	0.12	<0.01	0.61
LM area, cm ²	85.81	89.03	85.97	93.55	89.11	87.82	88.39	0.45	0.53					
KPH, %	2.16	2.06	1.96	1.98	2.53	1.87	2.00	0.21	0.09	0.45	0.73	0.52	0.05	0.02
USDA Calculated YG ³	3.3 ^{cd}	3.8 ^{bc}	3.3 ^{cd}	4.3 ^{ab}	4.7 ^a	3.6 ^{bcd}	3.1 ^d	0.27	<0.01	0.46	0.05	0.12	<0.01	0.81
Marbling Score ⁴	420	441	419	453	446	445	398	49.6	0.38					
Skeletal Maturity ⁵	138	145	140	145	146	145	160	10.4	0.79					
Color Score ⁶	5	5.4	5.1	5.0	4.9	5.1	5.4	0.5	0.90					
Canadian Ribeye length ⁷	1.38	1.75	2.00	2.13	1.88	1.75	1.75	0.25	0.49					
Canadian Ribeye width ⁷	2.13	2.25	2.00	1.88	2.00	1.50	1.75	0.24	0.43					
Canadian Muscle Score ⁷	1.50	2.00	2.00	2.13	1.88	1.38	1.75	0.30	0.53					
Canadian Fat Class ⁷	5.88 ^{cd}	8.13 ^{abc}	6.76 ^{cd}	8.63 ^{ab}	9.13 ^a	7.63 ^{bcd}	6.63 ^d	0.58	<0.01	0.36	0.05	0.20	<0.01	0.62
Canadian Lean Yield, % ⁸	53.9 ^{ab}	52.6 ^{abc}	54.6 ^a	51.9 ^{bc}	51.1 ^c	52.6 ^{abc}	54.6 ^a	0.92	0.03	0.81	0.17	0.16	<0.01	0.47

¹ Hot carcass weight includes kidney, pelvic and heart fat

² BW used was unshrunk as steers were off feed and water for 9 h

³ USDA calculated yield grade (YG) = $2.5 + (2.5 \times \text{FT}) + (0.2 \times \text{KPH}) + (0.0038 \times \text{HCW}) - (0.32 \times \text{REA})$, where FT = adjusted 12th rib fat depth in cm, KPH = percentage of kidney, pelvic, and heart fat, HCW = hot carcass weight in kg, and REA = longissimus muscle area in cm²

⁴ 100 = Practically Devoid⁰⁰, 300 = Slight⁰⁰, 500 = Modest⁰⁰, 700 = Slightly Abundant⁰⁰, and 900 = Abundant⁰⁰

⁵ 100 = A⁰⁰ and 500 = E¹⁰⁰

⁶ 1 = light pink, 2 = pink, 3 = dark pink, 4 = light cherry red, 5 = cherry red, 6 = dark red, 7 = very dark red (1/3 dark cutter), 8 = maroon (2/3 dark cutter) and 9 = dark maroon (full dark cutter)

⁷ Canadian ribeye length and width, muscle score and fat class calculated using Yield Ruler developed by Lacombe Research Station (CGBA, 2001)

⁸ Canadian calculated lean yield % = $63.65 + (1.05 \times \text{muscle score}) - (0.76 \times \text{fat class})$

CHAPTER 5

ESTIMATING CARCASS COMPOSITION IN STEERS FED DIFFERENT ENERGY LEVELS AND ZILPATEROL HYDROCHLORIDE

5.1 Abstract

A trial was conducted to examine the linear relationship between composition of the 9-10-11 rib and the carcass. Steers were commercially harvested ($n = 56$, Final BW = $612 \text{ kg} \pm 52.2$) and graded following a 24h chill. Carcass sides were fabricated 48h post-harvest; samples of 9-10-11 rib primals were dissected into lean, fat, and bone with lean and fat ground and sampled for analysis; all long bones were sliced and sampled for analysis. The relationship between individual 9-10-11 rib and carcass parameters were evaluated via correlation and simple linear regression. Multiple-linear regression was used to evaluate the predictive ability of 9-10-11 rib composition, HCW, LM area (LMA), 12th rib s.c. fat depth (FT), percentage kidney-pelvic-heart fat, marbling score (QG), Canadian ribeye length and width (REL and REW, respectively), fat class and muscle score (FC and MS, respectively), treatment (TRT; control C=0, zilpaterol Z=1), and diet (maintenance M=0, *ad libitum* A=1) to estimate edible carcass chemical composition. Separable fat of the carcass (SFC; $r = 0.89$; $P < 0.01$) and separable lean of the carcass (SLC; $r = 0.83$; $P < 0.01$) were strongly correlated to 9-10-11 rib separable fat (SFR) and to 9-10-11 rib separable lean (SLR), respectively and are estimated using the equations: $\{(SFC = 4.375 + 0.471 * SFR); P < 0.01, \text{Adj. } R^2=0.78, \text{RMSE } 1.462\}$

and $\{(SLC = 42.962 + 0.428 * SLR); P < 0.01, \text{Adj. } R^2=0.68, \text{RMSE } 1.446\}$. Ether extract of the edible carcass (EEC_e) exhibited a moderate correlation ($r = 0.67, P < 0.01$) to ether extract of the edible 9-10-11 rib (EER_e) and was estimated as $\{(EEC_e = 13.351 + 0.439 * EER_e); P < 0.001, \text{Adj. } R^2=0.44, \text{RMSE } 3.110\}$. Edible carcass moisture (MC_e) was correlated ($r = 0.48, P < 0.01$) to 9-10-11 edible rib moisture (MR_e) and estimated from the equation $\{(CM_e = 37.570 + 0.308 * RM_e); P < 0.001, \text{Adj. } R^2=0.25, \text{RMSE } 3.073\}$. Protein of the edible carcass (PC_e) was weakly correlated ($r = 0.30, P < 0.01$) to the 9-10-11 edible rib protein (PR_e) $\{(PC_e = 13.527 + 0.208X); P = 0.02, \text{Adj. } R^2= 0.07, \text{RMSE } = 1.418\}$. An equation to predict EEC_e was developed $\{P < 0.001; \text{Adj. } R^2=0.75, \text{RMSE } 2.068; (EEC_e = 51.476 - (1.440 * TRT) + (0.009 * QG) + (0.616 * FC) - (0.581 * SLR))\}$. Carcass protein (PC_e) was estimated $\{P < 0.01, \text{Adj. } R^2=0.38, \text{RMSE } 1.182; (PC_e = 9.997 + (0.694 * DOF) - (1.389 * TRT) - (1.117 * FT) - (0.988 * KPH) + (0.448 * FC) + (0.131 * SLR))\}$. Results indicate strong correlations ($r > 0.80, P < 0.01$) between separable lean and fat of the 9-10-11 rib to separable lean and fat of the carcass and models were developed that moderately predict ($\text{Adj. } R^2 > 0.50, P < 0.01$) carcass chemical composition by 9-10-11 rib physical and chemical composition, using carcass grading traits.

5.2 Introduction

Predicting physical and chemical carcass composition has been extensively researched by animal scientists for almost a century largely due to the labor, cost and waste involved with fabrication, grinding and sampling of whole carcass tissue. Thus, multiple methods have sought to eliminate the need for whole carcass dissection and analysis. Specific gravity measurements (Kraybill et al., 1952; Orme et al., 1958; Cole et

al., 1960) and 9-10-11 rib dissection (Hankins and Howe, 1946; Shackelford et al., 1995; Johnson et al., 1996) have been utilized as composition estimators in lieu of whole carcass dissection, grinding and chemical analysis. The 9-10-11 rib dissection has been used to predict physical and chemical carcass composition since its inception (Hankins and Howe, 1946) with more recent research using 9-10-11 rib composition as well as carcass characteristics to predict beef carcass chemical and physical composition (Crouse and Dikeman, 1977). Other research has focused on relationships between carcass attributes of loin eye area, HCW, carcass length and width and separable carcass lean (Cole et al., 1960 and 1962) whereas Powell and Huffman (1973) utilized carcass grading factors to determine carcass physical composition. Furthermore, both the USDA and Canadian grading system seek to not only predict customer eating satisfaction with quality grades but also saleable red meat yield from a calculated yield grade (USDA, 1997; CBGA, 2001).

Limited North American feeder cattle supplies have fostered an increase in carcass weights to meet beef demand. Additionally, new growth technologies have focused on improving lean muscle deposition and carcass cutability, which has prompted producers to utilize higher dose combination estradiol-trenbolone acetate implants and β -adrenergic agonists. To date, research has not focused on predicting carcass composition in heavier cattle (> 550 kg) with and without β -adrenergic agonist supplementation. Additionally, no studies have utilized both Canadian and USDA grading factors coupled with 9-10-11 rib physical and chemical composition to predict physical and chemical carcass composition. Therefore, the objective of this experiment was to examine the linear relationship between composition of the 9-10-11 rib, carcass grading factors and

the physical and chemical carcass composition in beef steers fed at maintenance or *ad libitum* level of intake with or without ZH supplementation.

5.3 Materials and Methods

All experimental procedures involving live animals were approved by the Animal Care and Use Committee at West Texas A&M University and adhered to the regulations in the Guide for the Care and Use of Agricultural Animals in Agricultural Research and Teaching (Federation of Animal Science Societies, 2010, Savoy, IL).

5.3.1 Live Cattle Procedures and Experimental Design

Live cattle procedures and experimental design are explained in detail in a previous manuscript (Walter et al., 2015. *Effect of dietary energy level and zilpaterol hydrochloride on the live performance, grading performance and harvest yields of beef steers*. Chapter II.). Briefly, single sired steers (n=60; initial BW = 574±36 kg) were purchased and transported to the research feedyard where they were held in two pens (n = 30 per pen) until further weighing and sorting. Cattle sort weights were recorded on d -5 and d -6 with 28 steers selected per block (Block 1 initial BW = 589±25 kg and Block 2 initial BW = 614±39 kg). Within each block, two steers with weights furthest from the mean (heaviest and lightest steer in Block 1 and two lightest steers in Block 2) were held in individual pens as alternates for the duration of the trial resulting in 28 head per Block. Steers were sorted into individual pens on d -4 with Block 2 starting trial 28 days after Block 1. Cattle were weighed off feed and water (9 h withdrawal) on d -1, 1, 27, 28, 55 and 56.

The trial was constructed as a multi-factorial treatment design with three harvest dates, two dietary energy intake levels and Z supplementation or control (C). Cattle were

blocked by implant status and randomized to pairs on the sort days (d -5 and d -6) by weight. Pairs were randomized to harvest day (0, 28 or 56 d) and dietary energy intake (maintenance or *ad libitum*) within harvest date. Within d 56 harvest date, one animal within each pair for both maintenance (M) and *ad libitum* (A) level of feeding was randomly assigned to zilpaterol hydrochloride supplementation (ZH; 90 mg/d; Merck Animal Health; Summit, NJ).

5.3.2 Harvest and Carcass Grading

All steers were harvested at Caviness Packing Company (Establishment 675; Hereford, TX). At harvest, mass of all harvest components (non-carcass and carcass) were recorded. Twenty-four hours post-harvest, a detailed carcass evaluation was conducted which included marbling score, lean and skeletal maturity, 12th rib s.c. fat depth (cm) and LM area (cm²). A final quality grade and calculated yield grade was determined (USDA, 1997). In addition, Canadian grading factors were evaluated for muscle score (matrix of maximum length and width) and grade fat (fat class) using the Canadian grading ruler to calculate the Canadian yield grade measurement (Canadian Beef Grading Agency, 2001). A further detail of carcass evaluation is included in Walter et al., 2015 (*Effect of dietary energy level and zilpaterol hydrochloride on the live performance, grading performance and harvest yields of beef steers*. Chapter II.).

5.3.3 Carcass Fabrication and Sampling

At 48 h post-harvest, left carcass sides were fabricated into primals and weighed to determine cold carcass side weight. Carcass primals were fabricated according to industry standards (IMPS, 2014) and weighed individually to ± 0.02 kg accuracy with all bone and fat trim weighed separately to determine cutability. Rib primals were weighed;

the 9-10-11 rib sections were removed according to Hankins and Howe (1946). Portions of 9-10-11 rib were kept separate from the remainder of the 112A rib subprimal, however both were fabricated to meet IMPS standards for the rib subprimal and weighed.

Subsequently, the 9-10-11 rib portion was further dissected into separable bone, lean and fat as well as the *longissimus dorsi* muscle. The muscle was weighed separately as well as with the lean trim to compute total separable lean for the 9-10-11 rib.

After all retail cuts were weighed, bone-in retail cuts (#130A and #124) were again dissected to remove bone. All remaining retail cuts and lean trim were weighed as separable lean, separable bone (inclusive of major tendons and ligamentum nuchae), and all the fat trim was weighed as separable fat. The soft tissue of the 9-10-11 rib was mixed by hand, ground once with a coarse plate (9.5 mm) through a meat grinder (548SS, Biro[®], Marblehead, OH), again mixed by hand and ground through a fine grind plate (3.2 mm). The fine grind mixture was sampled three times during the grind process for each animal; the three samples were thoroughly mixed together to form a homogenous sample and then divided into three separate samples (~100 g each). The retail cuts and lean trim were ground in a similar fashion as 9-10-11 rib but after the coarse grind, lean was put into a large stainless steel tub, the remaining 9-10-11 rib grind was added and the mixture was thoroughly mixed for 5 min before being ground through a fine grind plate (3.2 mm). Fat trim was ground, mixed and re-ground in an identical fashion to lean trim without the addition of 9-10-11 rib soft tissue. The same triplicate sampling and further sub-sampling procedures were used for separable lean and fat as was used with the 9-10-11 rib soft tissue samples. The triplicate, homogenous samples of each 9-10-11 rib, carcass

lean and carcass fat for individual animals were packaged separately in Whirl-Pak bags, vacuum sealed and frozen at -20 °C.

All long bones (tibia/fibula, femur, humerus, radius/ulna) were retained, packaged by individual animal, vacuum sealed and frozen (-20 °C) for further sampling. Long bones were then thawed for 12 h, weighed, trimmed of any remaining fat or lean and weighed after trimming. Trimmed bones were cut into 2.54 cm cross-sections, perpendicular to the long axis of the bone with a meat saw (116-F, Butcher Boy, Lasar Mfg. Las Angeles, CA). The saw was cleaned with a brush and saw dust collected after every animal, mixed and sub-sampled in three separate Whirl-Pak bags, vacuum sealed and frozen at -20 °C for further analysis.

5.3.4 Sample Analysis

All tissue samples were lypohphilized (Bulk Tray Dryer, Labconco Corp, Kansas City, MO) ground in a Waring blender and dried at 105°C for 9 h for further determination of DM. Samples were then analyzed by SDK Laboratories (Hutchinson, KS) for determination of ash (#942.05, AOAC, 2000), protein (#976.06, AOAC 2000) and crude fat (#Ba-3-38, AOCS, 2013).

5.3.5 Statistical Analysis

All regression analysis was performed using the REG procedure of SAS (SAS Institute, Inc. Cary, NC). Simple correlation and linear regression was calculated to quantify the relationship between the physical and chemical 9-10-11 rib composition and the physical and chemical dressed carcass composition. Multiple-linear regression using the best fit model as determined by stepwise, backward or forward regression was used to evaluate the predictive ability of 9-10-11 rib composition, HCW, LM area (LMA), 12th

rib subcutaneous fat depth (FT), percentage kidney-pelvic-heart fat, marbling score (QG), Canadian ribeye length and width measurements (REL and REW, respectively), Canadian fat class and muscle score (FC and MS, respectively), TRT (C=0, Z=1), and DIET (M=0, A=1) to estimate edible carcass chemical composition. Collinearity diagnostics using the TOL, VIF and COLLIN options of SAS was used to evaluate and prevent the use of multicollinearity if variable combinations resulted in a condition index > 10.

5.4 Results and Discussion

5.4.1 Physical and Chemical 9-10-11 Rib and Dressed Carcass Composition Relationship

Separable fat of the 9-10-11 rib cut (SFR) and the dressed carcass (SFC) averaged 33.45 and 20.14%, respectively (Table 1). Concomitantly, separable lean was greater in the dressed carcass (63.70%) than the 9-10-11 rib cut (48.45%). Separable bone was more closely associated the 9-10-11 rib cut and the dressed carcass (18.10 vs. 16.16%, respectively), however the range (13.84 to 25.87%) and variation (2.17 SD) was greater in the 9-10-11 rib cut than the dressed carcass (13.42 to 18.84%; 1.13 SD). The original research of Hankins and Howe (1946) reported the average SFR to be $25.37\% \pm 6.87$ and $23.77\% \pm 5.88$ for SFC. The SFR in our trial was 8.08% greater than the original research whereas SFC was slightly less than previously reported. The discrepancy could largely be due to the change in cattle population over the last 7 decades. Increased genetic selection for growth and muscling has likely led to increased saleable red meat yield. Separable lean of the 9-10-11 rib (SLR) and dressed carcass (SLC) in the original research was $52.82\% \pm 5.05$ and $58.27\% \pm 4.47$, respectively (Hankins and Howe, 1946).

Thus the dressed carcasses in the current study were leaner, resulting in increased separable lean and reduced separable fat when compared to the original work of Hankins and Howe (1946).

Ether extract of the edible portion of the 9-10-11 rib (EER_e) ($41.28\% \pm 6.35$) averaged 9.81% more than the ether extract of the edible portion of the dressed carcass (EEC_e) ($31.47\% \pm 4.16$). Protein and moisture of the edible portion of the 9-10-11 rib (PR_e and MR_e) (12.96 and 44.95%, respectively) was 3.26 and 6.47% less than the protein and water content of the edible portion of the dressed carcass (PC_e and MC_e) (16.22 and 51.42%, respectively). Ash of the edible portion did not differ between the 9-10-11 rib (0.62%) and the edible portion of the dressed carcass (0.44%). Parallel to physical composition, chemical composition of the current study differed from the original research. Ether extract of the edible portions of the 9-10-11 rib and dressed carcass averaged $32.75\% \pm 7.70$ and $27.73\% \pm 6.23$, respectively, a difference of 5.03% in the study by Hankins and Howe (1946). In contrast, protein and water differed slightly between 9-10-11 rib and dressed carcass in the study by Hankins and Howe (1946) and were thus similar to the results of the dressed carcass in the current study. The protein and water content of the 9-10-11 rib in the current study was reduced relative to the original research whereas ether extract was increased, suggestive of differing composition of gain likely due to genetic selection or extended days on feed. Similar to the current trial, EER_e was greater than EEC_e ($40.7\% \pm 6.0SD$ vs. $35.2\% \pm 4.9$) whereas MR_e was reduced ($45.2\% \pm 4.6SD$ vs. $49.6\% \pm 3.7$) vs. the dressed carcass in the study by Crouse and Dikeman (1974). Additionally, Nour and Thonney (1994) reported greater

levels of EER_e vs. EEC_e (45.2 vs. 37.6%) and reduced MR_e vs. MC_e (41.5 vs. 47.3%) in Angus steers.

Correlations and simple linear relationships were examined between the physical characteristics of the 9-10-11 rib and chemical and physical carcass composition (Table 2). Separable fat and lean of the 9-10-11 rib were strongly correlated ($r = 0.89$ and $r = 0.83$, respectively) and used to predict SFC $\{(Y = 4.375 + 0.471X); P < 0.001, \text{Adj. } R^2 = 0.78, \text{RMSE} = 1.462\}$ and SLC $\{(Y = 42.962 + 0.428X); P < 0.001, \text{Adj. } R^2 = 0.68, \text{RMSE} = 1.446\}$. Separable bone of the 9-10-11 rib was moderately correlated ($r = 0.59$) and able to predict SBC $\{(Y = 10.629 + 0.306X); P < 0.001, \text{Adj. } R^2 = 0.33, \text{RMSE} = 0.916\}$. Additionally, SFR was strongly correlated ($r = 0.83$) and able to predict ether extract of the dressed carcass $\{(Y = 14.463 + 0.489X); P < 0.001, \text{Adj. } R^2 = 0.68, \text{RMSE} = 1.980\}$ and EEC_e $\{(Y = 12.020 + 0.581X); P < 0.001, \text{Adj. } R^2 = 0.68, \text{RMSE} = 2.361\}$. Separable lean of the 9-10-11 rib was strongly correlated ($r = 0.72$ and $r = 0.73$) to the MC and the MC_e, respectively whereas SLR exhibited only moderate correlation ($r = 0.41$ and $r = 0.42$) to protein of the dressed carcass and the PC_e, respectively. Moisture of the edible portion of the dressed carcass was predicted by SLR $\{(Y = 25.761 + 0.530X); P < 0.001; \text{Adj. } R^2 = 0.53, \text{RMSE} = 2.439\}$ and PC_e was poorly predicted by SLR $\{(Y = 10.122 + 0.126X); P < 0.01; \text{Adj. } R^2 = 0.16, \text{RMSE} = 1.351\}$. Previous literature in steers, (Hankins and Howe, 1946) reported similar correlations between SFR and SFC ($r = 0.93$) and between SLR and SLC ($r = 0.90$) vs. correlations of $r = 0.89$ and $r = 0.83$, respectively for this study. Additionally, Hankins and Howe (1946) reported a strong correlation ($r = 0.92$) between SFR and the EEC_e as well as between SLR and PC_e ($r = 0.82$) relative to correlations of ($r = 0.83$ and $r = 0.42$) in the current study. Discrepancy

between SLR and PC_e between the original and current study may indicate changes in muscle composition because SLR was strongly correlated to MC_e ($r = 0.73$) and because moisture composes 73% of skeletal muscle, moisture maybe a stronger chemical indicator of separable lean vs. actual protein.

Correlations and simple prediction equations were also developed to predict chemical dressed carcass composition from chemical characteristics of the 9-10-11 rib (Table 3). Ether extract of the 9-10-11 rib whole (EER) and edible only portions (EER_e) exhibited positive moderate correlations ($r = 0.69$ and $r = 0.67$) to EEC and EEC_e , whereas EER and EER_e displayed negative moderate correlations to MC and MC_e ($r = -0.57$ and $r = -0.63$). Ether extract of the edible portion of the 9-10-11 rib predicted EEC_e $\{(Y = 13.351 + 0.439X); P < 0.001; \text{Adj. } R^2 = 0.44, \text{RMSE} = 3.111\}$ as well as MC_e $\{(Y = 65.746 - 0.347X); P < 0.001; \text{Adj. } R^2 = 0.38, \text{RMSE} = 2.791\}$. Protein of the dressed carcass was weakly correlated to EER in a negative fashion ($r = -0.34$) whereas PC_e was also weakly correlated to EER_e in a negative fashion ($r = -0.32$). Weak, positive correlations were exhibited between PR and PC ($r = 0.33$) and between PR_e and PC_e ($r = 0.30$). Moisture of the 9-10-11 rib was moderately correlated ($r = 0.48$) to MC and MR was used to predict MC $\{(Y = 33.992 + 0.297X); P < 0.001; \text{Adj. } R^2 = 0.22, \text{RMSE} = 2.567\}$. Additionally, MR_e was correlated ($r = 0.51$) to MC_e with MR_e predicted MC_e with $\{(Y = 37.570 + 0.308X); P < 0.001; \text{Adj. } R^2 = 0.25, \text{RMSE} = 3.073\}$. Ash of the 9-10-11 rib (AR) was moderately correlated ($r = 0.65$) to ash of the dressed carcass (AC), whereas ash of the edible portion of the 9-10-11 rib (AR_e) was poorly correlated ($r = 0.12$) to ash of the edible portion of the edible carcass (AC_e). Ash of the rib predicted AC $\{(Y = 3.842 + 0.353X); P < 0.01; \text{Adj. } R^2 = 0.42, \text{RMSE} = 0.382\}$ but AR_e was a poor

predictor ($P = 0.40$) of AC_e . Hankins and Howe (1946) reported stronger correlations between all chemical edible rib composition and chemical composition of the edible carcass including EER_e and EEC_e than the current study ($r = 0.91$ vs. $r = 0.69$, respectively), PR_e and PC_e ($r = 0.83$ vs. $r = 0.30$, respectively), EER_e and MC_e ($r = -0.99$ vs. $r = -0.63$, respectively) and MR_e and PC_e ($r = 0.91$ vs. $r = 0.24$). Additionally, Crouse and Dikeman (1974) reported strong correlations between MR_e and MC_e ($r = 0.88$), EER_e and EEC_e ($r = 0.94$) and PR_e and PC_e ($r = 0.95$), similar to the work by Hankins and Howe (1946). Nour and Thonney (1994) reported strong correlations between MR_e and MC_e ($r = 0.81$) and EER_e and EEC_e ($r = 0.85$) but reported a moderate correlation between PR_e and PC_e ($r = 0.71$). Alternatively, Vance et al. (1971) reported moderate correlations between chemical composition of the rib and carcass, similar to the results of the current study, MR_e and MC_e ($r = 0.74$), EER_e and EEC_e ($r = 0.71$) and PR_e and PC_e ($r = 0.51$). Discrepancies may be due to the diversity of the populations of cattle as noted by Crouse and Dikeman (1974) as well as total number of animals in the sample population. Hankins and Howe (1946) utilized 84 steers and 36 heifers from three study sites and reported cold carcass weights ranging from 129 to 389 kg whereas Crouse and Dikeman (1974) utilized 27 steers slaughtered on three different time points resulting in a HCW of 308.4 ± 35.1 kg. Furthermore, Nour and Thonney (1994) utilized both Angus ($n = 74$) and Holstein steers ($n = 69$) harvested on 5 time points with a BW range of 363 – 544 kg for Angus and 454 – 635 kg for Holstein whereas Vance et al. (1971) used a total of 16 animals with HCW ranging from 162 to 334 kg. The current study consisted of 56 steers of single sire origin, harvested across 84 d with a dressed HCW of 373.0 ± 32.1 kg and therefore may have resulted in reduced variation as evidenced by the smaller standard

deviations for physical and chemical composition of the rib and carcass vs. Hankins and Howe (1946) and Crouse and Dikeman (1974). The reduced variation across the study likely resulted in the lower coefficient of correlations as previously noted to be a cause of discrepancy by Crouse and Dikeman (1974).

5.4.2 Prediction of the Physical and Chemical Carcass Composition

Multiple studies have estimated physical and chemical carcass composition on diverse populations of cattle (Crouse and Dikeman, 1974; Guiroy et al., 2001; Marcondes et al., 2012) but to date, little research has focused on predicting physical and chemical carcass composition in cattle fed a β -adrenergic agonist and using dietary treatments. The current study investigated the ability of DOF, ZH treatment, DIET, USDA and Canadian carcass grading factors and physical and chemical rib composition to predict physical carcass composition (Table 4). Physical composition of the SFC was strongly predicted $\{(SFC = 45.261 + (0.961*FT) - (0.228*SBR) - (0.472*SLR)); P < 0.001, RMSE = 1.360 \text{ Adj. } R^2 = 0.82\}$ whereas SLC $\{(SLC = 44.043 - (0.868*FT) + (0.055*LMA) + (0.336*SLR)); P < 0.001, RMSE = 1.374 \text{ Adj. } R^2 = 0.69\}$ and SBC $\{(SBC = 12.612 - (0.011*HCW) + (0.494*SBR) + (0.083*SLR) - (0.730*AR)); P < 0.001, RMSE = 0.690 \text{ Adj. } R^2 = 0.66\}$ were moderately predicted by carcass grading factors and rib physical composition.

Carcass chemical composition was also predicted by diet, treatment, DOF, U.S. and Canadian carcass grading factors and rib physical and chemical composition. With respect to the edible portion of the carcass, EEC_e and MC_e were moderately predicted with $EEC_e = ((51.476 - (1.440*TRT) + (0.009*QG) + (0.616*FC) - (0.581*SLR)) (P < 0.001; RMSE = 2.068, \text{ Adj. } R^2 = 0.75)$ and $MC_e = 62.560 + (1.999*TRT) - (1.039*FC) -$

$(0.384 \cdot \text{SBR}) + (0.169 \cdot \text{SLR}) - (0.166 \cdot \text{EER}_e) + (2.691 \cdot \text{AR}_e)$ ($P < 0.001$; RMSE = 2.140, Adj. $R^2 = 0.64$). The protein fraction of the edible carcass was poorly estimated $\text{PC}_e = 9.997 + (0.694 \cdot \text{DOF}) - (1.389 \cdot \text{TRT}) - (1.117 \cdot \text{FT}) - (0.988 \cdot \text{KPH}) + (0.448 \cdot \text{FC}) + (0.131 \cdot \text{SLR})$ ($P < 0.001$; RMSE = 1.182, Adj. $R^2 = 0.38$). Crouse and Dikeman (1974) using carcass traits of (QG, FT, KPH, HCW and muscling score) were able to account for 90, 89 and 87% of the variation in MC_e , EEC_e and PC_e , respectively. Marcondes et al. (2012) was able to predict 83% of the variation in EEC_e utilizing EER_e and empty body weight. Therefore results of the current trial are weakly to moderately predicted vs. stronger predictions made by previous researchers (Crouse and Dikeman, 1974). This is likely a result of increased uniformity and thus reduced variation in the current study.

Results from the current study indicate strong correlations ($r > 0.80$, $P < 0.01$) between separable lean and fat of the 9-10-11 rib to separable lean and fat of the carcass, however chemical carcass composition was moderately correlated to 9-10-11 rib chemical composition ($r < 0.50$, $P < 0.01$). Models were developed utilizing Canadian and USDA grading factors as well as 9-10-11 rib physical and chemical composition that strongly (Adj. $R^2 > 0.80$, $P < 0.01$) predict SFR and moderately predict (Adj. $R^2 > 0.50$, $P < 0.01$) carcass chemical fat and moisture as well as SLC and SBC. Carcass protein and ash was poorly predicted (Adj. $R^2 < 0.50$, $P < 0.01$) by 9-10-11 rib physical and chemical composition and carcass grading traits. Further research is warranted to examine the improved predictive ability of physical and chemical carcass composition utilizing live cattle performance factors as well as to examine the ability of the 9-10-11 rib and grading factors to predict empty body physical and chemical composition.

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Table 5.1. Ranges, means and standard error of the composition and chemical analysis of the 9-10-11 rib cut and dressed beef carcasses.

Item	Min	Max	Mean	SD	SE
9-10-11 Rib Cut, %					
Separable Fat	21.45	53.68	33.45	5.90	0.792
Separable Lean	30.52	59.02	48.45	4.88	0.653
LM (eye muscle)	15.25	33.90	19.76	2.76	0.379
Separable Bone	13.84	25.87	18.10	2.17	0.288
Dressed Carcass, %					
Separable Fat	11.90	29.26	20.14	3.17	0.420
Separable Lean	56.56	69.27	63.70	2.55	0.339
Separable Bone	13.42	18.84	16.16	1.13	0.150
9-10-11 Rib Cut, %					
Ether extract	25.72	52.79	38.81	5.38	0.72
Ether extract, edible portion	24.73	57.19	41.28	6.35	0.85
Protein	10.90	20.69	13.87	1.75	0.23
Protein, edible portion	9.14	21.53	12.96	2.10	0.28
Water	28.01	53.49	39.84	4.73	0.63
Water, edible portion	30.50	63.70	44.95	5.89	0.79
Ash	5.77	10.89	7.30	0.93	0.12
Ash, edible portion	0.37	1.53	0.62	0.17	0.02
Dressed Carcass, %					
Ether extract	20.78	39.46	30.82	3.49	0.47
Ether extract, edible portion	19.23	41.92	31.47	4.16	0.56
Protein	14.44	20.06	16.51	1.26	0.17
Protein, edible portion	13.77	20.69	16.22	1.48	0.20
Water	40.50	51.46	45.84	2.86	0.39
Water, edible portion	44.58	58.86	51.42	3.52	0.47
Ash	5.17	7.76	6.42	0.51	0.07
Ash, edible portion	0.18	1.03	0.44	0.14	0.02

Table 5.2. Correlation and relationship between the physical composition of the 9-10-11 rib cut and the physical and chemical composition of the dressed carcass and the chemical composition of the 9-10-11 rib cut

Item	Coefficient of correlation	Estimating Equation	<i>P</i> - value	RMSE	Adj R ²
Separable fat of the 9-10-11 rib cut					
Separable fat of the dressed carcass	0.89	$Y = 4.375 + 0.471X$	<0.001	1.462	0.78
Ether extract of dressed carcass	0.83	$Y = 14.463 + 0.489X$	<0.001	1.980	0.68
Ether extract of dressed carcass, edible only	0.83	$Y = 12.020 + 0.581X$	<0.001	2.361	0.68
Separable lean of the 9-10-11 rib cut					
Separable lean of the dressed carcass	0.83	$Y = 42.962 + 0.428X$	<0.001	1.446	0.68
Protein of dressed carcass	0.41	$Y = 11.433 + 0.105X$	<0.01	1.158	0.15
Protein of dressed carcass, edible only	0.42	$Y = 10.122 + 0.126X$	<0.01	1.351	0.16
Moisture of dressed carcass	0.72	$Y = 25.154 + 0.427X$	<0.001	2.042	0.51
Moisture of dressed carcass, edible only	0.73	$Y = 25.761 + 0.530X$	<0.001	2.439	0.53
Separable bone of the 9-10-11 rib cut					
Separable bone of dressed carcass	0.59	$Y = 10.629 + 0.306X$	<0.001	0.916	0.33
Ash of dressed carcass	0.59	$Y = 3.937 + 0.137X$	<0.001	0.408	0.34
Ash of dressed carcass, edible only	0.14	$Y = 0.279 + 0.009X$	0.31	0.137	0.01

Table 5.3. Correlation and relationship between the chemical composition of the 9-10-11 rib cut and chemical composition of the dressed carcass and the 9-10-11 rib cut

Item	Coefficient of correlation	Estimating Equation	<i>P</i> - value	RMSE	Adj R ²
Ether extract of 9-10-11 rib cut					
Ether extract of dressed carcass	0.69	$Y = 13.605 + 0.444X$	<0.001	2.572	0.46
Protein of dressed carcass	-0.34	$Y = 19.583 - 0.079X$	0.01	1.193	0.10
Moisture of dressed carcass	-0.57	$Y = 57.785 - 0.308X$	<0.001	2.406	0.32
Ether extract of 9-10-11 rib cut, edible only					
Ether extract of dressed carcass, edible only	0.67	$Y = 13.351 + 0.439X$	<0.001	3.111	0.44
Protein of dressed carcass, edible only	-0.32	$Y = 19.262 - 0.074X$	0.02	1.410	0.09
Moisture of dressed carcass, edible only	-0.63	$Y = 65.746 - 0.347X$	<0.001	2.791	0.38
Protein of 9-10-11 rib cut					
Protein of dressed carcass	0.33	$Y = 19.583 - 0.079X$	0.01	1.973	0.09
Protein of 9-10-11 rib cut, edible only					
Protein of dressed carcass, edible only	0.30	$Y = 13.527 + 0.208X$	0.02	1.418	0.07
Moisture of 9-10-11 rib cut					
Moisture of dressed carcass	0.48	$Y = 33.992 + 0.297X$	<0.001	2.567	0.22
Protein of dressed carcass	0.22	$Y = 14.172 + 0.059X$	0.10	1.237	0.03
Moisture of 9-10-11 rib cut, edible only					
Moisture of dressed carcass, edible only	0.51	$Y = 37.570 + 0.308X$	<0.001	3.073	0.25
Protein of dressed carcass, edible only	0.24	$Y = 11.639 + 0.108X$	<0.01	1.379	0.15
Ash of 9-10-11 rib cut					
Ash of dressed carcass	0.65	$Y = 3.842 + 0.353X$	<0.01	0.382	0.42
Ash of 9-10-11 rib cut, edible only					
Ash of dressed carcass, edible only	0.12	$Y = 0.381 + 0.092X$	0.40	0.137	0.01

Table 5.4. Prediction equations derived from 9-10-11 rib physical and chemical (whole and edible portions) composition, USA and Canadian grading factors, HCW, DOF, Diet and ZH for the determination of carcass separable fat, lean and bone (SFC, SLC and SBC) and whole or edible carcass ether extract (EEC and EEC_e), carcass protein (PC and PC_e), carcass moisture (MC and MC_e) and carcass ash (AC and AC_e).

Item	Estimating Equation	P - value	RMSE	Adj R ²
<i>Physical Composition using Whole Rib Chemical Composition and Grading Factors</i>				
SFC	$Y = 45.261 + (0.961*FT) - (0.228*SBR) - (0.472*SLR)$	<0.001	1.360	0.82
SLC	$Y = 44.043 - (0.868*FT) + (0.055*LMA) + (0.336*SLR)$	<0.001	1.374	0.69
SBC	$Y = 12.612 - (0.011*HCW) + (0.494*SBR) + (0.083*SLR) - (0.730*AR)$	<0.001	0.690	0.66
<i>Chemical Composition using Whole Rib Chemical Composition and Grading Factors</i>				
EEC	$Y = -5.077 - (1.653*TRT) + (0.011*QG) + (1.522*REL) - (0.892*MS) + (0.776*FC) + (0.434*SFR) + (1.397*AR)$ $Y = 18.252 + (0.543*DOF) - (1.320*TRT) + (0.011*HCW) - (0.721*FT) - (0.914*KPH) - (0.411*REL) +$	<0.001	1.630	0.78
PC	$(0.225*FC) - (0.105*SFR) + (0.109*PR) - (0.226*AR)$	<0.01	0.998	0.39
MC	$Y = -58.526 + (1.786*TRT) - (0.946*FC) - (0.187*LMW) + (0.196*SLR) - (0.123*EER) - (0.972*AR)$	<0.001	1.770	0.61
AC	$Y = 1.894 + (0.167*DOF) - (0.262*TRT) + (0.030*SLR) + (0.023*MR) + (0.279*AR)$	<0.001	0.320	0.60
<i>Chemical Composition using Edible Rib Chemical Composition and Grading Factors</i>				
EEC _e	$Y = 51.476 - (1.440*TRT) + (0.009*QG) + (0.616*FC) - (0.581*SLR)$	<0.001	2.068	0.75
PC _e	$Y = 9.997 + (0.694*DOF) - (1.389*TRT) - (1.117*FT) - (0.988*KPH) + (0.448*FC) + (0.131*SLR)$	<0.001	1.182	0.38
MC _e	$Y = 62.560 + (1.999*TRT) - (1.039*FC) - (0.384*SBR) + (0.169*SLR) - (0.166*EER_e) + (2.691*AR_e)$	<0.001	2.140	0.64
AC _e	$Y = -1.144 + (0.062*DOF) - (0.150*TRT) + (0.001*HCW) + (0.021*SLR)$	<0.001	0.108	0.42
LM area = LMA; 12 th rib s.c. fat depth (FT); fat class (FC); % kidney-pelvic-heart fat (KPH); marbling score (QG); ribeye length (REL); muscle score (MS); TRT (C=0, Z=1); DIET (M=0, A=1); % of longissimus muscle in 9-10-11 rib (wt/wt) (LMW); Separable fat of the carcass = SFC; Separable lean of the carcass = SLC; Separable bone of the carcass = SBC; Separable fat of 9-10-11 rib = SFR; Separable lean of the 9-10-11 rib = SLR; Separable bone of the 9-10-11 rib = SBR; Ether extract carcass = EEC; ether extract of the 9-10-11 rib = EER; Moisture of carcass = MC; Moisture of 9-10-11 rib = MR; Protein of carcass = PC; Protein of 9-10-11 rib = PR; Ash of the carcass = AC; Ash of the 9-10-11 rib = AR; Ether extract of edible carcass = EEC _e ; ether extract of the 9-10-11 rib = EER _e ; Moisture of edible carcass = MC _e ; Moisture of edible 9-10-11 rib = MR _e ; Protein of edible carcass = PC _e ; Protein of 9-10-11 edible rib = PR _e ; Ash of the edible carcass = AC _e ; Ash of the 9-10-11 rib = AR _e				

CHAPTER 6
EFFECT OF DIETARY ENERGY LEVEL AND ZILPATEROL
HYDROCHLORIDE ON THE EMPTY BODY COMPOSITION
AND ENERGETICS OF BEEF STEERS

6.1 Abstract

A trial was conducted to examine the empty body (EB) composition, nutrient accretion and energetics of steers fed at maintenance (M) or an *ad libitum* (A) level of intake with zilpaterol hydrochloride (Z) supplementation. Single-sired, beef steers (n=56; 590 ± 36 kg) blocked (n=2) by BW and terminal implant were further sorted into pairs (n=14 per block) by BW. Pairs of steers were assigned to 0, 28 or 56 d of feeding, sorted into individual pens, and within 28 and 56 d to M or A feeding. Within 56d of feeding, steers within a pair were randomly assigned to either 20 d of Z (90 mg/hd daily) plus a 4 d withdrawal or to no ZH supplementation (C). Weights of all carcass and non-carcass components were recorded at slaughter; samples of hide and internal ground cavity were retained for proximate analysis. Carcasses were graded, following a 24 h chill then fabricated 48 h post-harvest. Samples of lean, fat and bone were retained for analysis. Data were analyzed using a mixed model; the fixed effect of treatment combination and random effects of block and pair with significance discussed at $P \leq 0.05$ and trends discussed at $P \leq 0.10$. Individual contrasts were constructed for d 0 vs. d 28, d 0 vs. d 56, d 28 vs. d 56, M vs. A and C vs. Z. Multiple-linear regression was used to evaluate the

predictive ability of live performance and carcass grading factors to estimate empty body (EB) composition. Fat content of the EB (EBF) and carcass (CF) were impacted by treatment with 56 d AC having greater EBF and CF ($P < 0.05$; 34.1 and 30.2%, respectively) than all other treatments except 28 d A. In addition, M steers had less ($P < 0.05$) EBF (30.44 v. 32.30%) and CF (30.29 v. 32.23%) than A steers. Moisture of the EB (EBM) and carcass (CM) tended to be impacted by diet ($P < 0.10$) in an inverse fashion to EBF and CF with Z increasing ($P < 0.05$) EBM and CM. Protein of the EB (EBP) and carcass (CP) tended to be affected by treatment on an absolute basis with Z steers having greater protein yield than C. Additionally, EBM, EBF and EBP daily gains were impacted by treatment ($P < 0.05$) with M having lower gains of all components than A ($P < 0.01$) and Z tending to increase moisture vs. C ($P = 0.06$). Heat production (HP) of AZ was greater than AC steers ($P < 0.001$; 0.211 vs. 0.184 Mcal/EBW^{0.75}) whereas MZ did not differ from MC ($P > 0.05$). As a result, efficiency of utilization of metabolizable energy intake (Kr) was 0.499 and 0.293 for C and Z steers respectively.

6.2 Introduction

Chemical composition, tissue accretion and energetic efficiency of domestic animals have been a focus of researchers for over a century. Tissue accretion of fat and protein differ as the animal's physiological age advances as well as with differing plane of nutrition (Koch et al., 1978). Maintenance energy requirements are a direct result of metabolic BW (BW^{0.75}; Fox and Black, 1984), however gain requirements are a result of the composition of gain; the proportion of fat and protein tissue accretion (Garrett, 1959). While consuming high energy dense diets, fat accretion of both empty body (EB) and carcass, occurs at an accelerated rate late in the growth phase (Simpfendorfer et al. 1974;

Fox and Black, 1984; Bruns et al., 1992). The increase in fat accretion as a proportion of gain results in an increased energy requirement for BW gain in the latter phases of the feeding period, resulting in reduced efficiency.

Zilpaterol hydrochloride (Z) is a β -adrenergic agonist fed 20 to 40 d prior to harvest that has been shown to increase gain efficiency, BW gain, HCW, dressed yield and carcass cutability (Montgomery et al., 2009ab; Elam et al., 2009; Vasconcelos et al., 2009; Hilton et al., 2010; Rathmann et al., 2012). Additionally, Z has been shown to increase carcass moisture and protein while decreasing empty body fat in Holstein steers (McEvers et al., 2014). Research with other β -adrenergic agonists including cimaterol and clenbuterol have reported increased protein deposition, decreased fat accretion and increased heat production in the first few days of administration (Ricks et al., 1984; Macrae et al., 1988; Reeds and Mersmann, 1990; Rikhardsson et al., 1991). Therefore, Z may potentially improve the efficiency of metabolizable energy used for gain by changing the composition of tissue growth.

Understanding and predicting growth composition at latter stages of finishing with or without β -adrenergic agonists using live and carcass performance factors is important to the beef industry. Equations exist to predict empty body fat, carcass fat, quality grade and yield grade by accounting for frame size, previous plane of nutrition, breed, growth implants and ionophores (Fox and Black, 1984). Multiple studies have sought to estimate EBW and composition based upon carcass grading factors and carcass composition (Lofgreen et al., 1962; Garrett and Hinman, 1969; Fox et al., 1976; Guiroy et al., 2001). To date, little research has focused on the impact of extended days on feed on the EB tissue changes and growth energetics with or without a β -adrenergic agonist in

beef steers. There is a need to understand the impact of Z on the efficiency of gain as well as to predict EB and carcass chemical composition in cattle fed to later physiological end points than previously researched. Therefore, the objectives of this experiment were to quantify EB composition and energetics in beef steers fed at maintenance (M) or *ad libitum* (A) levels of intake with or without Z supplementation.

6.3 Materials and Methods

All experimental procedures involving live animals were approved by the Animal Care and Use Committee at West Texas A&M University and adhered to the regulations in the Guide for the Care and Use of Agricultural Animals in Agricultural Research and Teaching (Federation of Animal Science Societies, 2010, Savoy, IL).

6.3.1 Live Cattle Procedures and Experimental Design

Live cattle procedures, experimental design, carcass grading and harvest procedures were previously reported (Walter et al., 2015. *Effect of dietary energy level and zilpaterol hydrochloride on the live performance, grading performance and harvest yields of beef steers*. Chapter II). Briefly, single sired steers (n=60; BW 574±36 kg) were purchased and transported to a commercial research feedlot (Canyon, TX) where steers were penned in two pens (n = 30 per pen) until further weighing and sorting. Cattle BW were recorded on d-5 and d-6 with 28 steers selected per block (Block 1 initial BW = 589±25 kg and Block 2 initial BW = 614±39 kg). Within each block, two steers with BW furthest from the mean (heaviest and lightest steer in Block 1 and two lightest steers in Block 2) were penned in individual pens as alternates for the duration of the trial resulting in a total of 56 steers being randomized to treatments. Steers were sorted into individual pens on d-4 with Block two starting on trial 28 days after block 1. Cattle were

weighed after 9 h withdrawal from feed and water on d -1 and 1, 27 and 28 and 55 and 56.

The trial was constructed as a multi-factorial treatment design with three harvest dates, two dietary energy levels and with or without Z supplementation. Cattle were blocked by terminal implant status (Revalor[®] XS or Revalor[®] S, Merck Animal Health, Summit NJ) and randomized to pairs on the sort days (d -5 and d -6) by weight. Pairs were randomized to harvest day (0, 28 or 56 d) and dietary level (maintenance or *ad libitum*) within harvest date. Within d 56 harvest date, one animal within each pair for both maintenance and *ad libitum* level of feeding was randomly assigned to Z supplementation (90 mg per head daily; Merck Animal Health; Summit, NJ). All steers were harvested at a commercial packing plant (Caviness Packing Company, Establishment 675; Hereford, TX).

6.3.2 Sample Collection

Hide samples (200 cm²) were obtained at harvest from the ventral side of the animal mid-way between the flank and naval. Individual hide samples were placed in whirl-Pak bags, vacuum sealed and frozen at -20 °C. Prior to further processing, hide samples were thawed at 4 °C for 8 h and then cut into 1 cm wide strips and further into 1 cm² pieces. Hide pieces were then mixed and sub-sampled into three homogenous Whirl-Pak bags with two of the bags vacuum sealed and frozen at -20 °C. One set of each hide sample was sent to a commercial lab (Servi-Tech, Amarillo, TX) where the samples were individually dried under pressure at 40 °C for 96 h using a Blue M DCRI-326-F-PM oven to determine initial loss of moisture upon drying. Hide samples were then ground through

a 1 mm screen using a Retsch GmbH-Rotor Beater Mill SR 300 (Retsch GmbH, Haan Germany).

At harvest, internal viscera and offal components were collected, weighed and transported to the Meat Lab at West Texas A&M University. As discussed in Walter et al. (Walter et al., 2015. *Effect of dietary energy level and zilpaterol hydrochloride on the live performance, grading performance and harvest yields of beef steers*. Chapter II) components were weighed, omental fat trimmed and weighed separately from stomachs, gastrointestinal tract weighed full, cleaned and weighed empty to determine gut fill and conversely EBW. After all internal cavity components were weighed, viscera was placed on dry ice, cut into smaller chunks and ground after removal from dry ice through a grinder (Butcher Boy Machines International LLC, Selmer, TN). The entire offal components were ground through a 9.5 mm plate, mixed extensively for 5 minutes and a ~20 kg sample was then passed through a 3.2 mm plate. During the fine grind, the mixture was sampled three times (~100 g amounts), the three samples were then mixed thoroughly and sampled into three composite samples, placed in Whirl-Pak bags and frozen at -20°C for further analysis.

At 24 h post-harvest, carcasses were evaluated for marbling score, lean and skeletal maturity, 12th rib s.c. fat depth (cm) and LM area (cm²). A final quality grade and calculated yield grade was determined (USDA, 1997). At 48 h post-harvest, left sides of the carcasses were fabricated into primals and weighed to determine a cold carcass side weight. Carcass primals were fabricated according to industry standards (IMPS) with all bone and fat trim weighed separately. Sampling of carcass lean, fat and bone is discussed in Walter et al., 2015. (*Effect of dietary energy level and zilpaterol*

hydrochloride on the live performance, grading performance and harvest yields of beef steers. Chapter II).

6.3.3 Sample Analysis

Feed and orts were analyzed for energy (Automatic Isoperibol Calorimeter, Parr Instrument Company, Moline, IL). Tissue samples (carcass lean, carcass fat, carcass bone, internal cavity) were lyophilized (Bulk Tray Dryer, Labconco Corp, Kansas City, MO), ground in a 1 L stainless steel Waring blender (700S, Waring Commercial, Torrington, CT) and dried at 105°C for 9 h for further determination of lab DM and analyzed for energy (Automatic Isoperibol Calorimeter, Parr Instrument Company, Moline, IL). Internal cavity, carcass lean, carcass fat and carcass bone samples were then analyzed for crude protein (AOAC method 976.06), ether extract (AOCS #Ba-3-38) and ash (AOAC #942.05) by SDK Laboratories (Hutchinson, KS). Additionally, for internal cavity samples, crude fiber was analyzed on the ether extract free sample (AOAC #962.09). Hide samples were analyzed by ServiTech Laboratories (Amarillo, TX) for crude protein (AOAC #990.03), ether extract (AOAC #2003.06), ash (AOAC #942.05) and crude fiber (AOAC #978.10). Hide samples were also analyzed for energy (Automatic Isoperibol Calorimeter, Parr Instrument Company, Moline, IL).

Blood samples (2×10 mL) were obtained at harvest in duplicate (15 mL tube, 93000-028 VWR, Radnor, PA). Blood samples were not available for analysis due to malfunction of the freeze drier and loss of sample integrity. Therefore, blood composition was based on the results of Duarte et al. (1999) with 80.9% moisture, 17.3% protein, 0.23% fat, 0.07% carbohydrates and 0.62% minerals (ash). The energetic

composition of blood was based on data from Merrill and Watt (1973) as presented by FAO (2003) with 9.02 kcal/g for fat and 4.27 kcal/kg for protein.

6.3.4 Statistical Analysis

The mixed procedure of SAS (SAS Institute, Inc. Cary, NC) was used for model analysis with the fixed effects of treatment combinations and the random effects of block and pair. Single df contrasts of d0 v. d28, d0 v. d56, d28 v. d56, M v. A and C v. Z were calculated. All regression analysis was performed using the REG procedure of SAS (SAS Institute, Inc. Cary, NC). Multiple-linear regression using the best fit model as determined by stepwise, backward or forward regression was used to evaluate the predictive ability of TRT (C=0, Z=1) DIET (M=1, A=2), DOF (28=1, 56=2), BW, DMI, daily metabolizable intake (MEI), ADG, gain:feed (GF), BW gain (BWG), hip length (HL), hip width (HW), shoulder height (SH), length from point of shoulder to hip (BL), hip height (HH), HCW, LM area (LMA), 12th rib s.c. fat depth (FT), percent kidney-pelvic-heart fat (KPH) and marbling score (QG), to estimate empty body chemical composition. Collinearity diagnostics using the TOL, VIF and COLLIN options of SAS was used to evaluate and prevent the use of multicollinearity if variables resulted in a condition index > 10.

6.4 Results

6.4.1 Empty Body Composition

Weights of empty body, carcass and internal cavity are discussed in Walter et al. (*Effect of dietary energy level and zilpaterol hydrochloride on the live performance, grading performance and harvest yields of beef steers*. Chapter II). Empty body ether extract (EBF_a), moisture (EBM_a) and ash (EBA_a) were all effected by treatment as an

absolute weight ($P < 0.01$; Table 1) whereas treatment tended ($P = 0.07$) to effect EB protein (EBP_a) yield. Relative to EBW, treatment impacted ($P \leq 0.05$) ether extract (EBF), moisture (EBM) and ash (EBA) and tended to impact ($P = 0.08$) moisture (EBM) as a % of EBW. Maintenance steers had lower EBF_a ($P < 0.01$; 164.9 vs. 192.4 kg, respectively) than A steers; whereas 56 d steers tended to have greater EBF_a ($P = 0.09$; 181.8 vs. 160.7 kg, respectively) than steers harvested on d0. As a percent of EB, EBF was again lower in M than A fed steers ($P = 0.03$; 30.4 vs. 32.2 % EBW, respectively). Additionally, d 56 AC cattle had more ($P = 0.05$) EBF (34.1%) than all other treatments except 28 d A steers (31.8 %).

Similar to ether extract, EBM_a increased ($P = 0.03$) over days on feed from d 28 to 56 (250.3 vs. 264.0 kg, respectively) and with increasing dietary energy intake, A fed (270.4 kg) cattle had greater EBM_a ($P < 0.01$) than M fed steers (249.7 kg). Conversely, EBM decreased over days on feed ($P = 0.04$) with d 28 and d 56 cattle exhibiting having lower EBM than d 0 steers (45.9 and 45.8, respectively vs. 47.8%). Dietary energy intake had a similar effect on EBM_a as EBF_a; steers fed M intakes had less EBM_a ($P < 0.001$; 249.7 vs. 270.1 kg, respectively) than A steers. Expressed as a percentage, EBM tended to be lower in A than M fed steers ($P = 0.08$; 45.1 vs. 46.3%, respectively), thus inverse to increased EBF in A than M fed steers. Additionally, Z supplementation increased ($P < 0.01$) EBM_a compared to control (271.3 vs. 256.8 kg, respectively) cattle and increased moisture as a percent of EBW ($P = 0.04$; 46.6 vs. 45.1% for ZH and control, respectively). In parallel fashion to EBM_a, EBP_a tended ($P = 0.09$) to be greater in cattle fed 56 d than d 0 (95.2 vs. 88.6 kg, respectively) whereas A intake increased ($P < 0.01$; 97.1 vs. 90.6 kg, respectively) EBP_a vs. M fed cattle. As a percent of EBW,

protein was not affected by treatment ($P = 0.49$), indicating that protein is constant relative to EBW. Empty body ash content was impacted by treatment; 56 d cattle had greater ($P < 0.05$) EBA_a as compared to d 0 and d 28 cattle (35.0 vs. 30.9 and 33.0 kg for 56, 0 and 28 d, respectively); similarly EBA as a percent of EBW increased over days on feed with 28 and 56 d cattle having greater ash than d 0 ($P = 0.02$; 6.0 and 6.1 vs. 5.7 %). Additionally, M cattle exhibited higher ash content than cattle fed A intakes ($P < 0.01$; 6.3 vs. 5.8%, respectively).

Early research examining the compositional growth of cattle noted that protein and ash increase while water content decreases rapidly in young animals but as an animal reaches physiological age, proportional changes of protein, ash and water in the fat-free empty body are relatively constant (Armsby and Moulton, 1925 and Blaxter, 1962). Therefore, the composition of the fat free EB (FFEB) was examined (Table 1). Treatment differences existed for fat free empty body weight (FFEBW) ($P < 0.01$) with A and Z treatment increasing FFEBW ($P = 0.04$) by 29 and 17 kg vs. M and C treatments, respectively. While d 56 cattle had greater FFEBW than d 28 cattle ($P = 0.05$), this was primarily due to the effect of Z at d 56 as well as reduced BW of M steers at 28 d. With respect to chemical composition of the FFEB, ash was affected by treatment ($P < 0.01$); whereas, moisture tended to be affected by treatment ($P = 0.08$). Moisture was increased ($P < 0.05$) in d0 steers than d 28 and d 56 while ash was decreased in d 0 than d 28 and d 56. Additionally, Z tended to increase ($P = 0.08$) moisture and decrease ($P = 0.01$) ash as a percent of the FFEB; whereas, M increased the percentage of ash in the FFEB relative to A ($P < 0.01$). Percentage of protein in the FFEB was unaffected by treatment ($P = 0.29$).

6.4.2 Carcass Composition

Treatment also impacted whole and edible carcass composition similarly to EB composition (Table 2). Absolute weights of carcass moisture (CM_a), ether extract (CF_a) and ash (CA_a) were affected by treatment ($P < 0.05$) whereas carcass protein (CP_a) tended to be impacted by treatment ($P = 0.08$). As a percentage of edible carcass, fat (CF_E) and moisture (CM_E) were impacted by treatment ($P \leq 0.05$, Table 2). As a percentage of carcass weight, carcass moisture (CM), ash (CA) and ether extract (CF) were impacted by treatment ($P < 0.05$). Carcass ether extract was increased in steers harvested on d 56 vs. d 0 ($P = 0.05$, 118.0 vs. 100.7 kg, respectively) primarily because A fed steers exhibited increased CF_a vs. M fed cattle ($P < 0.01$; 124.4 vs. 106.7 kg, respectively). As a percentage of carcass weight, CF was reduced ($P = 0.04$) in M vs. A fed steers (30.3 vs. 32.1 %) similar to EBF (Table 1). Additionally, CF tended ($P < 0.10$) to be higher in steers harvested on d 28 (31.3 %) and d 56 (31.2 %) vs. d 0 (29.0 %) steers. As a percentage of edible tissue, M cattle had lower CF_E than A cattle ($P = 0.05$; 33.0 vs. 30.8%, respectively) and similar to the whole carcass, AC steers had 4.13% more CF_E than AZ steers.

Similar to EBM_a , CM_a was increased in d 56 vs. d 28 steers ($P < 0.01$; 170.9 vs. 158.5 kg, respectively) and in cattle fed at A vs. M intake ($P < 0.001$; 172.9 vs. 161.1 kg, respectively). Conversely, steers supplemented with Z exhibited increased CM_a ($P < 0.001$; 177.9 vs. 163.8 kg, respectively) over C steers. Steers fed A for 56 d and supplemented with Z had higher CM_a than all other treatments ($P < 0.05$) whereas MZ steers did not differ in CM_a from 56 d MC fed cattle but did have a higher CM_a ($P < 0.05$) than 28 d M steers. As a percentage of the carcass, moisture was increased in d0

cattle vs. d 28 and d56 ($P < 0.01$; 48.0 vs. 44.7 and 45.6%, respectively) and CM was also increased in Z vs. C steers ($P = 0.04$; 46.5 vs. 44.7%). Moisture of the edible portion of the carcass decreased in cattle fed 28 d vs. d0 ($P = 0.01$; 53.58 vs. 50.13% , respectively) and tended to decrease ($P = 0.07$) in cattle fed 56 d vs. d0. Inverse to CF_E , cattle fed M had increased ($P = 0.03$) CM_E vs. A (51.92 vs. 49.97%) whereas cattle fed Z exhibited a tendency for increased CM_E vs C ($P = 0.08$; 52.34 vs. 50.37%, respectively)

Additionally, CP_a tended ($P = 0.08$) to be affected by treatment; A steers exhibited increased CP_a vs. M fed steers ($P = 0.02$; 63.7 vs. 58.9 kg, respectively). Steers fed for 56 d exhibited higher CP_a vs. d 0 steers ($P = 0.04$; 61.8 vs. 55.7 kg, respectively). As a percentage of the carcass, CP was not different ($P = 0.26$). Carcass ash was greater ($P \leq 0.05$) in d 56 (24.3 kg) harvested steers vs. d 0 (21.5 kg) and d 28 (22.9 kg) whereas as a percent of the carcass, ash was increased ($P < 0.01$) in steers fed M (6.7%) vs. A (6.2%) and tended to be reduced ($P = 0.07$) in steers supplemented with Z.

6.4.3 Internal Cavity Tissue Composition

Internal cavity (IC) chemical composition components of ether extract (ICF_a), moisture (ICM_a) and ash (ICA_a) were affected by treatment ($P \leq 0.05$; Table 3). In addition, treatment also affected ($P < 0.01$) ash (ICA) and tended ($P = 0.07$) to alter protein (ICP) when expressed as percentage of IC. Internal cavity ether extract was reduced in M vs. A steers ($P < 0.001$; 39.6 vs. 47.6 kg) and tended to be reduced in Z vs. C steers ($P = 0.07$). Day 56 AC steers had greater ICF_a than d 0 steers ($P < 0.05$; 51.7 vs. 42.0 kg, respectively) and all M steers. Internal cavity moisture was increased ($P < 0.01$) in A vs. M (37.0 vs. 31.6 kg, respectively) whereas d0 steers had intermediate internal cavity moisture (34.0 kg). The intermediary value for d0 steers vs. similar values for A

steers is likely due to the abundant handling at the start of trial with weights taken off feed and water d -4 and d -5 as well as d 0 and d 1 with extensive handling and penning as individual steers on d-4. Protein as a percentage of internal cavity tended to be impacted by treatment ($P = 0.07$); cattle fed at M exhibited increased ICP ($P = 0.03$; 8.38 vs. 7.64% for M and A, respectively) whereas d 56 steers tended to exhibit decreased protein as a percentage of internal cavity. This effect was primarily due to reduced ICP percentages in A steers at 56 d and inversely numerical increases in ICF with A steers. Ash of the internal cavity increased over days on feed with d 28 and d 56 cattle exhibiting higher ash of the internal cavity ($P < 0.01$) than d0 harvested steers and M expressed a higher ash content vs. A fed steers ($P = 0.04$; 0.8 vs. 0.6 kg, respectively).

6.4.4 Energy and Nutrient Accretion

With respect to energy, total energetic content of the empty body was affected by treatment ($P < 0.01$, Table 4) with M steers having reduced ($P < 0.01$; 2039 vs. 2337 Mcal, respectively) retained energy vs. A steers and d 56 steers tending to have a greater retained energy vs. d 0 ($P = 0.09$; 2220 vs. 2009 Mcal, respectively). Retained energy (kcal/kg of EB) was decreased in M cattle vs. A ($P = 0.01$) fed steers and in Z vs. C steers ($P = 0.05$). The effect of Z was primarily driven by the difference between 56 d AC (4067.7 kcal/kg EB) vs. 56 d AZ steers (3781.1 kcal/kg EB). Carcass energy was increased in d 56 vs. d 0 ($P = 0.05$) as AC steers at d 56, had higher ($P = 0.01$) energy in the carcass (1590.4 Mcal) than all other treatments except d 56 AZ steers (1514.1 Mcal) and 28 A fed cattle (1145.6 Mcal). Concurrently, M steers had reduced ($P < 0.01$) energy in the carcass vs. A steers (1327.6 vs. 1516.7 Mcal, respectively). Energy (kcal/kg of carcass) was reduced ($P = 0.04$) in M fed steers vs. A (3771.8 vs. 3917.2 kcal/kg,

respectively) steers and tended to be reduced in steers supplemented with Z vs. C ($P = 0.08$; 3748.8 vs. 3915.5 kcal/kg, respectively) cattle. Additionally, cattle fed 28 d exhibited ($P = 0.04$) increased energy composition per kg of carcass vs. d 0 cattle. Energy of the internal cavity was decreased in cattle fed M vs. A ($P < 0.001$; 413.8 vs. 494.9 Mcal, respectively).

Nutrient accretion and retained energy were impacted by dietary energy level, days of harvest and ZH treatment ($P < 0.05$, Table 5). Nutrient accretion of empty body ether extract (EBF), moisture (EBM), protein (EBP) and energy was higher in cattle fed A vs. M ($P < 0.001$). Empty body fat accretion was highest in A cattle harvested on d 28 (895 g/d) and AC cattle harvested on d 56 (833 g/d) whilst AZ cattle (530 g/d) were not different from all other A cattle ($P > 0.05$) but were also not different from 28d M cattle (247 g/d). Empty body moisture was increased in cattle fed A vs. M ($P < 0.001$; 212 vs. -216 g/d, respectively). Additionally, empty body moisture (EBM) accretion was increased in cattle fed to 56 d vs. 28 ($P = 0.04$; 80 vs. -167 g/d, respectively) and tended to be increased in cattle fed Z vs. C ($P = 0.06$; 193 vs. -66 g/d, respectively). Empty body protein accretion was increased in A vs. M fed cattle regardless of harvest d or Z treatment ($P < 0.001$; 208 vs. 57 g/d for A vs. M, respectively). Empty body energy accretion was decreased in cattle harvested on d 56 vs. d 28 ($P = 0.03$; 3.79 vs. 6.06 Mcal/d, respectively) primarily due to AZ (5.529 Mcal/d) cattle having reduced empty body energy accretion vs. d 28 A (10.15 Mcal/d) cattle. As expected, M had reduced empty body energy accretion vs. A ($P < 0.001$; 8.00 vs. 1.09 Mcal/d).

With respect to overall energetics, metabolizable energy intake and subsequently retained energy (RE) and heat production (HP) were higher in cattle fed A vs. M intakes.

Retained energy when expressed on a metabolic body weight basis ($EBW^{0.75}$) was reduced ($P < 0.05$) in 56d AZ cattle ($0.046 \text{ Mcal/kg } EBW^{0.75}$) vs. 28 d A ($0.087 \text{ Mcal/kg } EBW^{0.75}$) while 56 d AC ($0.071 \text{ Mcal/kg } EBW^{0.75}$) cattle were not different ($P > 0.05$) from all other A fed cattle. Conversely, HP in AZ cattle was greater than AC cattle ($P < 0.05$; 0.211 vs. $0.184 \text{ Mcal/kg } EBW^{0.75}$, respectively). No difference ($P > 0.05$) was detected between M cattle fed Z or C and harvested on d 28 or 56 for RE or HP with overall averages of (0.009 and $0.116 \text{ Mcal/kg } EBW^{0.75}$ for RE and HP of M fed cattle, respectively).

The regression of RE on MEI resulted in an equation for C cattle of $\{P < 0.001, \text{RMSE} = 27.525, \text{Adj. } R^2 = 0.65; (\text{RE} = -53.322 + 0.499 \cdot \text{MEI})\}$ and for Z cattle of $\{P < 0.01, \text{RMSE} = 19.429, \text{Adj. } R^2 = 0.52; (\text{RE} = -28.781 + 0.293 \cdot \text{MEI})\}$. Therefore, the efficiency of metabolizable energy used for growth (K_r) was 0.499 for control fed steers and 0.293 for zilpaterol steers. Heat production was log transformed with C cattle as $\{P < 0.001, \text{RMSE} = 0.097, \text{Adj. } R^2 = 0.57; (\log \text{HP} = -1.864 + 0.002 \cdot \text{MEI})\}$ and for Z $\{P < 0.001, \text{RMSE} = 0.075, \text{Adj. } R^2 = 0.77; (\log \text{HP} = 1.814 + 0.002 \cdot \text{MEI})\}$.

6.4.5 Carcass and Empty Body Prediction

The current study also investigated the ability of DOF, ZH treatment, diet and live performance feeding factors (final BW, ADG, gain:feed, DMI, daily metabolizable energy intake and biometric data) to predict carcass grading attributes (Table 6). In addition, treatment and live performance feeding factors were used with USA grading factors to predict empty body weight and composition (Table 7). Hot carcass weight was strongly predicted from treatment and live performance factors $\{P < 0.001, \text{RMSE} = 6.800 \text{ Adj. } R^2 = 0.96; (\text{HCW} = 10.475 + (13.184 \cdot \text{TRT}) + (0.571 \cdot \text{BW}) - (11.233 \cdot \text{ADG}) +$

(3.404*DMI)} whereas adjusted 12th rib subcutaneous fat thickness and USDA calculated yield grade (YG) $\{P < 0.001, \text{RMSE} = 0.684, \text{Adj. } R^2 = 0.44; (YG = -3.964 + (0.317*\text{DOF}) + (1.700*\text{DIET}) + (0.008*\text{BW}) - (0.929*\text{ADG})\}$ were moderately predicted from treatment and live performance factors. Both LMA $\{P < 0.01, \text{Adj. } R^2 = 0.18, \text{RMSE} = 6.418 (Y = 53.745 + (0.057*\text{BW}))\}$ and marbling score (MARB) $\{P = 0.04, \text{RMSE} = 54.006, \text{Adj. } R^2 = 0.07 (Y = 392.290 + (6.027*\text{DMI}))\}$ were weakly correlated from live performance factors.

Empty body weight (EBW) was strongly correlated with treatment and live performance factors as well as with carcass grading factors and a combination of live and carcass grading factors $\{(P < 0.001, \text{RMSE} = 6.781, \text{Adj. } R^2 = 0.99 (EBW = 40.628 + (5.150*\text{DIET}) - (11.065*\text{TRT}) + (1.425*\text{HCW}) - (0.267*\text{LMA}))\}$. Empty body fat (EBF) was moderately predicted using treatment and live performance factors ($P < 0.001, \text{RMSE} = 2.530, \text{Adj. } R^2 = 0.31$) or carcass grading factors ($P < 0.001, \text{RMSE} = 2.015, \text{Adj. } R^2 = 0.54$) with the combination of both live and carcass grading factors and treatments resulting in the best validation of EBF $\{(P < 0.001, \text{RMSE} = 1.790, \text{Adj. } R^2 = 0.63) (EBF = 14.788 - (1.641*\text{DIET}) - (1.964*\text{TRT}) + (0.041*\text{HCW}) + (1.962*\text{FT}) - (0.094*\text{LMA}) + (1.957*\text{KPH}) + (0.010*\text{MARB}))\}$. Empty body moisture (EBM) using carcass grading factors and live performance resulted in the highest prediction ability for EBM $\{(P < 0.001, \text{RMSE} = 1.651, \text{Adj. } R^2 = 0.50) (EBM = Y = 56.867 + (1.616*\text{DIET}) + (1.922*\text{TRT}) - (0.041*\text{HCW}) - (1.264*\text{FT}) + (0.072*\text{LMA}) - (1.020*\text{KPH}))\}$. Empty body protein (EBP) was poorly predicted ($P < 0.01, \text{Adj. } R^2 = 0.12$) with live performance factors and treatments whereas the addition of carcass grading factors resulted in moderate prediction of EBP $\{(P < 0.001, \text{RMSE} = 0.797, \text{Adj. } R^2 = 0.35) (EBP$

= 19.349 + (0.300*DOF) – (0.513*TRT) – (0.428*FT) – (1.075*KPH)}. Empty body ash (EBA) was weakly predicted by carcass grading factors only ($P < 0.001$, Adj. $R^2 = 0.30$) whereas the addition of live performance and treatment factors resulted in moderate prediction of EBA $\{(P < 0.001, \text{RMSE} = 0.291, \text{Adj. } R^2 = 0.57) (\text{EBA} = 8.809 + (0.268*\text{DOF}) - (0.171*\text{DIET}) - (0.004*\text{HCW}) - (0.165*\text{FT}) - (0.162*\text{KPH}) - (0.002*\text{MARB}))\}$.

6.5 Discussion

Results from the current study detail changes to empty body composition primarily due to dietary energy intake and Z supplementation at A level of intake. Empty body fat and EBM exhibited an inverse relationship. Empty body fat increased with increased dietary energy intake and in AC steers EBF increased 15.5% from d 0 to d 56 whereas EBW increased 58.4 kg for every 1% increase in EBF, EBW increased 12.58 kg. Guiroy et al. (2001) reported that for every 14.26 kg increase in empty body weight, EBF increased by 1%. Differences between trials may reflect physiological age differences of cattle; as steers in the current trial were harvested within a 56 d window in an extended day feeding trial whereas Guiroy et al. (2001) compiled data from multiple trials spanning 1977 to 2001. Steers fed at M and AZ steers did not exhibit an increase in EBF vs. d 0 steers while on an actual basis, AZ steers had increased empty body fat vs. d 0 but not d 28A steers. Therefore, Z supplementation effectively halted fat accretion whereas M cattle essentially maintained BW as well as tissue composition. The decrease in EBF and concurrent increase in EBM with Z totaled 3.12 and 2.47% for EBF and EBM, respectively in A fed steers with no difference between M fed steers. With respect to previous research on empty body composition and Z, McEvers et al. (2014) reported

Holstein steers fed Z had increased EBM (1.3%) and reduced EBF (1.7%). The difference between the studies may again be due to physiological age of cattle with the Holsteins harvested over 11 time points separated by 28 d and thus encompassing cattle that were lighter and at a reduced physiological age than cattle in the current study. Other differences may be due to inherent breed and composition of gain differences between Holsteins and beef type steers.

In contrast to A steers, M steers retained constant EB tissue composition across days and treatment. Koch et al. (1978) examined the growth of metabolically active tissue (skeletal and gastrointestinal tract muscle) and fat over time and with differing energy intakes. The authors reported fat to increase linearly over time across all dietary intakes but fat accretion also remained parallel to energy intake. The authors concluded that energy intake resulted in changes to physiological composition when harvested at a time constant endpoint as energy level and age resulted in differences to protein and fat synthesis (Koch et al., 1978). In the current study, EBF did not change over days in M cattle and remained constant to EBW. Essentially, M intake halted growth and cattle harvested 56 d later exhibited similar tissue composition as cattle harvested on d0. Cattle fed AZ had a similar empty body composition as cattle harvested 28 d prior but with additional weight gain. Previous results of Rathmann et al. (2012) observed a similar trend in heifers supplemented with Z and serially harvested.

Carcass and internal cavity exhibited similar composition changes as the empty body. Carcass fat of the whole and edible carcass remained constant in M cattle regardless of d and Z treatment whereas AC steers increased CF 5.25% over 56 d and AZ steers did not increase CF above d 0 or d28 A steers. As a result, carcass fat decreased by

3.45% while carcass moisture increased 2.97% for AZ steers with similar results on the edible only portion as well. McEvers et al. (2014) reported a similar but reduced response of 2.0% decrease in fat and 1.4% increase in moisture of the edible carcass. Ricks et al. (1984) fed clenbuterol for 98 d at 10 or 500 mg/head/d. In steers fed 10 mg/head/d of clenbuterol, carcass protein increased by 2% and moisture increased by 4.6% whereas fat decreased by 7.2 % as estimated by 9-10-11 rib dissection. With respect to dietary energy intakes, Bond et al. (1982) observed an 8% decrease in CF (36 vs. 44%) and a 6% increase in carcass lean in cattle fed restricted intakes vs. A at 48 months of age (when cattle were of similar weight range to the current trial). Differences in overall CF content are likely due to different physiological ages of the cattle although at similar weights thus exemplifying the difference in genetic selection over the last several decades.

The current study quantified daily accretion rates of EBF, EBP, EBM and empty body energy across dietary treatments. Steers fed A accreted 753 g of fat and 208 g of protein per day whereas steers fed M accreted 133 g of fat and 57 g of protein per day. Similar to the current study, Murphy and Loerch (1994) reported daily protein and water growth of the carcass remained constant in steers fed 80, 90 or 100% of A; an increasing linear effect of fat growth (0.28 to 0.52 kg/d) was observed as intake increased from 80 and 100% A. Carstens et al. (1991) examined the growth of body components and reported allometric accretion of carcass chemical components to be similar across previously restricted and unrestricted steers. Additionally, Koch et al. (1978) reported cattle synthesized 105 g of protein and 274 g of fat daily on a high plane of intake whereas cattle fed a low plane of nutrition accreted 65 g of protein and 43 g of fat daily.

Differences between trials are likely the result of time on feed and physiological end point of harvested cattle.

Maintenance fed steers fed Z or C exhibited no difference in HP in the current study. Previous research by Rikhardsson et al. (1991) observed no difference in heat production after d 4 of cimaterol treatment whereas MacRae (1988) reported an increase in heat production in the first 5 d of clenbuterol administration in wethers. Additionally, in steers fed zilpaterol at maintenance intake levels, heat production on a metabolic BW basis did not change when measured on d 12 -16 of Z feeding (Walter et al., 2015). As reported by Rikhardsson et al. (1991), Caine and Mathison (1987) reported cimaterol to increase Kr from 0.41 to 0.67 in lambs treated with cimaterol for 75 d. In contrast, Rikhardsson et al. (1991) observed no difference in Kr between ewe lambs treated with cimaterol (0.49) or control (0.48). With respect to the current study, steers fed A and supplemented with Z had a reduced Kr vs. AC (0.293 vs. 0.499, respectively) when retained energy was regressed on metabolizable energy intake. This difference may be due to the difference in the composition of gain even though Z results in increased overall gain. With respect to the composition of energy gain, Koch et al. (1978) reported in cattle fed at high planes of intake, protein energy was 18% of total energy gain whereas it comprised 46% of total energy gain for cattle on a low plane of intake. Furthermore, in ewe lambs treated with cimaterol, Rikhardsson et al. (1991) reported an 11% increase in energy stored as protein on a metabolic BW basis when estimated from HP and nitrogen balance. Thus the shift of empty body composition resulting in decreased fat accretion and a reduced physiological age at a higher EBW may result in reduced efficiency of gain.

Research has sought to standardize live weight by conversion to an EBW in order to account for large differences in gut fill or different planes of growth (Lofgreen et al., 1962). Methods to do so include carcass adjusted performance utilizing standard dressing percentages for initial BW and final dressing percentages to adjust final BW or the prediction of EBW and composition based upon carcass and live performance factors (Fox et al., 1976; Fox and Black, 1984; Guiroy et al., 2001). Results of the current trial illustrate that 99% of the variation in EBW could be predicted utilizing treatment factors, live performance and carcass grading ($P < 0.001$). The addition of live performance marginally improved the prediction of EBF, EBP and EBA to 64, 48 and 61% of the variation vs. treatment and carcass grading factors alone. Empty body moisture prediction was not improved with the addition of live performance factors; treatment and carcass grading factors accounting for 50% of the variation. Garrett and Hinman (1969) reported 96 to 99% of EBW and composition could be predicted using HCW and carcass chemical composition. Fox et al. (1976) reported a linear relationship between HCW and EBW $\{Y = (1.40*HCW) + 40.2\}$ that was similar to research by Garrett and Hinman $\{Y = 1.36*HCW + 30.3\}$. Fox and Black (1984) using data from previous trials (Simpfendorfer, 1974) reported an equation for $EBF = -0.61 + 0.037*EBW + 0.00054*EBW^2$. Crouse et al. (1974) used live performance factors to predict carcass grading performance; quality grade was poorly estimated with only 13% of the variation predicted by live performance factors while yield grade prediction was 51% between slaughter groups.

Results of the current study indicate that dietary intake level, days and Z treatment result in compositional differences in the empty body and carcass. Steers fed M had

reduced EBF and CF but increased EBM and CM vs. A steers. Steers supplemented with Z had increased CM and EBM and tended to have increased actual protein of the empty body and carcass vs. C steers. Additionally, EB moisture, fat and protein daily gains were impacted by treatment ($P < 0.05$) with M decreasing gains of all components v. A ($P < 0.01$) and Z tended to increase moisture v. C ($P = 0.06$). Heat production did not differ between C and Z steers at M intake but HP of AZ was higher than AC steers. As a result, efficiency of gain was reduced in steers supplemented with Z.

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Table 6.1. Chemical analysis of the empty body (EB) of beef steers fed to 0, 28 or 56 days on feed and given maintenance (M) or *ad libitum* (A) intake and control (C) or zilpaterol hydrochloride (Z) treatment.

Item	Treatments							SEM	P-value	P – values Contrasts				
	0	28 A	28 M	56 AZ	56 AC	56 MZ	56 MC			0 v. 28	0 v. 56	28 v. 56	M v. A	C v. Z
<i>n</i>	8	8	8	8	8	8	8							
EBW, kg	543.3 ^c	572.8 ^{bc}	528.9 ^c	615.1 ^a	601.7 ^{ab}	551.6 ^c	540.0 ^c	15.891	<0.01	0.74	0.10	0.09	<0.001	0.36
Ether Extract														
kg	160.7 ^c	181.4 ^{abc}	164.4 ^{bc}	191.1 ^{ab}	205.9 ^a	167.9 ^{bc}	162.3 ^c	12.08	<0.01	0.38	0.09	0.33	<0.01	0.58
% EBW	29.56 ^b	31.75 ^{ab}	31.02 ^b	31.02 ^b	34.14 ^a	30.26 ^b	30.03 ^b	1.421	0.05	0.19	0.14	0.98	0.03	0.12
Moisture														
kg	259.5 ^{bc}	259.6 ^{bc}	242.7 ^c	285.54 ^a	264.31 ^b	257.0 ^{bc}	249.3 ^{bc}	6.02	<0.01	0.33	0.54	0.03	<0.001	<0.01
% EBW	47.78 ^a	45.19 ^{bc}	45.96 ^{abc}	46.46 ^{ab}	43.99 ^c	46.71 ^{ab}	46.21 ^{ab}	1.233	0.04	0.04	0.04	0.69	0.08	0.04
Protein														
kg	88.6	95.3	88.7	100.5	97.0	90.7	92.4	2.99	0.07	0.43	0.09	0.27	<0.01	0.74
% EBW	16.32	16.49	16.78	16.34	16.11	16.49	17.09	0.408	0.49					
Crude Fiber														
kg	0.8	0.6	0.9	0.7	0.9	0.6	1.0	0.40	0.94					
% EBW	0.14	0.11	0.17	0.12	0.15	0.11	0.18	0.069	0.95					
Ash														
kg	30.9 ^c	33.8 ^{abc}	32.3 ^{bc}	35.5 ^a	34.5 ^{ab}	34.4 ^{ab}	35.6 ^a	0.99	0.01	0.11	<0.001	0.02	0.48	0.91
% EBW	0.53 ^d	0.82 ^{bcd}	1.02 ^{ab}	0.67 ^{cd}	0.65 ^d	0.95 ^{abc}	1.18 ^a	0.123	<0.01	0.02	0.02	0.55	<0.001	0.32
Fat Free Empty Body														
EBW fat free, kg	383 ^{bc}	392 ^{bc}	364 ^c	424 ^a	396 ^b	384 ^{bc}	378 ^{bc}	8.5	<0.01	0.75	0.26	0.05	<0.001	0.04
% Moisture	67.83	66.16	66.60	67.34	66.80	66.96	66.05	0.502	0.08	0.02	0.04	0.29	0.50	0.08
% Protein	23.17	24.24	24.33	23.71	24.47	23.65	24.42	0.412	0.29					
% Ash	8.09 ^d	8.59 ^{bcd}	8.86 ^{bc}	8.38 ^{cd}	8.72 ^{bc}	8.98 ^b	9.44 ^a	0.205	<0.01	<0.01	<0.001	0.32	<0.001	0.01

^{a-d} Least squares means within a row with differing superscripts differ ($P \leq 0.05$)

Table 6.2. Chemical analysis of the carcass of beef steers fed to 0, 28 or 56 days on feed and given maintenance (M) or *ad libitum* (A) intake and control (C) or zilpaterol hydrochloride (Z) treatment.

Item	Treatments							SEM	P-value	P – values Contrasts				
	0	28 A	28 M	56 AZ	56 AC	56 MZ	56 MC			0 v. 28	0 v. 56	28 v. 56	M v. A	C v. Z
<i>n</i>	8	8	8	8	8	8	8							
Cold Carcass, kg	347.4 ^c	370.1 ^{abc}	339.8 ^c	402.4 ^a	387.2 ^{ab}	365.3 ^{bc}	348.9 ^c	10.39	<0.001	0.64	0.05	0.05	<0.001	0.10
Ether extract														
kg	100.7 ^c	116.5 ^{abc}	105.4 ^{bc}	124.2 ^{ab}	133.0 ^a	111.4 ^{bc}	103.3 ^c	8.07	0.01	0.32	0.05	0.26	<0.01	0.95
% Carcass	28.98 ^b	31.68 ^{ab}	30.93 ^b	30.78 ^b	34.23 ^a	30.30 ^b	29.63 ^b	1.477	0.04	0.09	0.07	0.94	0.04	0.21
Moisture														
kg	166.7 ^{bc}	164.2 ^{bc}	153.9 ^c	186.4 ^a	167.5 ^b	169.5 ^b	169.0 ^{bc}	4.60	<0.001	0.20	0.43	<0.01	<0.01	<0.001
% Carcass	48.00 ^a	44.14 ^{bc}	45.35 ^{bc}	46.36 ^{ab}	43.39 ^c	46.53 ^{ab}	45.91 ^{ab}	1.239	<0.01	<0.01	0.01	0.29	0.07	0.04
Protein														
kg	55.7	62.8	57.5	65.2	63.0	58.6	60.6	2.19	0.08	0.16	0.04	0.43	0.02	0.96
% Carcass	16.03	16.89	16.92	16.21	16.26	16.09	17.31	0.476	0.26					
Ash														
kg	21.5 ^c	23.5 ^{abc}	22.3 ^{bc}	24.7 ^a	24.0 ^{ab}	23.9 ^{ab}	24.7 ^a	0.79	0.04	0.17	<0.01	0.05	0.43	0.99
% Carcass	6.18 ^b	6.34 ^b	6.57 ^b	6.16 ^b	6.19 ^b	6.56 ^b	7.08 ^a	0.156	<0.01	0.21	0.10	0.78	<0.01	0.07

^{a-c} Least squares means within a row with differing superscripts differ ($P \leq 0.05$)

Table 6.3. Chemical analysis of the internal cavity of beef steers fed to 0, 28 or 56 days on feed and given maintenance (M) or *ad libitum* (A) intake and control (C) or zilpaterol hydrochloride (Z) treatment.

Item	Treatments							SEM	P-value	P – values Contrasts				
	0	28 A	28 M	56 AZ	56 AC	56 MZ	56 MC			0 v. 28	0 v. 56	28 v. 56	M v. A	C v. Z
<i>n</i>	8	8	8	8	8	8	8							
Internal Cavity, kg	85.2 ^{bc}	92.5 ^{ab}	81.1 ^c	91.9 ^{ab}	96.8 ^a	77.6 ^c	79.5 ^c	4.73	<0.001	0.70	0.73	0.90	<0.01	0.23
Ether extract														
kg	42.0 ^{bc}	46.5 ^{ab}	41.1 ^{bc}	46.0 ^{ab}	51.7 ^a	37.8 ^c	40.0 ^{bc}	3.67	<0.01	0.59	0.52	0.96	<0.001	0.07
% Internal Cavity	49.13	49.83	50.52	50.04	53.23	48.46	50.20	1.741	0.15					
Moisture														
kg	34.0 ^{ab}	36.9 ^a	31.6 ^b	37.0 ^a	36.8 ^a	31.8 ^b	31.3 ^b	1.18	<0.001	0.78	0.88	0.83	<0.01	0.73
% Internal Cavity	40.03	40.54	39.08	40.20	38.08	41.25	39.49	1.522	0.46					
Protein														
kg	7.3	7.6	7.0	7.1	6.7	6.5	6.4	0.48	0.39					
% Internal Cavity	8.67	8.25	8.70	7.69	6.97	8.37	8.08	0.841	0.07	0.72	0.07	0.07	0.03	0.21
Crude Fiber														
kg	0.7	0.6	0.8	0.5	0.7	0.4	0.9	0.37	0.75					
% Internal Cavity	0.77	0.59	0.92	0.51	0.72	0.50	1.17	0.415	0.64					
Ash														
kg	0.5 ^c	0.7 ^{ab}	0.8 ^{ab}	0.6 ^{bc}	0.6 ^{bc}	0.7 ^{ab}	0.9 ^a	0.097	0.03	0.01	0.03	0.37	0.04	0.26
% Internal Cavity	0.53 ^d	0.82 ^{bcd}	1.02 ^{ab}	0.67 ^{cd}	0.65 ^d	0.95 ^{abc}	1.18 ^a	0.122	<0.01	0.02	0.01	0.55	<0.001	0.32

^{a-d} Least squares means within a row with differing superscripts differ ($P \leq 0.05$)

Table 6.4. Energy of tissue components of beef steers fed to 0, 28 or 56 days on feed and given maintenance (M) or *ad libitum* (A) intake and control (C) or zilpaterol hydrochloride (Z) treatment.

Item	Treatments							SEM	P-value	P – values Contrasts				
	0	28 A	28 M	56 AZ	56 AC	56 MZ	56 MC			0 v. 28	0 v. 56	28 v. 56	M v. A	C v. Z
<i>n</i>	8	8	8	8	8	8	8							
Energy Empty Body														
Mcal Total	2009.2 ^b	2241.0 ^{ab}	2024.0 ^b	2327.8 ^a	2457.2 ^a	2070.7 ^b	2023.2 ^b	118.21	<0.01	0.37	0.09	0.34	<0.001	0.63
kcal/kg	3696.5 ^b	3935.4 ^{ab}	3820.5 ^b	3781.1 ^b	4076.7 ^a	3740.5 ^b	3743.4 ^b	114.38	0.01	0.06	0.11	0.53	0.01	0.05
Energy Carcass														
Mcal	1268.8 ^c	1456.2 ^{abc}	1307.2 ^c	1514.1 ^{ab}	1590.4 ^a	1371.9 ^{bc}	1303.9 ^c	77.53	0.01	0.26	0.05	0.34	<0.01	0.95
kcal/kg	3651.6 ^c	3955.6 ^{ab}	3840.0 ^{abc}	3757.2 ^{bc}	4095.9 ^a	3740.3 ^{bc}	3735.1 ^{bc}	135.13	0.02	0.04	0.08	0.42	0.04	0.08
Energy Internal Cavity														
Mcal	443.6 ^{bc}	489.2 ^{ab}	427.5 ^{bc}	478.1 ^{ab}	530.8 ^a	398.8 ^c	415.3 ^c	34.36	<0.01	0.64	0.66	0.90	<0.001	0.10
kcal/kg	5188.9	5249.9	5259.5	5204.2	5474.6	5121.8	5214.2	133.29	0.35					

^{a-c} Least squares means within a row with differing superscripts differ ($P \leq 0.05$)

Table 6.5. Empty body nutrient accretion or loss of beef steers fed to 0, 28 or 56 days on feed and given maintenance (M) or *ad libitum* (A) intake and control (C) or zilpaterol hydrochloride (Z) treatment..

Item	Treatments						SEM	P-value	P – values Contrasts		
	28 A	28 M	56 AZ	56 AC	56 MZ	56 MC			28 v. 56	M v. A	C v. Z
<i>n</i>	8	8	8	8	8	8					
Nutrient Accretion and loss (g/d)											
Ether extract	895 ^a	247 ^{bc}	530 ^{ab}	833 ^a	122 ^c	29 ^c	134.6	<0.001	0.13	<0.001	0.43
Moisture	81 ^{ab}	-414 ^c	441 ^a	113 ^{ab}	-55 ^b	-179 ^{bc}	163.7	<0.01	0.04	<0.001	0.06
Protein	259 ^a	67 ^{bc}	204 ^{ab}	162 ^{abc}	35 ^c	69 ^{bc}	57.2	0.02	0.31	<0.001	0.94
Ash	108	70	160	138	121	168	32.2	0.14			
Energy (Mcal/d)	10.145 ^a	1.965 ^c	5.529 ^b	8.325 ^{ab}	1.022 ^c	0.276 ^c	1.1648	<0.001	0.03	<0.001	0.38
Energy (Mcal/kg EBW ^{0.75}) ¹											
ME Intake	0.279 ^a	0.125 ^c	0.257 ^{ab}	0.255 ^b	0.126 ^c	0.125 ^c	0.009	<0.001	0.10	<0.001	0.83
Retained Energy	0.087 ^a	0.017 ^c	0.046 ^b	0.071 ^{ab}	0.008 ^c	0.002 ^c	0.010	<0.001	0.03	<0.001	0.36
kcal from fat accretion ²	70.33 ^a	19.56 ^{bc}	40.02 ^{ab}	63.72 ^a	8.81 ^c	2.34 ^c	9.997	0.001	0.08	<0.001	0.39
kcal from protein accretion ³	9.72 ^a	2.54 ^{bc}	7.36 ^{ab}	5.89 ^{abc}	1.35 ^c	2.56 ^{bc}	2.14	0.03	0.28	<0.01	0.95
Heat Production	0.193 ^{ab}	0.108 ^c	0.211 ^a	0.184 ^b	0.118 ^c	0.123 ^c	0.012	<0.001	0.30	<0.001	0.25

^{a-c} Least squares means within a row with differing superscripts differ ($P \leq 0.05$)

¹ mid-point empty body weight used with initial EBW derived from d0 harvested steers ratio of EBW / BW for each block

² kcal / g estimated at 9.02 kcal/g, FAO

³ kcal / g estimated at 4.27 kcal/g, FAO

Table 6.6 Prediction equations for carcass factors derived from live performance factors

Item	Estimating Equation	P - value	RMSE	Adj R ²
<i>Carcass Factor Prediction using DOF, Diet, Treatment and Live Performance Factors</i>				
HCW	$Y = 10.475 + (13.184*TRT) + (0.571*BW) - (11.233*ADG) + (3.404*DMI)$	<0.001	6.800	0.96
FT	$Y = -3.252 + (0.206*DOF) + (1.140*DIET) + (0.006*BW) - (0.542*ADG)$	<0.001	0.502	0.45
REA	$Y = 53.745 + (0.057*BW)$	<0.01	6.418	0.18
MARB	$Y = 392.290 + (6.027*DMI)$	0.04	54.006	0.07
YG	$Y = -3.964 + (0.317*DOF) + (1.700*DIET) + (0.008*BW) - (0.929*ADG)$	<0.001	0.684	0.44

TRT (C=0, Z=1); DIET (M=1, A=2); DOF (0=0, 28=1, 56=2); BW = final BW; DMI = dry matter intake; MEI = daily metabolizable energy intake; ADG = average daily gain; GF = gain:feed; BWG = BW gain; LM area = LMA; adjusted 12th rib s.c. fat depth (FT); % kidney-pelvic-heart fat (KPH); marbling (MARB);

Table 6.7. Prediction equations for empty body weight (EBW) and empty body fat (EBF), protein (EBP), moisture (EBM) and ash (EBA) derived from live and grading performance factors

Item	Estimating Equation	P - value	RMSE	Adj R ²
<i>Empty Body Weight and Composition Prediction using DOF, Diet, Treatment and Live Performance Factors Only</i>				
EBW	$Y = 24.740 + (6.906*TRT) + (0.829*BW) + (5.755*DMI) - (17.738*ADG)$	<0.001	5.968	0.99
EBF	$Y = 12.737 - (1.829*TRT) + (0.0313*BW)$	<0.001	2.530	0.31
EBM	$Y = 62.457 + (1.737*TRT) - (0.029*BW) + (0.012*BWG)$	<0.001	1.906	0.32
EBP	$Y = 16.931 - (3.803*DMI) + (1.143*MEI)$	0.04	0.952	0.09
EBA	$Y = 7.063 - (0.159*DMI) + (1.808*GF)$	<0.001	0.323	0.46
<i>Empty Body Weight and Composition Prediction using USA Carcass Grading Factors only</i>				
EBW	$Y = 40.752 + (1.392*HCW) - (0.235*LMA) + (6.972*KPH)$	<0.001	8.414	0.97
EBF	$Y = 21.147 + (0.029*HCW) + (1.663*FT) - (0.010*LMA) + (2.193*KPH)$	<0.001	2.015	0.54
EBM	$Y = 53.094 - (0.025*HCW) - (0.958*FT) + (0.077*LMA) - (1.155*KPH)$	<0.001	1.881	0.35
EBP	$Y = 19.382 - (0.417*FT) - (0.967*KPH)$	<0.001	0.814	0.32
EBA	$Y = 8.684 - (0.003*HCW) - (0.149*FT) - (0.239*KPH) - (0.002*MARB)$	<0.001	0.372	0.30
<i>Empty Body Weight and Composition Prediction using DOF, Diet, Treatment and USA Carcass Grading Factors only</i>				
EBW	$Y = 40.628 + (5.150*DIET) - (11.065*TRT) + (1.425*HCW) - (0.267*LMA)$	<0.001	6.781	0.99
	$Y = 14.788 - (1.641*DIET) - (1.964*TRT) + (0.041*HCW) + (1.962*FT) - (0.094*LMA) + (1.957*KPH) + (0.010*MARB)$	<0.001	1.790	0.63
EBF				
EBM	$Y = 56.867 + (1.616*DIET) + (1.922*TRT) - (0.041*HCW) - (1.264*FT) + (0.072*LMA) - (1.020*KPH)$	<0.001	1.651	0.50
EBP	$Y = 19.349 + (0.300*DOF) - (0.513*TRT) - (0.428*FT) - (1.075*KPH)$	<0.001	0.797	0.35
EBA	$Y = 8.809 + (0.268*DOF) - (0.171*DIET) - (0.004*HCW) - (0.165*FT) - (0.162*KPH) - (0.002*MARB)$	<0.001	0.291	0.57

TRT (C=0, Z=1); DIET (M=1, A=2); DOF (0=0, 28=1, 56=2); BW = final BW; DMI = dry matter intake; ADG = average daily gain; GF = gain:feed; BWG = BW gain;

LM area = LMA; adjusted 12th rib s.c. fat depth (FT); % kidney-pelvic-heart fat (KPH); marbling (MARB); MEI = daily metabolizable energy intake

Appendix A:

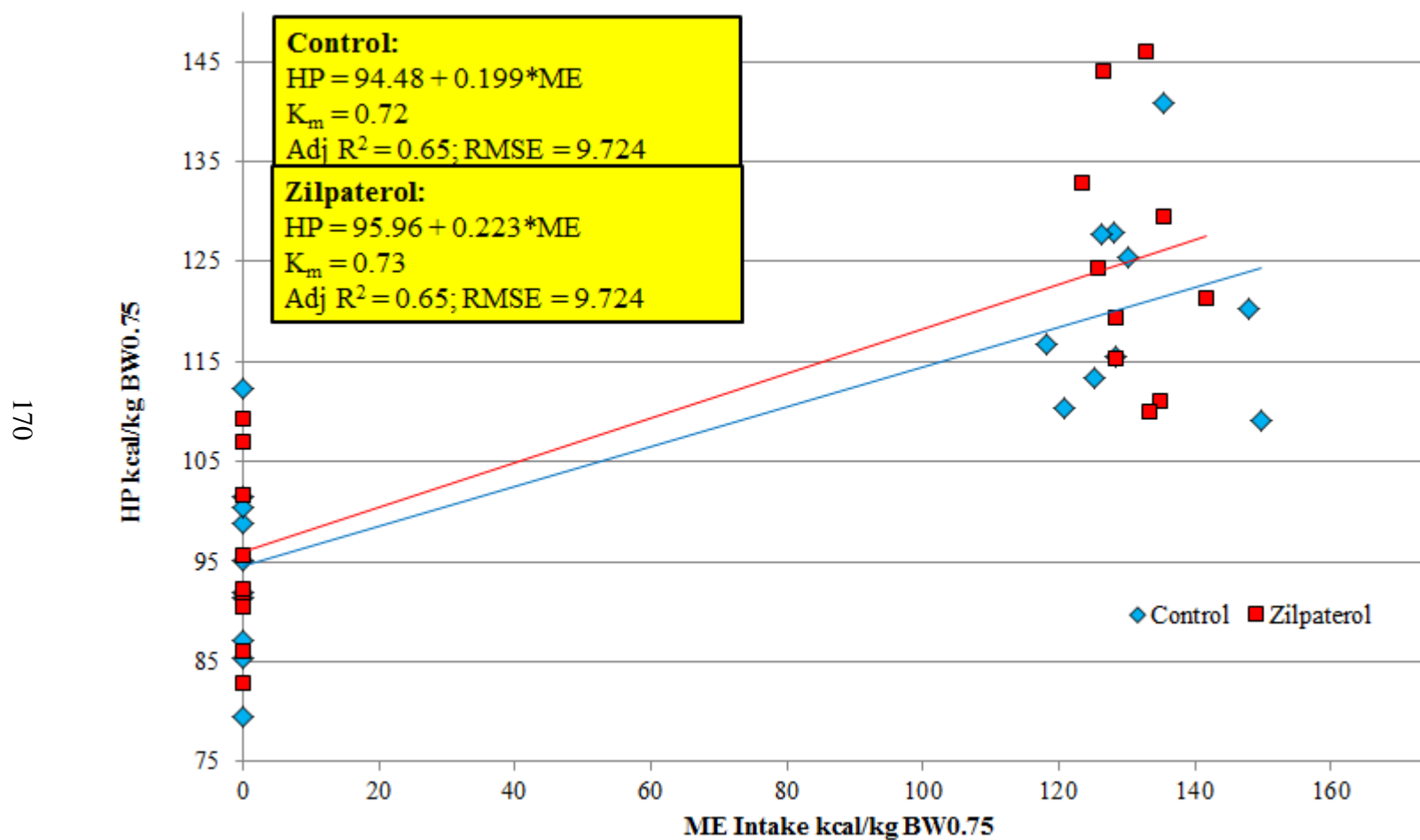


Figure A.1 Effect of zilpaterol hydrochloride on the regression of heat production on ME Intake and efficiency of maintenance measured using indirect calorimetry

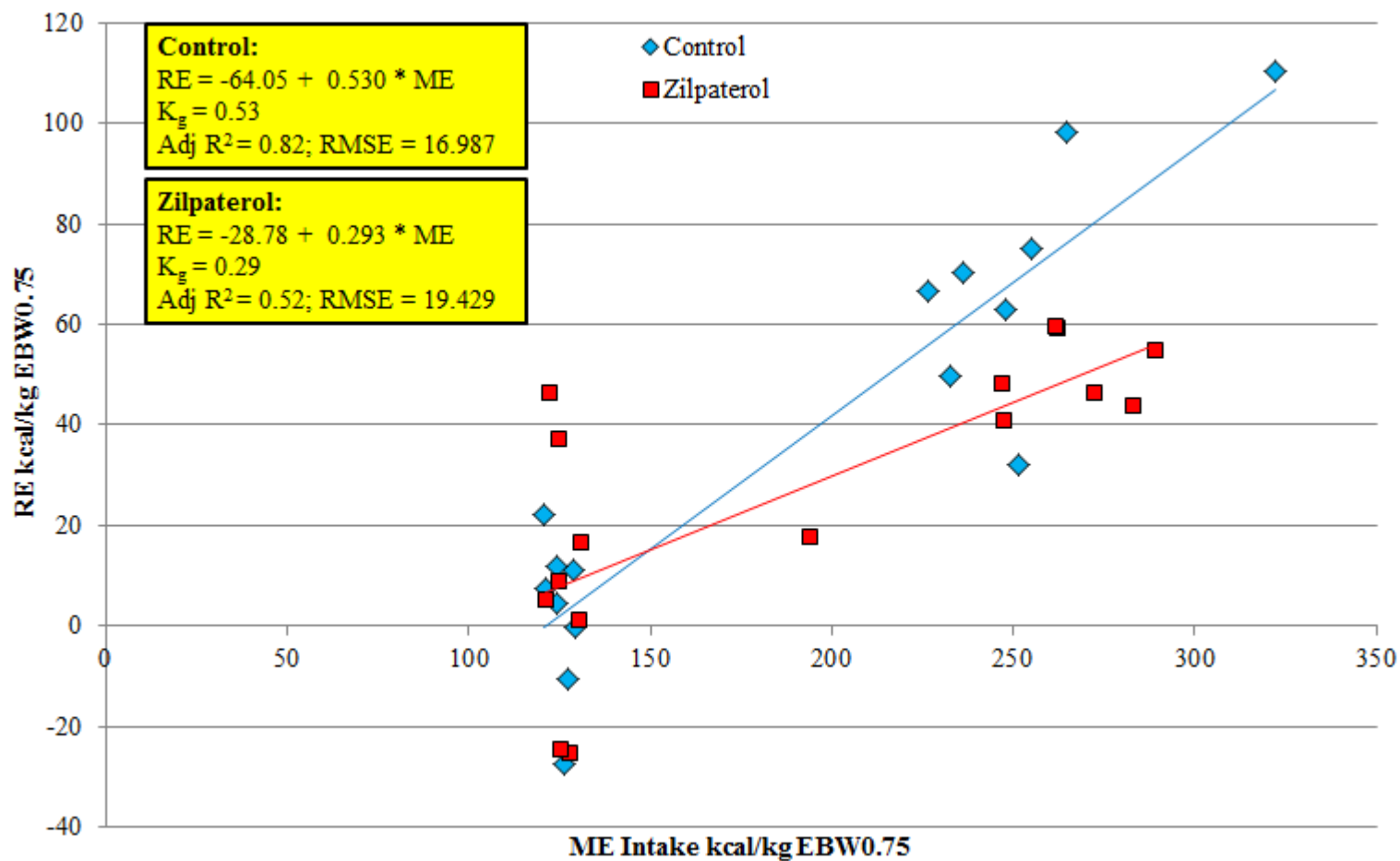


Figure A.2 Effect of zilpaterol hydrochloride on the regression of retained energy on ME Intake and efficiency of gain measured using comparative slaughter

Figure A.3 Effect of zilpaterol hydrochloride on the regression of heat production on ME Intake and fasting heat production measured using comparative slaughter