# THE EFFECTS OF ZILPATEROL HYDROCHLORIDE ON CARCASS CUTABILITY, TENDERNESS, AND COMPOSITION OF BEEF

By

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# THE EFFECTS OF ZILPATEROL HYDROCHLORIDE ON CARCASS CUTABILITY, TENDERNESS, AND COMPOSITION OF BEEF STEERS

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# CHAPTER I

## INTRODUCTION

Protein-energy malnutrition is listed as the most lethal form of malnutrition/hunger, and as of 2006, the United Nations Food and Agriculture Organization (FAO) estimated that 854 million (12.6% of the world population) were considered undernourished and protein deficient. Protein plays an essential role in maintenance of critical body functions and provides key amino acids for muscle function and growth. Meanwhile, beef is a highly nutritious food and is considered to be an excellent source of protein, iron, zinc, many of the B vitamins, and choline. In 1997, David Pimentel, professor of ecology at Cornell University's College of Agriculture and Life Science reported that beef cattle consume 54 energy units for every 1 protein unit output. Chickens, conversely, have a 4:1 energy to protein output ratio, indicating that beef production could benefit from improvements in efficiency in order to rival other protein sources.

Research concerning the improvement of beef efficiency has been ongoing in the U.S. for a number of years. Many new technologies have been developed to aid in the improvement of cattle feeding and beef production. One such technology new to the U.S. beef industry is beta-agonists. The first beta-agonist to be approved for use in the U.S. beef cattle industry was ractopamine hydrochloride sold under the trade name Optaflexx® (Elanco Animal Health, Greenfield, IN). Recently, zilpaterol hydrochloride,

sold under the trade name Zilmax® (Intervet/Schering Plough, Millsboro, DE) has been approved for use in the U.S. beef cattle industry as well. These beta-agonists are marketed very similarly as compounds that will improve body mass gain and feed conversion during the finishing phase, as well as improve the lean to fat ratio in carcasses and carcass cutability. The utilization of these compounds can decrease the amount of inputs needed during the finishing phase, as well as increase the total amount of lean per carcass for wholesale beef retailers. By using these compounds, the U.S. beef industry can increase the total amount of protein produced from beef while decreasing input levels. These new technologies could potentially allow the U.S. beef industry to make greater strides towards supplying a quality protein source throughout the world.

However, beta-agonists have been shown to have a negative effect on tenderness. While improvements in lean carcass mass are positive, the negative impact on tenderness may lead to some hesitation in adoption of these new feed additives by portions of the beef industry. The potential negative impact these compounds could have on consumer satisfaction has left some beef retailers concerned about losing customers. However, in recent years, the beef industry has begun to embrace the practice of enhancement, which the pork and poultry industries have been utilizing for years. Enhancement revolves around adding water, salt, and phosphates to meat, allowing for increased moisture retention. In addition to improvements in water holding capacity, both objective and subjective tenderness measurements are improved. Thus, the objectives of these studies were: (1) to determine the extent to which zilpaterol hydrochloride can improve the carcass cutability of beef, while determining which primals within the carcass are affected the most, (2) to determine the impact of zilpaterol hydrochloride, as well as

enhancement, on overall beef tenderness in steaks from the strip loin, top round, and bottom round, and (3) assess the impact of zilpaterol hydrochloride and enhancement on beef tenderness.

## CHAPTER II

## **REVIEW OF LITERATURE**

#### **INTRODUCTION**

The use of beta-adrenergic agonists ( $\beta$ -AA) to improve feed efficiency and enhance carcass composition in livestock species has been well documented since the early 1980's. Many  $\beta$ -AA act as repartitioning agents and have been shown to enhance lean meat production in many animal species. Beta-agonists have structures similar to that of the catecholamines, epinephrine, a hormone, and norepinephrine, a neurotransmitter. The  $\beta$ -AA act as analogues of these hormones which are responsible for regulating smooth muscle contraction, blood pressure, cardiac rate, lipolysis, and glycogenolysis (Mersmann, 1989). Thus,  $\beta$ -AA were initially used in human health for treating ashmatics, muscle atrophy, and obesity. The use of  $\beta$ -AA to address human health issues led to the investigation of  $\beta$ -AA in livestock species to improve carcass composition. Development of different types of  $\beta$ -AA led to the production of ractopamine hydrochloride (RAC), which is the first form of a  $\beta$ -AA to be approved by the FDA for use in livestock species. Along with RAC, other forms of  $\beta$ -AA have been developed and studied for use in livestock production. One such compound that was recently approved for use in the U.S. by the FDA is Zilpaterol hydrochloride (ZH).

Ractopamine hydrochloride (Optaflexx®, Elanco Animal Health, Greenfield, IN) is a phenethanolamine with beta-adrenergic properties that has been shown to increase

lean meat production in many animal species. The U.S. Food and Drug Administration approved the use of RAC in cattle finishing diets in June 2003. Its use has been shown to improve carcass lean growth and the efficiency of lean deposition in feedlot cattle through reductions in trimmable fat and increases in carcass muscling. Feeding steers RAC has consistently shown to increase average daily gain, gain:feed, carcass weight, and ribeye area. Ractopamine hydrochloride feeding appears to have its greatest impact on carcass muscling. However, feeding heifers RAC has yielded variable results. Similar responses to those of steers have been reported (Quinn et al., 2008); however, minimal response has also been noted (Schroeder et al., 2003a).

Zilpaterol hydrochloride (Zilmax®, Intervet/Schering Plough Animal Health, Millsboro, DE) was approved in South Africa in 1997 and Mexico in 1999; however, it was not until 2006 that ZH became approved for use in beef cattle in the U.S. and recently, in 2009, it became approved for use in Canada. Initial studies have indicated that ZH has a similar effect to RAC when fed as a supplement in finishing diets for beef cattle. However, unlike RAC, ZH requires a 3 day withdrawal period prior to harvest where RAC has no withdrawal time. Much like RAC, ZH fed animals exhibit increased efficiency during the finishing phase as well as increased carcass lean. Additionally, as seen with most  $\beta$ -AA, tenderness can be negatively impacted due to the feeding of these compounds.

## β-Adrenergic Agonist Mechanism

Beta-adrenergic agonists are synthetic compounds that are similar in structure to the natural occurring catecholamines, epinephrine, and norepinephrine. Research with norepinephrine and epinephrine, as well as other compounds, led to the identification and

nomenclature of two  $\beta$ -AA receptors known as  $\alpha$ -adrenergic receptors ( $\alpha$ AR) and  $\beta$ adrenergic receptors ( $\beta$ AR) in the late 1940's (Mersmann, 1989). These receptors were found to be over 400 amino acids in length and comprise a part of the seven-membrane receptor family. Their nomenclature is indicative of the family of receptors having seven relatively hydrophobic transmembrane domains that anchor the receptor to the plasma membrane. Moreover, the receptor contains two extracellular recognition sites which allow for recognition of the correct ligand as well as the appropriate G-protein (Mersmann, 1998; Garret and Grisham, 1999). Each of the different receptor sites is associated with a specific G-protein which allows for appropriate signaling for activity  $(G_s \text{ and } G_i)$  (Northup, 1985). The G stimulator protein is responsible for stimulating the production of adenylate cyclase upon activation, however once the stimulus has been sent, the G stimulatory protein becomes inactive  $(G_i)$  and adenylate cyclase production is inhibited. While the  $\alpha AR$  and  $\beta AR$  are stimulated by beta adrenergic agonists, the natural hormones norephinephrine and epinephrine can stimulate the receptors to increase activity however epinephrine has a greater affinity for binding to  $\alpha AR$  when compared to norepinephrine. Most all tissue within mammalian cells have a form of beta-agonist receptor however,  $\alpha AR$  are most predominantly associated with gut contractions and cerebral, skin, and salivary gland arterioles while  $\beta AR$  are associated with heart rate, bronchiodilation, and stimulation of lipolysis (Mersmann, 1989).

Further classification of the beta form of  $\beta$ -AA came nearly twenty years after the initial classification of  $\beta$ -AA. Lands et al. (1967) further classified the  $\beta$ -receptors into  $\beta_1$ -AR and  $\beta_2$ -AR. Numerous years later, Emorine et al. (1992) classified a third form of the receptor as  $\beta_3$ -AR. While little is known about this newly classified form of the  $\beta$ -

receptor, it may play an important role in mediating catecholamine action and it is also thought to be more sensitive to  $\beta$ -AA and may be less prone to agonist-induced desensitization when compared to the other two forms of  $\beta$ -AR (Ding et al., 2000).

While most mammalian tissues contain beta receptors, the distribution of these receptors is varied among different tissues. Cardiac tissue within the mouse contains mostly  $\beta_1$  receptors while lung and muscle tissues contain mostly  $\beta_2$  receptors (Mills, 2002). The  $\beta_3$  receptors have a limited expression pattern and are found predominantly in adipose tissue (Mills, 2002). Additionally, McNeel and Mersmann (1999) documented that porcine adipocytes are comprised of 70%  $\beta_1$ , 20%  $\beta_2$ , and 10%  $\beta_3$  – receptors. Also, in porcine muscle tissue,  $\beta_1$ -receptors make up 60% of the expressed receptors with  $\beta_2$ making up 39% and  $\beta_3$  comprising < 1% of the total receptors (McNeel and Mersmann, 1999). While the distribution of receptor types varies among tissues, each receptor type also differs in regulation as well. Each receptor has a specific G-protein, unique phosphorylation activity, and gene expression level (Strosberg, 1996). This variation in regulation and expression level can explain why varying results are seen with  $\beta$ -AA administration across different species. Mills (2002) concluded that the varying response seen to a specific ligand is due to the multiple subunits found within the specific Gprotein associated with each receptor and can explain the reasoning behind the varying responses seen between different tissues as well as between species. Mills (2002) also reported that the sequence homology for the various receptors is high across species; however, the ligand's ability to bind to a given receptor and to signal through a G-protein signaling pathway is highly dependent upon the amino acid sequence of the receptor

which can vary greatly across species. Therefore, results due to  $\beta$ -AA administration can vary greatly across species and can be hard to predict using different species as a model.

The primary signaling pathway is stimulated upon the binding of the  $\beta$ -AA binding to a  $\beta$ -AR which will activate the specific G-protein associated with that specific receptor. Once the G-protein becomes activated, the  $G_s$  protein stimulates adenylate cyclase production which initiates cyclic adenine 3', 5'- monophosphate (cAMP) production, which can combine with protein kinase A allowing the compound to become active. Once active, protein kinase A phosphorylates and activates proteins such as hormone sensitive lipase and glycogen phosphorylase (Buttery and Dawson, 1987; Mersmann, 1989 and 1998; Murry et al., 1996). Protein kinase A will also phosphorylate glycogen synthase and acetyl CoA carboxylase which inhibits each of these proteins. By activating hormone sensitive lipase and glycogen phosphorylase and deactivating glycogen synthase and acetyl CoA carboxylase, lipolysis and glycogenolysis become stimulated and glycogen and fatty acid biosynthesis are inhibited (Mills et al., 1990; Mersmann, 1998; Garrett and Grisham, 1999). When the  $\beta$ -AR becomes activated by  $\beta$ -AA binding, the receptor becomes phosphorylated and is removed from the cell surface (Ding et al., 2000). As the receptors are exposed to continued levels of  $\beta$ -AA, the level of receptor replacement becomes decreased to where the exposure is greater than receptor replacement and the cell becomes less sensitive to  $\beta$ -AA stimulation and the effects of the  $\beta$ -AA become attenuated (Spurlock et al., 1994).

The most important components to the  $\beta$ -AA pathway are G-proteins. Without the specific G-proteins available to transport the signal to adenylate cyclase the secondary signal cannot become activated (Rodbell, 1980). G-proteins are comprised of three

subunits ( $\alpha$ ,  $\beta$ , and  $\gamma$ ); the  $\alpha$  subunit is needed to bind to GTP to allow for signal transduction. When bound to GTP, the  $\alpha$  subunits disassociates from the G-protein and becomes associated with adenylate cyclase to cause cAMP production. Production of cAMP will continue as long as the  $\alpha$  subunit remains associated with adenylate cyclase; however, due to the  $\alpha$  subunits' intrinsic GTPase activity, GTP rapidly hydrolyses to GDP which leads to the dissociation of the  $\alpha$  subunit from adenylate cyclase and its reassociation with the  $\beta$  and  $\gamma$  dimer, regenerating the heterotrimeric G-protein complex (Garrett and Grisham, 1999).

#### β-Adrenergic Agonist Effects on Lipid and Protein Metabolism

Fat accretion, as well as protein accretion, are due to a balance between metabolism and catabolism of fatty acids and amino acids (Buttery and Dawson, 1987). Protein turnover has been shown to be a very inefficient process and has been estimated to comprise 15 to 20% of growing animal's energy requirements (Young et al., 1975). Beta-adrenergic agonist can significantly alter both lipid and protein metabolism as well as lipid and protein catabolism. For these reasons,  $\beta$ -AAs are being used in the livestock industry to increase the efficiency of lean production while reducing fat accretion within the animal.

Beta- adrenergic agonist increase the levels of cAMP with lipid cell, which leads to an activation of protein kinase A, then activating triacylglycerol lipase. Triacylglycerol lipase is responsible for hydrolyzing fatty acids from triacylglycerol, which eventually results in free fatty acids and glycerol. Fatty acids are then further broken down to fatty acyl-CoA which then can be broken down to form acetyl-CoA, FADH<sub>2</sub>, and NADH. The resulting acetyl-CoA can then enter the TCA cycle where it can be used as a substrate for amino acid biosynthesis. This up-regulation of fatty acid catabolism and down-regulation of fatty acid synthesis can result in a loss or decrease in fatty accretion with animals. Mills and Liu (1990) suggested that lipid biosynthesis is more sensitive to  $\beta$ -AA than lipolysis. Moreover, Mills et al. (1990) suggests that lipid biosynthesis is reduced by as much as 40% in pigs supplemented with the  $\beta$ -AA RAC. However, these suggestions have be contradicted by other studies (Mills et al., 1990; Dunshea, 1993; Lui et al., 1994) where either limited changes or increased lipolysis was seen in pigs supplemented with RAC. The exact effects of  $\beta$ -AA on lipid metabolism is still debated and due to varying effects across species as well as across the various types of  $\beta$ -AA, the mechanism by which  $\beta$ -AA alter lipid metabolism may be different across species as well as across the different  $\beta$ -AA compounds.

While elevated cAMP levels leads to  $\beta$ -AA responses in lipid metabolism, there are some internal inhibitors that may affect the  $\beta$ -AA response in some animals. Cyclic AMP is highly sensitive to intracellular adenosine which can prevent the activation of protein kinase A by cAMP. Without activated protein kinase A, downstream enzymes responsible for lipid metabolism and catabolism cannot become activated and, therefore, the  $\beta$ -AA response is blocked (Mills et al., 1990). Additionally, insulin has been shown to antagonize the  $\beta$ -AA response by decreasing the cells sensitivity to the  $\beta$ -AA (Mills and Liu, 1990). In an additional study, Peterla and Scanes (1990) reported that the decreased lipid biosynthesis effect seen with  $\beta$ -AA supplementation was completely blocked by insulin and the lipolysis effects were significantly decreased by insulin.

While protein turnover has been document to be a very inefficient process, it is a necessary process for maintaining available pools of amino acids, repairing erroneous proteins, and removing proteins not properly incorporated into their appropriate

subcellular locations (Reeds, 1989). The protein accretion response seen with  $\beta$ -AA supplementation is generally regarded as an increase in muscle hypertrophy (Mills, 2002). Moreover, Johnson (2009) reported that type II muscle fibers (white, large, fast twitch) were more sensitive to  $\beta$ -AA treatment when compared to type I fibers (red, small, slow twitch). An increase in muscle fiber hypertrophy is noted by no net change in DNA within the muscle and can indicate a change in the rate of protein synthesis, degradation, or both; however,  $\beta$ -AA are generally thought to have the greatest effect on protein degradation as opposed to synthesis. This is evident by a study by Pringle et al. (1993) where it was report that calpain (cysteine proteinase) activity of lambs supplemented with a  $\beta$ -AA was significantly decreased while calpastatin (calpain inhibitor) activity was increased by 73%. Moreover, Garber et al. (1976) documented a decrease in amino acid released from muscle with increased level of epinephrine, which has similar effects on adenylate cyclase as does  $\beta$ -AA.

Insulin promotes protein synthesis and inhibits protein degradation and  $\beta$ -AA have been shown to activate signaling cascades in common with insulin (Luttrell et al., 1998). Moreover, Gupta et al. (1996) documented a direct link between cAMP and gene regulation for myosin heavy chain cardiomyctes, and Cong et al. (1998) documented a relationship between cAMP and bovine calpastatin. While the exact mechanisms involved in the  $\beta$ -AA manipulation of protein turnover are still debated, it is understood that  $\beta$ -AA increase muscle hypertrophy by having a significant impact on the size of type II muscle fibers. Continued research is needed to fully understand the exact effect  $\beta$ -AA have on muscle turnover.

## β-AA Effects on Live Animal Performance and Carcass Composition

#### Ractopamine hydrocholoride

Ractopamine hydrochloride (RAC) feeding has been shown to improve average daily gain (ADG) and feed to gain ratios (F/G) in beef cattle (Gruber et al., 2007; Winterholler et al., 2007). In additional studies, hot carcass weight (HCW) significantly increases with RAC supplementation (Avendano-Reyes et al., 2006; Winterholler et al., 2007). Moreover, some studies have indicated an increase in dressing percentage (Avendano-Reyes et al., 2006), while others report that RAC supplementation has no impact on dressing percentage (Gruber et al., 2007; Winterholler et al., 2007). In the livestock industry, producers are paid on a value-based pricing system where carcass leanness and fat are the primary contributors to this system. All of the aforementioned improvements play a considerable role in increasing profits for producers. Schroeder et al. (2004) and Winterholler et al. (2007) reported improved dressing percentages and increased ribeye areas in cattle treated with RAC. This increase in dressing percentage, along with the increase seen in live body weights with RAC supplementation, would indicate an increase in edible carcass tissue and not an increase in visceral mass.

Initial studies have indicated that RAC has limited impact on 12<sup>th</sup> rib back fat in beef cattle. Schroeder et al. (2004) reported no effects on 12<sup>th</sup> rib backfat in feedlot steers supplemented with RAC. The same effects were also reported in feedlot heifers (Quinn et al., 2008). Additionally, Schroeder et al. (2004) reported a numerical decrease in carcass fat percent for all levels of RAC fed. Carcass fat percentage was significantly reduced in steers fed 200 mg/hd/d but those fed 300 mg/hd/d only tended to see a reduction in carcass fat percentage (Schroeder et al., 2004). In heifers, carcass fat percent

was numerically decreased across all RAC treatments but was only significantly reduced in those fed 300 mg/hd/d (Schroeder et al., 2004).

Many researchers have reported changes in carcass lean due to the effects of feeding RAC or any other  $\beta$ -AA. In many studies, the loin muscle area of pigs increased with supplementation of RAC (Anderson et al., 1989; Watkins et al., 1990; Yen et al., 1990; Stites et al., 1991, Armstrong et al., 2004; Schroeder et al., 2004). Additionally, Watkins et al. (1990) reported an increase in the muscle score in pigs (Watkins et al., 1990). However, when supplementing finishing cattle with RAC the impacts on carcass muscling are varied. Increased conformation scores were reported by Schroeder et al. (2004) and Gruber et al. (2007) with RAC supplementation; however, another study (Avendano-Reyes et al., 2006) reported no impact on ribeye size.

Weight and yield of trimmed and untrimmed carcass cuts and muscles are another way to express changes in composition due to RAC supplementation. Most studies report that RAC supplementation does not significantly affect the weights of untrimmed cuts in pork carcases (Yen et al., 1990; Stites et al., 1991; Utttaro et al., 1993; Crome et al., 1996). However, Stites et al. (1991) and Crome et al. (1996) reported an increase in the untrimmed weight of the ham and loin. Additionally, Crome et al. (1996) reported an increase in the weight of the untrimmed picnic and Boston butt. In pork carcasses, the weights of the trimmed primal cuts from the ham, loin, belly, Boston butt, and picnic shoulder increased with supplementation of RAC (Aalhus et al., 1990; Yen et al., 1990; Uttaro et al., 1993; Crome et al., 1996). Additionally, Crome et al. (1996) noted an increase in weight of boneless, trimmed retail cuts from the tenderloin, Boston butt,

picnic, ham, and loin due to supplementation of RAC. Furthermore, the ham was dissected and separated into individual muscles. The individual weights of the inside, outside, and knuckle increased with RAC supplementation (Crome et al., 1996). In addition, when trimmed primals were expressed as a percentage of the carcass side weight, Stites et al. (1991) and Crome et al. (1996) noted an increased percentage in the ham and loin.

#### Zilpaterol hydrochloride

Zilpaterol hydrochloride is a  $\beta$ -AA that has been approved in Mexico and South Africa for over 10 years; however, it was not until 2006 that the compound was approved by the FDA for use in feedlot cattle in the United States. Therefore, limited research is available on the effects of ZH in U.S. feedlot systems. However, since the approval of the compound, extensive research has begun to determine the effects of ZH on feedlot performance and its comparison to other  $\beta$ -AA compounds, such as RAC, that are currently being used in U.S. feedlot systems.

Initial studies conducted in Mexico and South America, where ZH is approved, found that ZH had similar effects to those found using RAC in U.S. studies. Much like RAC, ZH is marketed as a compound that will increase rate of gain, improve feed efficiency, and increase carcass leanness in cattle fed in confinement systems. Initial studies by Avendano-Reyes et al. (2006) show an increase in final body weight, increased average daily gain (ADG), and an improvement in gain to feed ratios (G:F) for cattle supplemented with ZH. These results are very similar to results seen when supplementing with RAC; however, most studies show a decrease in feed consumption with RAC inclusion, which is not documented with ZH inclusion (Avendano-Reyes et al.,

2006; Plascencia et al., 1999). Additionally, Avendano-Reyes et al. (2006) documented an increase in HCW, carcass yield, and loin muscle area. This again is very similar to results from numerous studies where RAC was supplemented to finishing cattle. However, a greater increase in loin muscle area has been shown when supplementing ZH compared to RAC supplementation (Avendano-Reyes et al., 2006). Moreover, while the study by Avendano-Reyes et al. (2006) indicated a tendency for ZH fed cattle to have decreased 12<sup>th</sup> rib fat; other studies (Plascencia et al., 1999; Montgomery et al., 2009a) have shown no effect on 12<sup>th</sup> rib back fat with ZH inclusion which is very similar to studies where RAC was used.

More recently, studies conducted in the U.S. have released information regarding the impacts of ZH on fed cattle production and carcass characteristic in the U.S. Initial studies have focused on the most effective length of time for ZH supplementation. Most studies have included ZH into the diet ranging from 20 to 40 d prior to harvest. While many of the earliest research trials focused on fed beef steers, current U.S. studies have included both fed beef steers and calf-fed Holstein steers, as well as beef heifers. Additionally, dairy cows comprise a majority of the cattle within the U.S., and these cows are bred and calved for their milk production abilities. While heifers from these parturitions can be recycled back into the dairy cow herds, bulls are typically castrated and placed into a calf-fed system and finished as beef. The resulting steers from the breeding of dairy cows comprise 8.7% of the harvested cattle in the U.S. each year (Smith et al., 2006). Calf-fed Holstein steers stay on feed for nearly one year prior to harvest, are typically very consistent, and grade with a high proportion of USDA quality grades of Choice or better. However, these steers have poor muscling, low dressing percentages, and reduced fabrication yields when compared to typical beef steers. Therefore, including a  $\beta$ -AA into the finishing protocol for calf-fed Holstein steers as well as typical beef steers could be an advantageous practice.

Early studies have indicated that the greatest benefit from ZH supplementation is seen with 20 d of supplementation as nearly 99% of the increase in final body weight is seen within this time frame (Elam, 2009). This increase in body weight was also reflected by an increase in carcass weight, where 99% of the increase was seen at 20 d of supplementation (Elam, 2009). Moreover, much like previous research, ZH supplementation did not alter dry matter intake, yet ADG was increased with increasing time fed ZH (Elam, 2009). Furthermore, at harvest, cattle supplemented with ZH had increased dressing percentages that responded linearly as d of supplementation increased (Elam, 2009). Cattle supplemented with ZH tended to show linear decreases in backfat as ZH supplementation time increased (Elam, 2009). While kidney, pelvic, and heart fat (KPH) percentages decreased with ZH supplementation, there was not a linear decease seen as supplementation time increased like that observed with the decrease in backfat (Elam, 2009). In addition to changes in fat within the carcass, a linear increase in ribeye area was seen with increasing d of ZH (Elam, 2009). These decreases in backfat and percentage KPH, as well as the increase in ribeye area, led to a linear decrease in calculated USDA yield grade as d of supplementation increased (Elam, 2009). This resulted in a linear increase in the percentage of carcasses that were classified as Yield Grades 1 and 2 and a linear decrease in those categorized as Yield Grades 4 and 5 as d of ZH supplementation increased (Elam, 2009). It was concluded by Elam (2009) that ZH

increased the overall production and efficiency of production of beef cattle as well as that supplementation for 20 d is sufficient to achieve the desired responses.

Similar to RAC, initial studies where red meat yields were determined in ZH supplemented beef cattle indicated that ZH supplementation can significantly increase carcass cutability. In studies that involved both beef and calf-fed Holstein steers, red meat yield was increased as much as 1.7% in beef steers and 1.0% in calf-fed Holstein steers with ZH supplementation (Hilton, 2009). Moreover, when expressed as a percentage of cold side weight, ZH supplemented carcasses had increased yields of several high value cuts such as the strip loin, tenderloin, top inside round, eye of round, bottom sirloin tri-tip, and top sirloin butt (Hilton, 2009). Additionally, Hilton (2009) reported that ZH increased the percent yield in USDA Yield Grade 2 carcasses by 1.5% and reported that this improvement in yield is not accurately depicted by the current USDA Yield Grade system. Finally, this increase in yield correlates to improved financial returns for red meat wholesalers. A 1.5% improved red meat yield with an average wholesale red meat price of \$2.00/lb can result in an improvement of \$3.00/cwt (Hilton, 2009).

#### **β- AA Effects on Carcass Quality**

#### *Ractopamine hydrocholoride*

Meat color is an important characteristic and is the single most important quality factor as it greatly affects point of purchase for the consumer. Color in fresh meat is typically measured using colorimeters or by using subjective color standards adopted by the meat industry. Colorimeters use three values, L\*, a\*, and b\*, which measure light reflectance. Subjective color can also be determined by using trained personnel to

evaluate the color of fresh beef on a predetermined scale. Quinn et al. (2008) reported no significant changes in L\* values, a\* values, or b\* values between control cattle and cattle fed 200 mg/d of RAC. In a study by Schroeder et al. (2004), a scale of 1 to 7 with 1 being least acceptable was used to determine fresh beef color. Schroeder et al. (2004) reported no change in muscle color of RAC-treated steers (0, 100, 200, 300 mg/hd/day) using the subjective color score scale. However, slight changes were reported in treated heifers (0, 100, 200, 300 mg/hd/day) as a darker color score was observed with all levels of RAC supplementation when compared to controls (Schroeder et al., 2004). In an additional study by Gonzalez et al. (2009), it was documented that RAC had no impact on subjective or objective color parameters of six muscles from the beef carcass. Finally, Avendano-Reyes et al. (2006) reported that hue angle ( $h^{\circ}$ ) was increased by RAC supplementaion as steaks were more red as compared to contol steaks as well as tended to have increased b\* values. Furthermore, Avendano-Reyes et al. (2006) reported that RAC supplementation had no impact on chroma (C\*) or a\* values; however, L\* values for control steaks indicated that they were darker at 5 d of retail display when compared to RAC supplemented steaks yet were similar at 1 d and 14 d or retail display.

Intramuscular fat (IMF) or marbling is associated with the eating quality of meat by many people. The amount of IMF can be measured subjectively, by using marbling score standards for cattle developed by the United States Department of Agriculture (USDA), or objectively by chemical analysis of the muscle tissue. Watkins et al. (1990) reported a significant increase in marbling scores in pigs treated with RAC. Schroeder et al. (2004) reported no differences in marbling score or quality grade between control and heifers and steers supplemented with RAC. There were also no differences in IMF

observed across the different levels of RAC supplementation (Schroeder et al., 2004). Additionally, in studies by Winterholler et al. (2007) and Quinn et al. (2008), it was documented that RAC supplementation had no impact on marbling score or the percentage of carcasses qualifying for USDA Choice or greater, USDA Select or USDA Standard. Collectively, RAC supplementation has not been shown to considerably affect IMF in beef.

Tenderness is a quality factor of great concern to most consumers as it relates to palatability. "The increased rate of protein synthesis and lipolysis in pigs fed RAC has resulted in some concern about the toughness of meat that is produced," (Uttaro et al. 1993). Tenderness is measured by both subjective sensory panels and objectively by Warner-Bratzler shear force (WBS) values.

Schroeder et al. (2004) reported all WBS values from strip loin steaks from each RAC treatment group (0, 100, 200, 300 mg/hd/d) were considered acceptably tender and were well within the ranges found in today's beef carcasses, although WBS force values for the 300 mg/hd/day group were significantly higher than that of the control. Shroeder et al. (2004) measured sensory panel tenderness values on a scale from 0 (not tender) to 150 (very tender) and no differences were reported for groups fed 0 mg/hd/d, 100 mg/hd/d and 200 mg/hd/d of RAC. However, the treatment group receiving 300 mg/hd/d RAC was rated significantly less tender than the other treatment groups (Schroeder et al., 2004). Avendano-Reyes et al. (2006) also reported a decrease in tenderness of loin muscle steaks from RAC supplemented animals. However, Quinn et al. (2008) documented that RAC supplementation had no impact on WBS values from strip loin steaks aged for 14 d when compared to steaks from control animals. Tenderness values

for beef, as evaluated by both WBS force and sensory panels, have been shown to decrease at higher concentrations of RAC supplementation (Gruber et al., 2007). These slight decreases in tenderness could be due to an increase in muscle protein synthesis resulting in muscle hypertrophy and larger muscle fibers.

#### Zilpaterol hydrochloride

While little data is available regarding the effects of  $\beta$ -AA on shelf life and meat color, some studies have documented a tendency for  $\beta$ -AA supplementation to produce paler meat (Geesink et al., 1993; Vestergaard et al., 1995). While the study by Avendano-Reyes et al. (2006) indicated little effect of ZH on muscle color or stability, ZH did decrease the time it took for loin steaks to darken. In an additional study by Strydom et al. (2000), ZH supplementation was shown to significantly increase the color stability of loin and rump steaks, as well as ground beef. In a more recent study, Hunt (2009) reported that feeding ZH for up to 40 d had no impact on pH, initial color or appearance, or display color stability of semimembranosus, gluteus medius, and longissimus steaks, as well as ground beef.

Much like RAC, ZH has been shown to significantly increase WBS values of loin muscle steaks. While this is a common finding among animals supplemented with various  $\beta$ -AA, the reported values still range from severe to mild increases in WBS values. As reported by Brooks and Miller (2009), longissimus steaks from both USDA Choice and Select carcass were tougher as the duration of ZH feeding increased from 0 to 40 d. Moreover, WBS force values for gluteus medius and triceps brachii were increased with ZH supplementation in both USDA Choice and Select carcasses (Brooks and Miller, 2009). Furthermore, Brooks and Miller (2009) reported that longissimus steaks from

calf-fed Holstein steers had increased WBS force values when supplemented with ZH at 7, 14, and 21 d of postmortem aging.

Sensory analysis has indicated similar results to those seen using WBS for tenderness. Trained panelists indicated a decrease in juiciness, tenderness, beef flavor intensity, and beef flavor when comparing steaks from ZH supplemented beef steers to steaks from control beef steers (Hilton et al., 2009). The initial and sustained tenderness scores were reduced by approximately 6 to 7% with ZH treatment; however, ZH supplementation did not affect sensory panel off-flavor scores (Hilton et al., 2009). In an additional study, Brooks and Miller (2009) reported a decrease in consumer scores for tenderness, juiciness, flavor and overall acceptability when comparing USDA Choice steaks from ZH fed steers to USDA Choice steaks from control steers both aged for 14 d. Additionally, these findings were the same when consumers were asked to make the same comparison using USDA Select steaks (Brooks and Miller et al., 2009). When comparing USDA Choice ZH steaks to USDA Choice control steaks, both aged for 21 d, consumers noted no difference in beef flavor (Brooks and Miller, 2009). However, steaks from steers that had been supplemented for 30 d had reduced consumer scores for tenderness, juiciness, and overall acceptability when compared to control steaks (Brook and Miller, 2009). When comparing USDA Select steaks from ZH supplemented and control steers aged for 21 d, consumers found no differences in any of the traits of interest (Brooks and Miller, 2009). Finally, when comparing USDA Choice steaks from calf-fed Holstein steers supplemented with ZH to steaks from calf-fed Holsteins that were not supplemented with ZH, consumers indicated that control and 20 d supplemented steaks were similar for juiciness, flavor and overall acceptability, yet steaks from 20d

supplemented animals were rated as tougher (Brooks and Miller, 2009). However, when comparing control to 30 d supplemented steaks, those from calf-fed Holstein steers supplemented for 30 d with ZH were tougher, less juicy, less flavorful and less acceptable overall than control steaks (Brooks and Miller, 2009).

# Zilpaterol Withdrawal

The overall effectiveness of ZH can depend upon several factors including the rate of absorption from the digestive tract or the half-life of the compound within various tissues within the body (Robles-Estrada et al., 2009). Additionally, the overall effectiveness of ZH can also depend upon the degree of transformation of the compound upon digestion and the rate of excretion (Robles-Estrada et al., 2009). Zilpaterol hydrochloride has been shown to have a short half-life and a rapid rate of excretion from the body upon removal of the supplement. In a study by Shelver and Smith (2006), it was reported that ZH levels climbed rapidly in sheep supplemented with ZH (0.15 mg/kg BW daily) until a plateau was reached at 5 d. Following the removal of the supplement, levels of ZH in the tissue dropped rapidly (muscle, liver, and kidney levels decreased by 95% after 3 d withdrawal) (Shelver and Smith, 2006). The elimination of ZH during the first 3 d of withdrawal followed a first-order excretion pattern averaging an elimination half-life of  $12.7 \pm 4.0$  h (Shelver and Smith, 2006). Currently, ZH is labeled requiring a 3 d withdrawal of the compound prior to slaughter. Due to the rapid elimination of ZH from the body within the first 72 h of withdrawal and since it may not always be feasible to harvest an entire lot of cattle on the same date resulting in longer than expected withdrawal from ZH, concern for reversal of growth performance and carcass yield has grown.

Limited research has addressed the concern over the potential for a diminishing effect of ZH after extender periods of withdrawal. Robles-Estrada et al. (2009) evaluated withdrawal times of 3, 6, and 12 d to determine the impact of an extended withdrawal of ZH on growth performance and carcass merit. It was concluded that prolonging the withdrawal time beyond the required 3 d tended to decrease ADG, dressing percentage, and percent lean yield (Robles-Estrada et al., 2009).

# **Enhancement Technology for Improving Beef Tenderness**

Many beef wholesalers and retailers have begun using marination to increase the tenderness, juiciness, and flavor of cooked beef. Marination is a technique that has been around for many years and has been used to increase the probability of consumers having a quality eating experience. Marination can be a very valuable tool for increasing the consumer acceptance of lower quality beef. Typically, the marination of meat is a slow process involving the passive transport of marination ingredients into the meat. In recent years, beef processors have been interested in the utilization of functional non-meat ingredients and their abilities to increase tenderness, juiciness, and flavor of beef products; however, they were not willing to sacrifice the time need to incorporate these ingredients into beef products. New technology has allowed for the decrease in marination time by allowing marinades to be directly injected into the meat. The direct injection of the marinade into the meat decrease the time needed for the marinade to penetrate the meat and increase the uniformity of marinade intake into the meat. This new technique is referred to as enhancement, and it is documented that 14% of the wholemuscle beef in U.S. retail cases has undergone a form of enhancement (NCBA, 2007).

New technologies are continually being developed to improve current enhancement techniques.

While most enhancement solutions used in the current industry are proprietary and unique to individual goals and operations, most enhancement solutions contain similar ingredient with slight variations among ingredient types and amounts. Generally, all brine solutions contain salt, alkaline phosphates, and water (Miller, 1998). Additional ingredients such as lactates (sodium or potassium lactate), natural flavorings (rosemary extract), natural tenderizers (papain), and other spices or flavoring may be added depending on the primary goal for utilizing the enhancement solution (Brooks, 2007). Typical enhancement solutions are incorporated into the product at levels ranging between 8 – 12% and can be increased to 15% of the final product with minimal impact on sensory characteristics (Brooks, 2007). Since sodium is a major component within the brine, maintaining acceptable levels of salt in the final product to maintain consumer acceptability is a determining factor when choosing a final pump percentage.

In recent years, enhancement technology has become more important within the fresh beef industry. Research has indicated that consumers are willing to pay a premium for a consistently tender product, and this is a market many beef merchandisers are interested in supplying (Boleman et al., 1997). Consumers are willing to pay for a guaranteed tender product; however, the industry is plagued with inconsistency in tenderness at the consumer level (Voges et al., 2007), and this guaranteed tender product cannot be supplied without further processing. Enhancement is a technology that the beef industry is turning to in order to improve the inconsistencies seen between carcasses, muscles, and locations within muscles (Hamling and Calkins, 2008).

Robbins et al. (2002) utilized enhancement technology in an effort to improve sensory and retail display characteristics of beef rounds. Robbins et al. (2002) reported that sensory scores for tenderness, as well as shear force values, were improved with enhancement; however, trained panelist indicated that enhanced round roasts were more salty than unenhanced roasts. Grobbel et al. (2008) reported that enhancement improved WBS values for longissimus lumborum, semitendinosus, and tricepts brachii steaks, and this improved tenderness was also verified by trained sensory panelist in each of the three muscles. Moreover, Grobbel et al. (2008) concluded that enhancement decreased beef flavor and increased off-flavors as reported by trained sensory panelists. Additionally, it has been suggested that phosphate containing enhancement solutions may contribute to off-flavors or soapy tastes in meat (Vote et al., 2000); however, as documented by Robbins et al. (2002), soapy flavors were almost undetectable when beef roasts were pumped to contain 0.4% on a weight/weight basis.

While the utilization of phosphates in enhancements has been shown to decrease cooking loss (Detienne and Wicker, 1999), Robbins et al. (2002) found that enhanced steaks had a significantly higher cook loss than control steaks. It was concluded that the enhanced steaks had a greater amount of moisture to lose when compared to control steaks and was the reason for the increased cook loss. While the enhanced steaks did lose a greater amount of moisture (3.5% more than controls), they did retain > 6% of the added moisture from enhancement. Grobbel et al. (2008) documented that enhancement had no impact on cook loss.

Enhancement has also been shown to have an impact on meat color and retail shelf-life. Grobbel et al. (2008) documented that enhanced steaks were initially darker in

color when compared to control steaks. Darker initial steaks colors were also reported by Robbins et al. (2002), and it was concluded that the increased pH found in enhanced steaks due to enhancement results in darker colored lean. Moreover, Robbins et al. (2002) found that enhanced steaks had a more rapid rate of discoloration and darken when compared to control steaks. Steaks that were enhanced had lower a\* and b\* values, increased hue angles, and lower chroma values by 4 d of retail display (Robbins et al., 2002). Lawerence et al. (2004) evaluated the usage of calcium lactate in replacement of salt and phospate for improving color stability of enhanced strip loin steaks. Lawerence et al. (2004) also incorporate the antioxidant rosemary into each enhancement formulation to aid in oxidation rate. When compared to steaks enhanced with a salt, phosphate, and rosemary solution, steaks enhanced with calcium lactate and rosemary had improved display color stability. However, steaks enhanced with calcium lactate were rated as tougher than steaks enhanced with the salt/phosphate solution by trained sensory panelists which was in contrast to WBS values that indicated no change in tenderness between the two treatments (Lawerence et al., 2004).

While beef continues to be inconsistent in regards to consumer attributes, enhancement appears to be a worthy tool for improving the consistency of beef products. Trained sensory panel evaluations have proven that the juiciness and tenderness of beef can be improved through enhancement technology; however, the shelf-life and color of the product may be negatively impacted. Alternative ingredients to phosphate and salt used in enhancement solutions may allow for increased shelf-life and improved fresh meat color. Moreover, enhancement may allow for a greater usage of animal products

such as  $\beta$ -AA that improve growth rate and performance but have been shown to decrease tenderness and marbling resulting in less juiciness and reduced consumer appeal.

# Conclusion

Muscle growth, fat deposition, and product quality are of great concern to the red meat industry. Past research has shown the positive effects of  $\beta$ -AA supplementation on carcass composition and consequently increased profit. The results from studies using  $\beta$ -AAs vary, but overall, its effect on the hypertrophy of muscle fibers and fat deposition is positive.

Additionally, research has proven that tissue receptors are different within tissues and between species. More research is needed in all livestock species as the effects of varying the concentrations, duration, and withdrawal of  $\beta$ -AA's on animal growth and carcass composition needs to be determined. Continued research focused on understanding the outcomes when utilizing these compounds in beef cattle finishing diets is crucial for maximizing efficiency and profitability. Additionally, once the negative effects from the utilization of these compounds is recognized, identifying methods for correcting or reversing these effects will be needed. The utilization of  $\beta$ -AA technology can allow for greater production protein with greater efficiency and finding a way to allow for continued and/or increased utilization of these compounds will greatly impact to beef industry. However, maintaining a quality, desirable product for consumers is also a must, and establishing a system where both goals can be achieved is critical. By determining the locations within the carcass most affected by  $\beta$ -AA supplementation in terms of both size and tenderness, technologies such as enhancement can be utilized to improve the overall tenderness of products negatively affected by  $\beta$ -AA supplementation.

Therefore, if product acceptability can be maintained by post-fabrication technologies, utilization of  $\beta$ -AA in beef finishing diets can allow for a greater, more efficiently produced, protein supply, with minimal effects to product quality.

## CHAPTER III

# EFFECTS OF ZILPATEROL HYDROCHLORIDE AND ZILPATEROL HYDROCHLORIDE WITHDRAWAL TIME ON BEEF CARCASS CUTABILITY, COMPOSITION, AND TENDERNESS

# ABSTRACT

The impact of zilpaterol hydrochloride (ZH) on carcass yield, composition, and tenderness was evaluated using 384 beef steers in a randomized complete block design. Main effects were the addition of 0 or 8.3 mg/kg ZH for the final 20 d of feeding and each inclusion level was paired with withdrawal periods of 3, 10, 17, or 24 d. The two animals with weights closest to the pen average were selected for carcass fabrication to determine carcass yield, composition, and tenderness. The carcasses from animals fed ZH had greater (P = 0.008) individual side weights. Carcass fat determinations were unchanged (P = 0.70) by ZH. Weights of the strip loin (P = 0.01), peeled tenderloin (P = 0.01) 0.02), and top sirloin butt (P < 0.001) were all improved with ZH. When expressed as a proportion of carcass weight, ZH increased percentage of carcass in the top sirloin butt (P = 0.006), bottom sirloin tri-tip (P = 0.02), top inside round (P = 0.002), bottom round flat (P = 0.001), and flank steak (P = 0.02). A longer withdrawal time (WT) increased (P < 0.001)0.001) carcass weights. Shoulder clod weights were greatest (P < 0.001) with 17 d WT from ZH, while chuck roll weights were greatest (P = 0.02) at 17 and 24 d of WT. Peeled tenderloins, top sirloin butts, and eve of rounds responded to WT, with increased

(P < 0.001) weights seen at 10 d of WT as compared to all other WTs. Shear force values were higher at each of the 3 aging times, 7 d (P < 0.001), 14 d (P < 0.001), and 21 d (P = 0.003), in steaks from ZH fed steers compared to control steers. Protein percentages were greater in ZH steaks (P = 0.03) and ZH ground beef trim (P < 0.001). Percent moisture was increased (P < 0.001) in strip loin steaks at 3 and 10 d WT. Ground beef trim had an increase (P = 0.04) in percent moisture and a decrease (P = 0.01) in percent fat at 10 d WT. Carcass weights and yields were improved with ZH feeding and may continue to improve even up to 10 d after withdrawal of the supplement. Tenderness was slightly reduced with ZH supplementation but was unaffected by WT. Zilpaterol hydrochloride can be a valuable supplement to finishing beef steers to improve carcass lean yields and composition.

Key Words: Beef, Zilpaterol Withdrawal, Cutability, Tenderness

## **INTRODUCTION**

The use of  $\beta$ -adrenergic agonists ( $\beta$ -AA) to improve feed efficiency and enhance carcass composition in livestock species has been well documented since the early 1980's. Many  $\beta$ -AA act as repartitioning agents and have been shown to enhance lean meat production in many animal species. Zilpaterol hydrochloride (Zilmax®, Intervet, Millsboro DE, USA) is a  $\beta$ -AA that has been approved in Mexico and South Africa for over 10 yrs; however, it was not until 2006 that the compound was approved by the FDA for use in feedlot cattle in the U.S.

Zilpaterol hydrochloride (ZH) is marketed as a compound that will increase rate of gain, improve feed efficiency, and increase carcass leanness in cattle fed in

confinement systems. Initial studies by Avendano-Reyes et al. (2006) documented an increase in final body weight, increased ADG, and an improvement in G:F for cattle supplemented with ZH. Additionally, Avendano-Reyes et al. (2006) documented an increase in hot carcass weight (HCW), carcass yield, and loin muscle area with ZH. While the study by Avendano-Reyes et al. (2006) indicated a tendency for ZH fed cattle to have decreased 12<sup>th</sup> rib fat, a study by Plascencia et al. (1999) found no effect on 12<sup>th</sup> rib fat thickness with ZH inclusion. However, Montgomery et al. (2009b) reported that feeding 8.3 mg/kg (DM basis) of ZH for 30 days significantly decreased 12-rib fat. When combined with an increase in loin muscle area and HCW, calculated yield grade was decreased (i.e., improved), which was an indication of ZH ability to improve lean yield. Leheska et al. (2009) reported significant increases in carcass muscle deposition/protein accretion.

Zilpaterol hydrochloride has also been shown to increase weight of gross primal and boneless closely trimmed primals, and boneless closely trimmed retail cuts as a percentage of carcass weight, when carcass weights were held constant (Plascencia et al., 1999). Additionally, harvesting a lot of cattle on the same date can be problematic and may require longer than expected withdrawal periods and may result in withdrawal periods in excess of 3 days. Problems such has equipment failure, over-scheduling of cattle, trucking issues, market conditions (price fluctuations) among others, can arise at processing facilities and in the industry which can alter expected slaughter dates from planned. If compounds such as ZH are used during the last 20 d of finishing and an appropriate 3 d WT has been scheduled, the extended time on feed can result in extended WT. Since ZH is

rapidly eliminated in the urine (> 95% in 72 h) (Shelver and Smith, 2006), a withdrawal period of greater than 72 h could result in a loss of performance or a reversal in carcass yields. Furthermore, ZH has been shown to significantly increase Warner-Bratzler shear (WBS) values of loin muscle steaks; however, these reported values still range from severe to mild increases in WBS values (Avendano-Reyes et al., 2006; Casey et al., 1997; and Hilton et al., 2009).

The objective of this study was to determine the effects of zilpaterol hydrochloride and WT on beef carcass lean to fat ratios as well as carcass tenderness and proximate analysis. The studies were conducted on carcasses from Holland et al. (In Review).

### **MATERIALS AND METHODS**

A subset (n = 128) selected from three hundred eighty four (BW =  $356 \pm 23.3$  kg) British and British × Continental steers was used in this experiment. Steers were separated into two weight blocks and randomly assigned to pens (32 pens per block; 4 pens per treatment combination; 6 steers/pen) using a computer generated schedule. Within each block, pens were randomly assigned to a  $2 \times 4$  factorial arrangement of treatments. Main effects were the addition of 0 or 8.3 mg/kg (100% DM basis) ZH fed for 20 d at the end of the feeding period and each supplementation level was paired with withdrawal periods of 3, 10, 17, or 24 d prior to slaughter (Holland et al., In Review). Zilpaterol treatment began at d 95 and d 123 for heavy and light blocks, respectively. Cattle in ZH and control groups assigned to the same WT were weighed and slaughtered on the same days. Cattle were fed identical diets until the final finishing phase when ZH supplementation began. Zilpaterol treatment

cattle were fed 8.3 mg/kg (100% DM basis) for 20 d and monensin and tylosin were removed from the diet of ZH treated cattle at this time for the remainder of the feeding period. Control cattle continued to be fed monesin and tylosin for the entire finishing phase.

## Harvest and Carcass Selection

For the 3-d withdrawal steers only, 2 steers from each pen assigned to the control and ZH treatments were selected for intensive sampling. Animals whose BW was closest to pen average were selected for the intensive fabrication (n = 32). These animals were loaded in the evening and hauled less than 5 km to the Robert M. Kerr Food and Agricultural Products Center (FAPC) located on the campus of Oklahoma State University, for slaughter the next morning. Furthermore, all 10-d, 17-d, and 24-d steers from each pen assigned to the control and ZH treatments, were loaded in the evening and shipped 431 km to a commercial abbatoir for harvest the next morning. The two steers whose final BW was closest to their respective pen average were selected for harvest (n = 96). After chilling for a similar time at both locations, carcasses were ribbed at the 12<sup>th</sup> rib, and USDA Quality and Yield Grades and carcass traits were records (USDA, 1997). All carcass data was collected by trained Oklahoma State University personnel. Carcass sides were transported 431 km via commercial refrigerated truck (0 to  $-2^{\circ}$ C) to the FAPC for further fabrication. One carcass was removed from the study upon arrival at FAPC due to excessive trimming at the commercial abbatoir.

### **Carcass Fabrication**

Once carcasses arrived at FAPC, they were stored in holding coolers (0° to 4°C) until fabrication. Cold side weights (CSW) were recorded prior to fabrication using a certified on-line rail scale. Carcasses were then fabricated into various NAMP subprimals which included: 114E Chuck Shoulder Clod (0.635 cm trim), 114F Chuck Shoulder Tender, 116B Chuck (Mock) Tender, 130A Chuck Short Ribs, 116A Chuck Roll, 109B Rib Blade Meat, 112A Ribeye Roll, Lip-on, 124 Rib Back Ribs, 120 Brisket Whole, 115D Pectoral Meat (trimmed to blue), Pastrami Meat (serratus ventralis from the plate), 121C Outside Skirt, 121D Inside Skirt, 180 Strip Loin (0.635 cm trim), 189A Peeled Tenderloin, 184 Top Sirloin Butt, 185B Bottom Sirloin Ball Tip, 185D Bottom Sirloin Tri-Tip, 167A Peeled Knuckle, 168 Top Inside Round (0.635 cm trim), 171B Bottom Round Flat (0.635 cm trim), 171C Eye of Round (0.635 cm trim), 185A Bottom Sirloin Flap (denuded), 171F Heel Meat, 193 Flank Steak, Shank Meat, and Elephant Ear (cutaneous trunci from the flank). All trim from fabrication was segregated into one of three lean trim categories: 90% lean/10% fat (90/10), 80% lean/20% fat (80/20), or 50% lean/50% fat (50/50). Kidney knob fat, all trimmed fat, and all bones were also collected and weighed. After all weights of each side were recorded and entered, fabrication yield was calculated to ensure that 99% to 100.5% of CSW was recovered. Weights were recorded for all previously mentioned products of fabrication and were expressed as a percentage of CSW. Once all weights were recorded, all three trim levels were combined together for each animal, ground, and analyzed for percent moisture, fat, and protein. All trim levels were combined to allow for an estimation of lean differences between control and ZH carcasses. Combining of trim levels allows for

the most consistent comparison of proximate analysis of beef trim between treatments because different treatments produced different amounts and/or percentages of trim (Hilton, et al. unpublished); therefore, to determine proximate analysis of all trim, 50/50, 80/20 and 90/10 needed to be combined.

# Postmortem Aging and Strip Loin Fabrication

Upon completion of fabrication, strip loins (IMPS 180) were fabricated for shear force analysis. The anterior end of the strip loin was cut perpendicular to the long axis of the strip loin to allow for consistent and uniform steaks, the meat removed was used for proximate analysis. Proximate samples were vacuum packaged and frozen in a  $(-20^{\circ} \text{ to } -40^{\circ} \text{C})$  blast freezer and then held in a freezer (-10°C) until further analysis. Once the strip loins were cut perpendicular to the long axis of the strip loin on the anterior end, three 2.54 cm steaks were cut and assigned to one of three aging times (7 d, 14 d, or 21 d) based on the order the steaks were cut from the strip loin. For each additional strip loin fabricated, the order was advanced by one so that on the second strip loin, the first steak was assigned to age 14 d, steak 2 was assigned to age 21 d, and steak 3 was assigned to age 7 d. This pattern was continued for all strip loins fabricated. After cutting, all steaks were individually vacuum packaged and aged for their respective time under refrigeration at  $0^{\circ}$  to  $4^{\circ}$ C. After the assigned aging period, samples were frozen in a blast freezer (-20° to -40°C); and frozen, samples were held in a freezer (-10°C) until further analysis.

# Warner-Bratzler Shear Force

Warner-Bratzler shear force was completed using the American Meat Science Association guidelines (1995), with the following modification: Internal

temperatures of each steak were recorded prior to cooking insuring that internal temperatures were > 0°C and < 5° C. If temperatures were outside this range, steaks were either allowed to thaw for a longer period of time or replaced in refrigeration to return internal temperature to the desired range. Steaks were cooked using "The Next Generation" George Foreman Digital Grill (Model GRP99) to a medium degree of doneness (~71°C). Steaks were then placed on trays, covered with poly-vinyl wrap and refrigerated overnight to achieve an ultimate steak temperature of 2°C to 5°C. Following the overnight chill, two cores from each of the lateral, middle, and medial portions, for a total of six cores (1.27 cm), from each steak were removed parallel to the longitudinal orientation of the muscle fibers. Cores were shorn using a Warner-Bratzler Shear Testing Machine (G-R Elec. Mfg. Co., Manhattan, KS), and the peak shear force was recorded in kg and the average was determined. After each sample, the shear knife was cleaned, and the machine was reset to zero.

### **Proximate Analysis**

Samples for proximate analysis were thawed similar to the previously mentioned method used for thawing WBS steaks. After thawing, samples were powder homogenized using a blender (model 51BL31, Waring, Torrington, CT). Samples were analyzed for nitrogen content using the combustion method procedures of the AOAC (1990). Samples were dried in an oven at 102°C for 24 h for moisture content determination. Lipid percentage was determined by ether extraction of the dried samples following the procedures of the AOAC (1990). *Data Analysis*  In this study the interaction of ZH and WT was not significant so only main effect means are reported. Data were analyzed using the mixed model procedures of SAS. Analysis of variance for a complete random design with the main effects of ZH and WT was analyzed. Two weight blocks (heavy weight and light weight) were included in the model as fixed variables; block was not significant so results were pooled over block effect. Carcass side was the experimental unit used for analysis. For ZH and WT, all carcasses (control and ZH fed) were included together for analysis. Least squares means were generated and separated using a pairwise t-test when the model displayed a treatment effect ( $\alpha < 0.05$ ). Miller et al. (2001) used a range from 3.92 to 4.50 kg as intermediate and tough as greater than 4.5 kg when comparing beef steaks. The frequency of tender, intermediate, and tough steaks was determined using the method established by Miller et al. (2001) and were analyzed using the Chi-square procedure.

#### **RESULTS AND DISCUSSION**

#### Zilpaterol Hydrochloride

Results for growth performance characteristics, as well as USDA grade data, were reported by Holland et al. (2009). Zilpaterol hydrochloride inclusion in the diet had a significant effect on individual side weights with ZH supplemented animals having heavier (P = 0.008) weights. These finding are in agreement with previous studies by Avendano-Reyes et al. (2006) and Plascencia et al. (1999) where carcass weights increased with ZH. Furthermore, there was an increase (< 0.001) in the percentage of total wholesale carcass lean (total side weight minus 50/50 trim, 80/20 trim, 90/10 trim, kidney knob fat, total fat trim, and total bone) with ZH

supplementation (Table 3.1). However, no effect of ZH treatment on lean trim percentages or percent carcass fat was observed. A significant reduction (P = 0.05) in total bone weight was documented, which resulted in a decreased (P < 0.001) percentage of bone in the carcass side (Table 3.1). Carcass fat also remained unchanged by ZH in a study by Avendano-Reyes et al. (2006); however, in the same study, it was reported that ZH had no significant impact on percent carcass bone, which contradicts the findings of this study. Due to a decrease in percent bone and with percent fat being unchanged, the increase in carcass weight is presumed to be due to an increase in carcass lean.

Although most major primals from the forequarter, such as the shoulder clod, chuck roll, and ribeye roll were not affected by ZH treatment, several cuts increased in weight with ZH inclusion. A significant increase in the weight of pectoral meat (P = 0.03), rib blade meat (P = 0.03), and pastrami meat (P = 0.04) was shown with ZH feeding. Zilpaterol hydrochloride inclusion also resulted in increased weight of the whole brisket (P < 0.001) (Table 3.2).

More of the muscles and primals from the hindquarter exhibited greater response to ZH as compared to those from the forequarter. Higher valued cuts, such as the strip loin (P = 0.01), peeled tenderloin (P = 0.02), and top sirloin butt (P < 0.001) all increased in weight with ZH supplementation. In addition to these cuts, several other cuts from the hindquarter were heavier with ZH, including the bottom sirloin tri-tip (P = 0.005), top inside round (P < 0.001), bottom round flat (P < 0.001), eye of round (P = 0.02), and flank steak (P = 0.005). Also, an increase in weight was observed in the heel (P = 0.02) and the shank (P < 0.01) with the inclusion of ZH. While several of the primal and cut weights significantly increased, only the top sirloin butt (P = 0.006), bottom sirloin tri-tip (P = 0.02), top inside round (P = 0.002), bottom round flat (P = 0.001), and the flank steak (P = 0.02) increased when cut weight was expressed as a percentage of CSW (Table 3). While Avendano-Reyes et al. (2006) did not report individual subprimal weights; they reported that carcass yields were increased with ZH. However, when carcasses were deboned, ZH steers yielded equal amounts of lean as control steers (Avendano-Reyes et al., 2006). In a study by Plascencia et al. (1999), where individual subprimals were weighed and expressed as a percentage of carcass weight, several subprimals such as the sirloin, knuckle, inside skirt, and inside round were increased with ZH supplementation. However, in the same study, several subprimals were significantly reduced either in weight or percentage by ZH supplementation (Plascencia et al., 1999). This contradicts current findings where no subprimals were significantly reduced in weight by ZH.

As shown in this study, the response to ZH seemed to be greater in hindquarter muscles as compared to those recovered from the forequarter. Previous studies indicate that type II fibers have a greater response to  $\beta$ -AA- stimulations as compared to other muscle fiber types (Miller et al., 1988; Smith et al., 1995). Furthermore, as reported by Kirchofer et al. (2002) there is a greater variation among fiber types within muscles of the chuck and the round is comprised mainly of white muscle fibers. These findings could explain the variation in response between muscles within this study, as those muscles with a greater proportion of white fibers had a greater response.

### Zilpaterol Hydrochloride Withdrawal Time

Cold carcass weight was significantly affected by ZH WT, with carcasses from animals withheld for 3 d having lighter (P < 0.001) weights as compared to all other WTs. However, this increase in weight most likely resulted from cattle being fed longer because of increased WT (Table 3.1).

The forequarter also responded to the various WTs. Shoulder clod weights were affected (P < 0.001) by WT with 3 d withdrawal weights being the lightest and 17 d withdrawal weights being the heaviest. Weights were similar between the 10 d and 24 d withdrawal. Chuck roll weights were impacted (P = 0.02) by WT as chuck rolls were heaviest with 24 d WT than with 3 d or 10 d WT. In addition to the shoulder clod and chuck roll, WT had a significant effect (P < 0.001) on brisket weight. Briskets were lightest at WTs of 3 and 10 d, and heaviest at withdrawal d 17 and 24 (Table 2). Additional minor cuts affected by WT included rib blade meat (P < 0.001), back ribs (P < 0.001), pastrami meat (P = 0.003), inside skirt (P = 0.03), and the outside skirt (P = 0.005) (Table 3.2).

Much like the forequarter, most major subprimals from the hindquarter were affected by ZH WT. Peeled tenderloins were affected (P < 0.001) by ZH WTs with tenderloin weights being the lowest at 3 d WT and greatest at 10 d WT. Weights at 17 d WT were similar to those documented at WT of 24 d, but were less than for 10 d WT. Top sirloin butts were also affected (P < 0.001) by WT with the greatest top sirloin butt weights occurring at 10 d WT and the lightest at 24 d WT. Top sirloin butt weights at 17 d were similar, with weights recorded at 17 d WT being similar to weights recorded at 24 d WT. Additionally, WT had a significant

effect (P = 0.03) on eye of round weights. Weights increased from 3 d WT to 10 d WT, but remained similar at 17 d WT. However, eye of round weights at 24 d WT were similar to those at 3 d WT. Moreover, knuckle weights were increased (P = 0.02) from 3 d to 10 d WT, but remained unchanged for the remaining WT. Furthermore, like with the forequarter, various minor cuts from the hindquarter were impacted by ZH WT. These cuts included the bottom sirloin ball tip (P = 0.01), bottom sirloin tri-tip (P < 0.001), shank meat (P = 0.05), bottom sirloin flap (P < 0.001), flank steak (P = 0.002), and the "elephant ear" (P < 0.001) (Table 3.3).

In a previous study where ZH withdrawal was investigated (Casey et al., 1997), it was reported that ZH had no lasting effect on carcass or meat characteristics. In the current study, numerous subprimals continued to increase in weight as WT moved from 3 d to 10 d while several other subprimals continued to increase in weight throughout the entire 24 d withdrawal period. While little data is available to support the idea of continued action of ZH with longer withdrawal periods, a study by Sissom et al. (2007) suggests that ZH alters mRNA and protein concentrations of  $\beta$  –adrenergic receptors of muscle cells which could impact the cellular response to ZH when exposed for an extended period.

Although it is understood that feeding  $\beta$ -AA to livestock increases lean: fat ratios, various theories remain as to how these compounds truly affect muscle and fat synthesis. While various researchers still debate the true mechanisms of increased muscle accretion as being an increase in muscle cell hypertrophy, a reduction in muscle protein degradation, or a combination of both, it is apparent that an increase

in lean mass is seen to a greater extent in ZH fed cattle as opposed to cattle fed ractopamine hydrochloride as was reported by Avendano-Reyes et al. (2006).

# Warner-Bratzler Shear Force

Shear force values of strip loin steaks were significantly lower for control animals as compared to ZH treated animals at 7 (3.84 kg vs. 4.65 kg) (P < 0.001), 14 (3.44 kg vs. 4.18 kg) (*P* < 0.001), and 21 (3.18 kg vs. 3.61 kg) (*P* = 0.003) d aging. At 7 d of aging, 17.44% of the control steaks exceeded the threshold for tender qualification and were considered intermediate/tough, as compared to 46.15% of the ZH steaks that were intermediate/tough. Even though ZH steaks produced higher WBS values through 14 and 21 d aging periods, the percentage exceeding the tenderness threshold dropped to 1.61% and 10.77% for control and ZH steaks, respectively, by d 21 (Fig. 3.1). Much like the present study, Strydom et al. (2009) reported an increase in WBS values with  $\beta$ -agonist supplementation and also indicated favorable responses to aging time up to 14 days. Moreover, Strydom et al. (2009) concluded that tenderness effects due to  $\beta$ -agonist inclusion were associated with their effect on calpastatin. While calpastatin levels were not measured in the current study, tenderness effects due to ZH inclusion could be due to the effects of ZH on calpastatin activity.

While WT tended (P = 0.06) to have an impact on WBS value at 7 d postmortem aging, it had little to no impact on WBS value at aging times of 14 (P = 0.76) and 21 d (P = 0.73). At 7 d aging, animals withdrawn from ZH for 3 and 24 d produced steaks with the highest shear force values with WBS values of 4.34 kg and 4.60 kg respectively, while the lowest values were seen in steaks from animals

withdrawn for 10 and 17 d as indicated by WBS values of 3.98 kg and 4.06 kg respectively.

Much like the results in this study, increased WBS of steaks from animals supplemented with  $\beta$ -AA is commonly reported. However, O'Neill (2001) concluded ZH did not cause tougher meat when compared to non-treated animals. This, however, is a rare finding, as several studies have found that numerous  $\beta$ -AA's including ZH increase WBS values (Pringle et al., 1993; Schroeder et al., 2003; Hilton et al., 2009).

# **Proximate Analysis**

Zilpaterol hydrochloride supplementation had no impact on moisture (P = 0.23) or fat (P = 0.27) content of ground beef trim or the moisture (P = 0.97) and fat (P = 0.11) content of strip loin steaks. A significant increase (P = 0.03) was found in percent protein of ground beef trim with ZH supplementation; however, this increase was less than one percentage unit (Table 3.4). Inclusion of ZH also increased (P < 0.001) percent protein in strip loin steaks (Table 3.4). While fat content was not statistically different between treatments, fat content in beef trim as well as in strip loin steaks was numerically lower in steaks and trim from ZH supplemented animals. This reduction in fat allows for a significant increase in protein of strip steaks and beef trim and was as expected due to the ability of ZH to increase protein accretion throughout the carcass.

Withdrawal time seemed to have a different effect on proximate analysis determinations as compared to those seen with ZH inclusion. Moisture percentage (P = 0.04) and fat percentage (P = 0.01) of ground beef trim were significantly

impacted by ZH WT, while protein percentages were unchanged (P = 0.56). Ground beef moisture content was the greatest at 10 d withdrawal from ZH. Fat percentages were highest in ground beef trim from animals withdrawn for 3 and 17 d, while fat levels were lowest in animals withdrawn for 10 d. Animals withdrawn for 24 d had intermediate fat percentages (Table 3.4). While ZH WT impacted moisture and fat percentages in ground beef trim, ZH WT had no impact on fat (P = 0.65) and protein (P = 0.14) percentages in strip loin steaks, but did affect moisture content (P < 0.001). Moisture content in strip loin steaks was greatest at WTs of 3 and 10 d, with withdrawal d 17 and 24 resulting in significantly lower moisture content (Table 3.4). While ZH WT did not significantly impact percent fat in strip loins steaks like seen in beef trim, the percentage fat did increase numerically as moisture content decreased which resembles the same relationship seen between percent moisture and percent fat in beef trim.

Few data exists to explain the differences seen in proximate analysis of strip loin steaks and ground beef trim, furthermore, the differences that were seen due to ZH were to be expected. Zilpaterol hydrochloride has been shown to increase lean yields in carcasses, so it is logical to see a concurrent increase in the percent protein found in ground beef trim as well as in strip loin steaks with ZH supplementation. Moreover, while percent fat was not significantly reduced in either of the samples measured, reported values were numerically lower for ZH when compared to controls. Furthermore, there were no differences seen in protein levels across the various WTs which seems illogical since increases in various carcass cuts were seen with increased WT. This could be due to less effect of ZH on lean within the entire

animal so there were minimal carryover effects of ZH after withdrawal on the lean in samples that were collected.

In conclusion, the repartitioning agent zilpaterol hydrochloride, when fed 20 d prior to slaughter, increased carcass weights and yields in beef steers which led to an increase in wholesale carcass lean. Warner Bratzler shear force values were increased with ZH supplementation, however, with appropriate aging, these values were reduced. Withdrawal time of ZH seemed to have no negative impact on product tenderness or proximate analysis of ground beef trim or strip loin steaks, while improvements in some carcass cuts were seen with 10 d of withdrawal from ZH.

| Item                                 |         | ZH        |         |                     | Withdrawal (d)      |                     |                     |         |      |  |
|--------------------------------------|---------|-----------|---------|---------------------|---------------------|---------------------|---------------------|---------|------|--|
|                                      | 0 mg/kg | 8.3 mg/kg | Pr > F  | 3                   | 10                  | 17                  | 24                  | Pr > F  | SEM  |  |
| Total Side Weight                    | 180.97  | 184.30    | < 0.01  | 178.16 <sup>b</sup> | 184.69 <sup>a</sup> | 184.12 <sup>a</sup> | 183.60 <sup>a</sup> | < 0.001 | 1.24 |  |
| Wholesale Carcass Lean <sup>1*</sup> | 50.03   | 51.49     | < 0.001 | 49.94               | 50.80               | 51.49               | 50.80               | 0.06    | 0.18 |  |
| 50/50 Trim, kg                       | 20.47   | 20.25     | 0.66    | 19.93               | 20.67               | 20.82               | 20.03               | 0.47    | 0.61 |  |
| % 50/50 trim <sup>1</sup>            | 11.31   | 10.98     | 0.19    | 11.18               | 11.20               | 11.31               | 10.91               | 0.71    | 0.31 |  |
| 80/20 Trim, kg                       | 6.10    | 6.17      | 0.82    | 6.06                | 6.44                | 5.98                | 6.06                | 0.78    | 0.60 |  |
| % 80/20 trim <sup>1</sup>            | 3.37    | 3.35      | 0.94    | 3.41                | 3.49                | 3.25                | 3.29                | 0.78    | 0.33 |  |
| 90/10 Trim, kg                       | 6.78    | 7.16      | 0.21    | 7.02                | 7.19                | 7.03                | 6.64                | 0.62    | 0.57 |  |
| % 90/10 trim <sup>1</sup>            | 3.74    | 3.89      | 0.38    | 3.93                | 3.88                | 3.82                | 3.63                | 0.57    | 0.31 |  |
| Kidney Knob Fat, kg                  | 5.53    | 5.24      | 0.21    | 5.12                | 5.26                | 5.56                | 5.62                | 0.37    | 0.27 |  |
| Total Fat Trim, kg                   | 15.03   | 17.46     | 0.70    | 14.77               | 14.54               | 14.46               | 14.75               | 0.50    | 0.74 |  |
| % Carcass Fat <sup>1</sup>           | 11.34   | 10.83     | 0.19    | 11.12               | 10.71               | 10.85               | 11.68               | 0.31    | 0.39 |  |
| Total Bone, kg                       | 35.96   | 35.27     | 0.05    | 35.42               | 35.97               | 35.24               | 35.83               | 0.40    | 0.35 |  |
| % Carcass Bone <sup>1</sup>          | 19.90   | 19.15     | < 0.001 | 19.92               | 19.51               | 19.15               | 19.53               | 0.07    | 0.20 |  |

Table 3.1. Effects of ZH inclusion into the diet and withdrawal time of ZH prior to slaughter on carcass cutout characteristics (n = 127).

<sup>a, b, c</sup> Within a row and main effect of withdrawal, means with different superscripts differ (P < 0.05). <sup>a, b, c</sup> Within a row and main effect of withdrawal, means with different superscripts differ (P < 0.05). <sup>1</sup>Percentage of cold side weight. <sup>\*</sup>total side weight minus 50/50 trim, 80/20 trim, 90/10 trim, kidney knob fat, total fat trim, and total bone.

|  | ZH      |           |         | Withdrawal (d)     |                    |                    |                    |         |      |
|--|---------|-----------|---------|--------------------|--------------------|--------------------|--------------------|---------|------|
| Item   | 0 mg/kg | 8.3 mg/kg | Pr > F  | 3                  | 10                 | 17                 | 24                 | Pr > F  | SEM  |
| Shoulder clod, trimmed, kg                   | 8.88    | 9.02      | 0.20    | 8.41 <sup>c</sup>  | 9.05 <sup>b</sup>  | 9.53 <sup>a</sup>  | 8.81 <sup>b</sup>  | < 0.001 | 0.11 |
| Shoulder Clod trimmed <sup>1</sup>           | 4.91    | 4.90      | 0.88    | 4.72 <sup>c</sup>  | $4.90^{b}$         | $5.18^{a}$         | $4.80^{bc}$        | < 0.001 | 0.06 |
| Chuck Shoulder Tender, kg                    | 0.51    | 0.51      | 0.88    | $0.48^{b}$         | 0.63 <sup>a</sup>  | $0.48^{b}$         | $0.46^{b}$         | < 0.001 | 0.02 |
| Chuck Shoulder Tender <sup>1</sup>           | 0.28    | 0.28      | 0.66    | $0.26^{b}$         | 0.34 <sup>a</sup>  | $0.26^{b}$         | $0.25^{b}$         | < 0.001 | 0.01 |
| Chuck Roll, kg                               | 13.74   | 14.12     | 0.22    | 13.45 <sup>b</sup> | 13.48 <sup>b</sup> | $14.18^{ab}$       | 14.61 <sup>a</sup> | 0.02    | 0.43 |
| Chuck Roll <sup>1</sup>                      | 7.59    | 7.66      | 0.69    | $7.56^{ab}$        | 7.29 <sup>b</sup>  | 7.69 <sup>ab</sup> | 7.96 <sup>a</sup>  | 0.02    | 0.23 |
| Chuck Mock Tender, kg                        | 1.53    | 1.75      | 0.23    | 1.47               | 1.61               | 1.90               | 1.58               | 0.39    | 0.18 |
| Chuck Mock Tender <sup>1</sup>               | 0.85    | 0.95      | 0.30    | 0.83               | 0.87               | 1.04               | 0.86               | 0.46    | 0.10 |
| Chuck Short Ribs, kg                         | 1.37    | 1.36      | 0.87    | 1.04               | 1.39               | 1.41               | 1.38               | 0.49    | 0.06 |
| Chuck Short Ribs <sup>1</sup>                | 0.76    | 0.74      | 0.84    | 0.72               | 0.76               | 0.76               | 0.75               | 0.81    | 0.03 |
| Pectoral Meat, trimmed to blue, kg           | 0.89    | 0.99      | 0.03    | 0.88               | 0.99               | 0.95               | 0.94               | 0.37    | 0.08 |
| Pectoral Meat, trimmed to blue <sup>1</sup>  | 0.49    | 0.54      | 0.06    | 0.49               | 0.54               | 0.52               | 0.51               | 0.60    | 0.04 |
| Rib Blade Meat, kg                           | 1.56    | 1.71      | 0.03    | $1.78^{a}$         | $1.81^{a}$         | 1.42 <sup>b</sup>  | 1.54 <sup>b</sup>  | < 0.001 | 0.09 |
| Rib Blade Meat <sup>1</sup>                  | 0.86    | 0.93      | 0.07    | $1.00^{a}$         | $0.98^{a}$         | $0.77^{b}$         | $0.84^{b}$         | < 0.001 | 0.05 |
| Ribeye Roll, kg                              | 5.62    | 5.72      | 0.20    | 5.63               | 5.72               | 5.71               | 5.63               | 0.80    | 0.10 |
| Ribeye Roll <sup>1</sup>                     | 3.10    | 3.11      | 0.88    | 3.16               | 3.10               | 3.10               | 3.07               | 0.44    | 0.05 |
| Rib Back Ribs, kg                            | 1.43    | 1.44      | 0.89    | 1.39 <sup>b</sup>  | 1.59 <sup>a</sup>  | 1.41 <sup>b</sup>  | 1.35 <sup>b</sup>  | < 0.001 | 0.34 |
| Rib Back Ribs <sup>1</sup>                   | 0.79    | 0.78      | 0.55    | $0.78^{b}$         | $0.86^{a}$         | $0.77^{b}$         | $0.74^{b}$         | < 0.001 | 0.02 |
| Pastrami Meat, kg                            | 0.59    | 0.65      | 0.04    | $0.56^{b}$         | $0.66^{a}$         | $0.59^{b}$         | $0.67^{a}$         | < 0.01  | 0.34 |
| Pastrami Meat <sup>1</sup>                   | 0.33    | 0.35      | 0.10    | 0.31 <sup>c</sup>  | 0.36 <sup>ab</sup> | $0.32^{bc}$        | 0.37 <sup>a</sup>  | 0.01    | 0.02 |
| Brisket Whole, bnls packer trim, kg          | 5.62    | 6.05      | < 0.001 | 5.30 <sup>b</sup>  | 5.48 <sup>b</sup>  | 6.76 <sup>a</sup>  | 6.31 <sup>a</sup>  | < 0.001 | 0.14 |
| Brisket Whole, bnls packer trim <sup>1</sup> | 3.11    | 3.28      | < 0.01  | 2.97 <sup>b</sup>  | 2.97 <sup>b</sup>  | $3.40^{a}$         | 3.44 <sup>a</sup>  | < 0.001 | 0.07 |
| Inside Skirt, kg                             | 1.03    | 1.10      | 0.09    | $0.98^{b}$         | 1.14 <sup>a</sup>  | 1.09 <sup>ab</sup> | 1.05 <sup>ab</sup> | 0.03    | 0.05 |
| Inside Skirt <sup>1</sup>                    | 0.57    | 0.60      | 0.20    | 0.55               | 0.62               | 0.59               | 0.57               | 0.12    | 0.03 |
| Outside Skirt, kg                            | 0.65    | 0.63      | 0.25    | $0.58^{b}$         | $0.66^{a}$         | $0.67^{a}$         | $0.65^{a}$         | < 0.01  | 0.02 |
| Outside Skirt <sup>1</sup>                   | 0.36    | 0.34      | 0.10    | 0.33               | 0.36               | 0.36               | 0.36               | 0.06    | 0.01 |

Table 3.2. Effects of ZH inclusion into the diet and withdrawal time of ZH prior to slaughter on various wholesale beef cuts from the forequarter (n = 127).

<sup>a, b, c</sup> Within a trait and main effect of withdrawal, means with different superscripts differ (P < 0.05). <sup>1</sup>Listed as a percentage of cold side weight.

|  | ZH      |           |         | Withdrawal (d)    |                   |                    |                    |         |      |
|--|---------|-----------|---------|-------------------|-------------------|--------------------|--------------------|---------|------|
| Item                                       | 0 mg/kg | 8.3 mg/kg | Pr > F  | 3                 | 10                | 17                 | 24                 | Pr > F  | SEM  |
| Strip Loin, kg                             | 4.87    | 5.09      | 0.01    | 4.93              | 5.12              | 4.94               | 4.94               | 0.41    | 0.0  |
| Strip Loin <sup>1</sup>                    | 2.69    | 2.76      | 0.09    | 2.76              | 2.76              | 2.68               | 2.69               | 0.36    | 0.04 |
| Peeled Tender, side muscle on, kg          | 2.66    | 2.76      | 0.02    | 2.52 <sup>c</sup> | $2.86^{a}$        | 2.72 <sup>b</sup>  | $2.74^{ab}$        | < 0.001 | 0.0  |
| Peeled Tender, side muscle on <sup>1</sup> | 1.47    | 1.50      | 0.22    | 1.42 <sup>c</sup> | 1.55 <sup>a</sup> | $1.48^{bc}$        | $1.49^{ab}$        | < 0.01  | 0.0  |
| Top Sirloin Butt, kg                       | 5.47    | 5.82      | < 0.001 | 5.69 <sup>b</sup> | 6.06 <sup>a</sup> | 5.49 <sup>bc</sup> | 5.34 <sup>c</sup>  | < 0.001 | 0.1  |
| Top Sirloin Butt <sup>1</sup>              | 3.03    | 3.16      | < 0.01  | 3.20 <sup>a</sup> | 3.28 <sup>a</sup> | $2.98^{b}$         | 2.91 <sup>b</sup>  | < 0.001 | 0.0  |
| Bottom Sirloin Ball Tip, denuded, kg       | 0.58    | 0.66      | 0.07    | $0.61^{ab}$       | 0.69 <sup>a</sup> | $0.68^{a}$         | $0.50^{b}$         | 0.01    | 0.05 |
| Sirloin Ball Tip, denuded <sup>1</sup>     | 0.32    | 0.36      | 0.12    | $0.34^{ab}$       | 0.37 <sup>a</sup> | 0.37 <sup>a</sup>  | $0.27^{b}$         | 0.02    | 0.02 |
| Bottom Sirloin Tri-Tip, denuded, kg        | 0.99    | 1.08      | < 0.01  | 1.02 <sup>b</sup> | 1.14 <sup>a</sup> | $1.08^{ab}$        | $0.90^{\circ}$     | < 0.001 | 0.04 |
| Sirloin Tri-Tip, denuded <sup>1</sup>      | 0.55    | 0.59      | 0.02    | $0.57^{a}$        | $0.62^{a}$        | $0.59^{a}$         | $0.49^{b}$         | < 0.001 | 0.02 |
| Knuckle, peeled, kg                        | 5.16    | 5.26      | 0.34    | 4.93 <sup>b</sup> | 5.27 <sup>a</sup> | 5.29 <sup>a</sup>  | 5.34 <sup>a</sup>  | 0.02    | 0.12 |
| Knuckle, peeled <sup>1</sup>               | 2.85    | 2.85      | 0.99    | 2.77              | 2.85              | 2.88               | 2.91               | 0.20    | 0.00 |
| Top Inside Round, kg                       | 9.71    | 10.36     | < 0.001 | 9.72              | 10.21             | 10.23              | 10.01              | 0.06    | 0.15 |
| Top Inside Round <sup>1</sup>              | 5.37    | 5.62      | < 0.01  | 5.46              | 5.53              | 5.56               | 5.43               | 0.62    | 0.08 |
| Bottom Round Flat, kg                      | 6.59    | 7.14      | < 0.001 | 6.75              | 6.76              | 6.90               | 7.04               | 0.42    | 0.14 |
| Bottom Round Flat <sup>1</sup>             | 3.64    | 3.87      | < 0.01  | 3.79              | 3.66              | 3.75               | 3.83               | 0.39    | 0.0  |
| Eye of Round, kg                           | 2.59    | 2.78      | 0.02    | 2.52 <sup>b</sup> | $2.80^{a}$        | $2.78^{a}$         | 2.63 <sup>ab</sup> | 0.03    | 0.08 |
| Eye of Round <sup>1</sup>                  | 1.43    | 1.50      | 0.08    | 1.41              | 1.52              | 1.51               | 1.43               | 0.16    | 0.04 |
| Heel Meat, kg                              | 2.27    | 2.39      | 0.02    | 2.29              | 2.36              | 2.39               | 2.29               | 0.31    | 0.0  |
| Heel Meat <sup>1</sup>                     | 1.26    | 1.30      | 0.10    | 1.28              | 1.28              | 1.30               | 1.25               | 0.54    | 0.02 |
| Shank Meat, kg                             | 2.58    | 2.71      | < 0.01  | 2.59 <sup>b</sup> | 2.61 <sup>b</sup> | 2.62 <sup>b</sup>  | $2.75^{a}$         | 0.05    | 0.0  |
| Shank Meat <sup>1</sup>                    | 1.43    | 1.47      | 0.08    | $1.46^{ab}$       | 1.41 <sup>b</sup> | 1.42 <sup>b</sup>  | $1.50^{a}$         | 0.05    | 0.04 |
| Bottom Sirloin Flap, denuded, kg           | 1.32    | 1.34      | 0.69    | $1.20^{b}$        | 1.37 <sup>a</sup> | $1.40^{a}$         | 1.37 <sup>a</sup>  | < 0.001 | 0.09 |
| Bottom Sirloin Flap, denuded <sup>1</sup>  | 0.73    | 0.73      | 0.77    | $0.67^{b}$        | $0.74^{a}$        | $0.76^{a}$         | $0.74^{a}$         | < 0.01  | 0.0  |
| Flank Steak, kg                            | 0.82    | 0.89      | < 0.01  | 0.79 <sup>c</sup> | $0.90^{a}$        | $0.84^{bc}$        | $0.89^{ab}$        | < 0.01  | 0.0  |
| Flank Steak <sup>1</sup>                   | 0.45    | 0.48      | 0.02    | 0.44 <sup>c</sup> | $0.49^{a}$        | $0.46^{bc}$        | $0.48^{ab}$        | 0.02    | 0.02 |
| Elephant Ear, kg                           | 1.50    | 1.57      | 0.18    | 1.23 <sup>c</sup> | 1.52 <sup>b</sup> | $1.78^{a}$         | $1.60^{b}$         | < 0.001 | 0.0  |
| Elephant Ear <sup>1</sup>                  | 0.83    | 0.85      | 0.39    | 0.69 <sup>c</sup> | 0.83 <sup>b</sup> | $0.97^{a}$         | $0.87^{b}$         | < 0.001 | 0.0  |

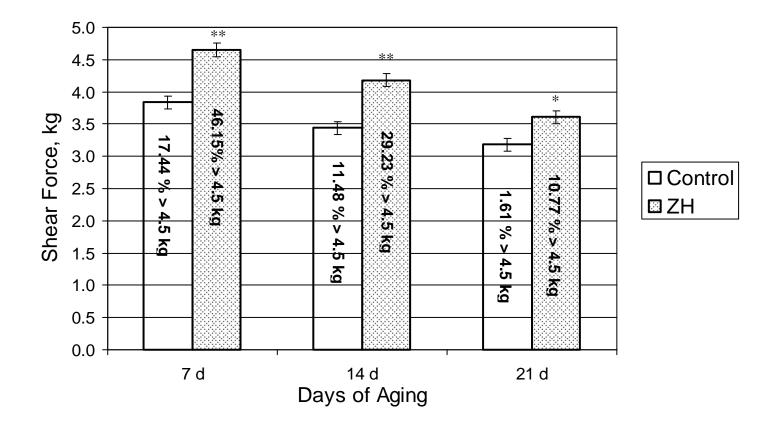
Table 3.3. Effects of ZH inclusion into the diet and withdrawal time of ZH prior to slaughter on various wholesale beef cuts from the hindquarter (n = 127).

| Item                          | ZH      |           |         |                    |                    |                    |                     |         |      |
|-------------------------------|---------|-----------|---------|--------------------|--------------------|--------------------|---------------------|---------|------|
|                               | 0 mg/kg | 8.3 mg/kg | Pr > F  | 3                  | 10                 | 17                 | 24                  | Pr > F  | SEM  |
| Strip Loin Steak              |         |           |         |                    |                    |                    |                     |         |      |
| Moisture %                    | 74.67   | 74.66     | 0.97    | $75.37^{a}$        | 75.19 <sup>a</sup> | 74.01 <sup>b</sup> | 74.09 <sup>b</sup>  | < 0.001 | 0.35 |
| Fat %                         | 4.47    | 3.61      | 0.11    | 3.50               | 4.44               | 4.09               | 4.14                | 0.65    | 0.54 |
| Protein %                     | 22.87   | 23.41     | < 0.001 | 22.95              | 23.40              | 23.02              | 23.20               | 0.14    | 0.25 |
| Ground Beef Trim <sup>1</sup> |         |           |         |                    |                    |                    |                     |         |      |
| Moisture %                    | 50.74   | 51.52     | 0.23    | 50.17 <sup>b</sup> | 52.63 <sup>a</sup> | $50.76^{b}$        | 50.96 <sup>ab</sup> | 0.04    | 0.66 |
| Fat %                         | 33.48   | 32.45     | 0.27    | 33.95 <sup>a</sup> | 30.62 <sup>b</sup> | 34.81 <sup>a</sup> | 32.49 <sup>ab</sup> | 0.01    | 1.20 |
| Protein %                     | 13.91   | 14.43     | 0.03    | 14.35              | 14.32              | 13.95              | 14.07               | 0.56    | 0.39 |

Table 3.4. Effects of ZH inclusion into the diet and withdrawal time of ZH prior to slaughter on meat quality characteristics (n = 127).

<sup>a, b, c</sup> Within a row and main effect of withdrawal, means with different superscripts differ (P < 0.05). <sup>1</sup>Combination of 50/50, 80/20, and 90/10 trim.

**Figure 3.1.** The main effect of ZH treatment on Warner Bratzler shear force values of strip loin steaks. Within a specific aging time, means with a single asterisk (\*) differ (P < 0.01), whereas means with a double asterisks (\*\*) differ (P < 0.001).



### CHAPTER IV

# THE EFFECTS OF ZILPATEROL HYDROCHLORIDE ON CARCASS CUTABILITY AND TENDERSS OF CALF-FED HOLSTEIN STEERS

# ABSTRACT

To evaluate the impact of zilpaterol hydrochloride (ZH) on carcass cutability and tenderness of calf-fed Holstein steers, calf-fed Holstein carcasses (n = 102) were selected from a pool of 2,300 steers that were either fed 0 or 8.3mg/kg (DM basis) of ZH. Zilpaterol hydrochloride was supplemented the last 20 d of the finishing period and withdrawn for 3 d prior to slaughter. Carcasses were selected based on carcass weight as well as predetermined USDA Yield Grade categories. For tenderness evaluation, steaks from the strip loin, bottom round, and top round were either used as a control or were injected with an enhancement solution and aged for 14 or 21 d. Carcasses from ZH fed steers had a greater (P < 0.0001) amount of wholesale carcass lean than control fed steers. Additionally, ZH fed steers had a greater ( $P \le 0.01$ ) subprimal yield from the shoulder clod, strip loin, peeled tenderloin, top sirloin butt, bottom sirloin tri-tip, peeled knuckle, top inside round, bottom round flat, eye of round, heel and shank. Furthermore, ZH decreased ( $P \le 0.0036$ ) the total amount of bone and fat trim from the carcass. Zilpaterol hydrochloride increased ( $P \le 0.001$ ) Warner-Bratzler Shear force (WBS) values in strip loin steaks, regardless of aging period. Bottom round steaks aged for 14 and 21 d postmortem had increased (P < 0.004) cook loss as a result of ZH, and bottom round steaks aged for 21 d had increased (P = 0.003) that loss due to ZH. In top round steaks aged for 21 d, ZH increased ( $P \le 0.04$ ) thaw loss, percent thaw loss, and percent cook loss. Enhancement decreased ( $P \le 0.05$ ) percent cook loss in strip loin steaks aged

for both 14 and 21 d. Enhancement also decreased ( $P \le 0.005$ ) thaw loss and percent thaw loss in 21 d aged strip steaks. Shear force values were improved ( $P \le 0.001$ ) with enhancement in both 14 and 21 d aged strip steaks. Enhancement also improved (P < 0.0001) WBS values for top round steaks aged for 21 d. Percent cook loss was also improved ( $P \le 0.009$ ) with enhancement in top round steaks aged for 14 and 21 d. Therefore, ZH can improve carcass cutability of calf-fed Holstein steers; however, ZH can negatively impact tenderness. Enhancement improved tenderness and some cooking characteristics and may be used to improve tenderness in ZH fed animals.

Key Words: Zilpaterol hydrochloride, carcass cutability, tenderness, beef

### INTRODUCTION

Zilpaterol hydrochloride (ZH), commercially available as Zilmax® (Intervet/Schering Plough, Millsboro,DE), is a beta-agonist recently approved for use in the U.S. in beef finishing diets. Zilpaterol hydrochloride is similar in nature to the catecholamines and acts as a repartitioning agent similar to other  $\beta$ -AA. Zilpaterol hydrochloride is being marketed as a compound that can increase lean deposition and decrease fat accretion as well as improve animal growth performance characteristics much like the  $\beta$ -AA clenbuterol and cimaterol (Ricks et al., 1984; Moloney et al., 1990; Chikhou et al., 1993).

As reported by Shook et al. (2009), ZH can increase the amount of wholesale lean recovered from the carcass; however, a reduction in tenderness in strip loin steaks resulting from ZH supplementation is also documented. While ZH has been shown to increase the lean yield in fed beef steers, limited research is available as to the effects on carcass yields from calf-fed Holstein steers. Calf-fed Holstein steers comprise nearly 9% of the total beef harvest in the U.S. (Smith et al., 2006) and typically are very consistent at producing a high percentage of USDA Choice or better carcasses. However, calf-fed Holstein steers typically have poor muscling, low dressing percentages, and reduced fabrication yields when compared to beef steers. Supplementing calf-fed Holstein steers with ZH in the final finishing phase may result in greater improvements in carcass yields than seen in traditional beef steers.

Improvements in beef tenderness have been reported using needle injected enhancement solutions in beef (Robbins et al., 2002). A recent report indicated that 14% of whole-muscle beef cuts at the retail level have received some form of enhancement (National Cattlemen's Beef Association, 2007). The objectives of this experiment were to determine the effect of ZH on carcass cutability and tenderness from calf-fed Holstein steers as well as the impact of enhancement on products derived from calf-fed Holstein steers.

## MATERIAL AND METHODS

## Animals

The live phase of this experiment regarding blocking, penning, and animal selection are described in Beckett et al. (2009). Carcasses for this experiment were acquired from 4 different sources during 2 phases (2 sources/phase) at a large commercial feedlot in the desert Southwest U.S. from approximately 2300 calf-fed Holstein steers. Steers were either fed 8.3 mg/kg ZH (DM basis) for 20 d and were removed from the supplement 3 d prior to harvest; or fed a control (no ZH) diet. Phase 1 cattle were harvested on two separate days, control animals were harvested on Oct. 7, 2008, and ZH cattle were harvested on Oct. 8, 2008. For phase II, harvest was again on two separate

days with control animals harvested on Oct. 21, 2008, and ZH steers harvested on Oct. 22, 2008. After harvest and chilling, carcasses were ribbed at the  $12^{th}$  rib and carcass traits to determine USDA Quality (QG) and Yield Grades (YG) were recorded (USDA, 1997). After grading, carcasses were selected based on carcass weight and calculated yield grade. Carcasses were selected based on carcasses that were  $\pm 1$  SD (11.36 kg) from the mean hot carcass weight of the pen and to fit one of six predetermined USDA YG categories: YG < 1.99, YG 2 - 2.49, YG 2.5 - 2.99, YG 3 - 3.49, YG 3.5 - 3.99, and YG 4+ for both ZH supplemented and control cattle. Due to a lack of availability of carcass meeting some of the categories not all categories had equal representation; the obtained carcass distribution is presented in Table 4.1. This reduction from the desired number of carcass sides were transported 2,095 km via commercial refrigerated truck (0 to  $-2^{\circ}$ C) to the Robert M. Kerr Food and Agriculture Products Center (FAPC) for further fabrication.

# **Carcass Fabrication**

Once carcasses arrived at FAPC, they were stored in holding coolers  $(2^{\circ} \pm 2^{\circ}C)$ until fabrication. Cold side weights (CSW) were recorded prior to fabrication using a certified rail scale. Carcasses were then fabricated into subprimals according to the North American Meat Processors Association (NAMP) guidelines to include: 114C Chuck Shoulder Clod (0.635 cm trim), 114F Chuck Shoulder Tender, 116B Chuck (Mock) Tender (trimmed to blue), 130A Chuck Short Ribs (trimmed to blue), 116A Chuck Roll (0.635 cm trim), 109B Rib Blade Meat (trimmed to blue), 112A Ribeye Roll, Lip-on, 124 Rib Back Ribs, 120 Brisket Whole (packer trim), 115D Pectoral Meat (trimmed to blue),

121 Short Plate, 121C Outside Skirt (denuded), 121D Inside Skirt (denuded), 180 Strip Loin (0.635 cm trim), 189A Peeled Tenderloin, 184 Top Sirloin Butt (0.635 cm trim), 185B Bottom Sirloin Ball Tip (trimmed to blue), 185D Bottom Sirloin Tri-Tip (trimmed to blue), 167A Peeled Knuckle, 168 Top Inside Round (0.635 cm trim), 171B Bottom Round Flat (0.635 cm trim), 171C Eye of Round (0.635 cm trim), 185A Bottom Sirloin Flap (denuded), 171F Heel Meat, 193 Flank Steak (trimmed to blue), shank meat, and Elephant Ear (*cutaneous trunci* from the flank). Lean trimmings from all components were categorized into three categories: 90% lean/10% fat (90/10), 80% lean/20% fat (80/20), or 50% lean/50% fat (50/50) according to industry standard sorting techniques. Kidney knob fat, all trimmed fat, and all bones were also collected separately and weighed. After all weights from each side were recorded and entered, fabrication yield was calculated to ensure that 99% to 100.5% of CSW was recovered. Weights for all previously mentioned products of fabrication were also expressed as a percentage of CSW.

# Muscle Selection and Enhancement

Upon completion of fabrication, strip loins (NAMP 180; n = 54), top inside rounds (NAMP 168; n = 54), and bottom round flats (NAMP 171B; n = 54) from Phase 1 were selected for subsequent enhancement and fabricated for shear force analysis. Strip loins were fabricated by making a transverse cut through the center of the muscle, leaving two equal halves. Top inside rounds and bottom round flats were also fabricated by making a transverse cut across the muscle to yield two equal halves.

Within each subprimal, sections were allocated to either receive enhancement or to serve as the negative control by selecting alternation halves. The enhancement

solution was formulated using a proprietary solution (Hi-Grade Evaporated Salt, Cargill, Inc., Minneapolis, MN; Brifisol 750, BK Giulini Corp., Simi Valley, CA; Vivox Antioxidant, Vitiva, Cannes, France; Purasal HighPure P Plus, PURAC, Lincolnshire, IL; Proliant B1301 Beef Stock, Proliant, Ankeny, IA; water, ice) based on ongoing research at Oklahoma State University. The enhancement solution was mixed using chilled water as well as ice to maintain a temperature of  $2^{\circ} \pm 2^{\circ}$ C during injection. Muscle halves allocated to enhancement were injected (10% by weight; Famaco, Model FGM 20/20, Copenhagen, Denmark) and allowed to equilibrate for 15 min prior to steak fabrication.

# Steak Fabrication and Aging

For each muscle sample, two 2.54 cm steaks were cut from the medial face and randomly assigned one of two aging times (14 or 21 d). After fabrication, all steaks were individually vacuum packaged and aged for their respective time under refrigeration at  $2^{\circ} \pm 2^{\circ}$ C. After steaks had completed their appropriate aging time, there were frozen in a blast freezer (-30° ± 10°C) for 24 h, and then held in a subsequent freezer at -10° ± 2°C until further analysis.

# **Calculations**

Cook loss was determined by calculating the difference between thawed weight and cooked weight. After cooked weights were recorded, steaks were placed on trays and covered and then cooled at  $2^{\circ} \pm 2^{\circ}$ C for 18 to 24 h. Prior to coring for WBS, chilled weights were recorded and chill loss was determined by calculating the difference between cooked weight and chilled weight. Additionally, for each weight taken, percent loss compared to initial frozen weight was determined by taking the loss and dividing it by the initial frozen steak weight and then expressing the answer as a percentage.

### Warner Bratzler Shear Force

Warner-Bratzler shear force (WBS) was completed using the American Meat Science Association guidelines (1995), with modifications. Prior to thawing, frozen weights were recorded for all steaks while in the vacuum packaged bag. For all samples, frozen steaks were allowed to temper at  $2^{\circ} \pm 2^{\circ}C$  for 24 h prior to cooking. Prior to cooking, steaks were blotted free of any purge and thawed weights were recorded. Thaw loss was calculated by determering the difference in thawed weight and frozen weight. Steaks were broiled on an impingement oven (XLT Impinger, Model 3240-TS, BOFI Inc., Wichita, KS) at 200°C to an internal temperature of 68°C. An Atkins AccuTuff 340 thermometer (Atkins Temtec, Gainesville, FL) was used to measure the temperature of each steak as it exited the oven. If the steak had not yet reached 68°C, it was put back on the conveyor until it reached 68°C. All final internal temperatures were recorded, steaks were immediately weighed, and cooked weight was recorded. Six cores, 1.27 cm in diameter, were removed parallel to muscle fiber orientation and sheared once, using a Warner-Bratzler head attached to an Instron Universal Testing Machine (Model 4502, Instron Corporation, Canton, MS). The Warner-Bratzler head moved at a crosshead speed of 200 mm/min. Peak load (kg) of each core was recorded by an IBM PS2 (Model 55 SX) using software provided by the Instron Corporation. Peak load (kg) for all six cores was averaged and peak load (kg) was analyzed for each sample.

# Statistical Analysis

# Carcass Cutout Characteristics

Data were analyzed using the mixed model procedures of SAS (SAS Inst. Inc., Cary, NC). Analysis of variance for a completely randomized design with the fixed main

effect of ZH and random effect of individual carcass ID were included in the model. Carcass side was the experimental unit used for analysis. Least squares means were generated and separated using a pairwise t-test when the model displayed a treatment effect ( $\alpha = 0.05$ ). All data for Phase 1 and Phase 2 were combined for analysis. *Shear Force and Steak Characteristics* 

Shear force and steak characteristics data were analyzed using the mixed model procedure of SAS using a completely randomized design with the fixed main effects of ZH, enhancement and ZH × enhancement interaction. Location (anterior vs. posterior) within each whole muscle was included into the model as a random effect. All data were analyzed by aging time. Least squares means were generated and separated using a pairwise t-test when the model displayed a treatment effect ( $\alpha = 0.05$ ).

### **RESULTS AND DISCUSSION**

# **Carcass Cutout**

While carcasses from ZH steers had numerically greater cold side weights, total side weight was not impacted by ZH inclusion into the diet (P = 0.27) (Table 4.2). However, total wholesale carcass lean (total side weight minus 50/50 trim, 80/20 trim, 90/10 trim, kidney knob fat, total fat trim, and total bone) was increased (P < 0.0001) by nearly 5 kg and this increase resulted in a 2% increase in percent wholesale carcass lean when expressed on a carcass weight basis (Table 4.2). A significant reduction in total fat trim (P = 0.001) and total bone (P = 0.004) was also documented from carcasses of steers receiving ZH supplementation, resulting in a decrease in the percent of fat trim (P = 0.0003) and percent of bone (P < 0.0001) when expressed as a percent of carcass weight (Table 4.2). Moreover, there was no impact of ZH on any of the three lean trim levels measured. Finally, there was a tendency for ZH to decrease (P = 0.07) the amount of kidney knob fat; however, when expressed as a percentage of carcass weight, ZH supplementation significantly decreased (P = 0.03) kidney knob fat (Table 4.2). While in this study, carcass weight was not influenced by ZH supplementation, but the ratio in which lean, fat, and bone contributed to carcass weight was affected by ZH inclusion into the diet of calf-fed Holstein steers. The decrease in percent fat trim, bone, and kidney knob fat were counteracted by an increase in carcass lean, which resulted in similar carcass weights as compared to carcasses from control animals.

When comparing the subprimal yields from the forequarter, few differences due to ZH supplementation were observed. Shoulder clod weights increased (P = 0.0002) with ZH inclusion into the diet (Table 4.3). Also, the total yield of rib blade meat (P = 0.04) and inside skirt (P = 0.0002) increased with ZH inclusion into the diet (Table 4.3). However, the yield from rib back ribs (P = 0.04) was reduced in ZH fed steers (Table 4.3). When expressed as a percentage of carcass weight, ZH increased the percentage of the shoulder clod (P = 0.0001) and inside skirt (P = 0.0006), but reduced the percent yield in rib back ribs (P = 0.02) (Table 4.3). Zilpaterol also tended to increase (P = 0.07) the percentage of rib blade meat (Table 4.3).

In the hindquarter, numerous subprimals and muscles responded to ZH inclusion into the diet as recorded weights significantly increased. Weights increased for the strip loin (P < 0.0001), peeled tender (P < 0.0001), top sirloin butt (P = 0.01), bottom sirloin tri-tip (P < 0.0001), peeled knuckle (P = 0.001), top inside round (P < 0.0001), bottom round flat (P = 0.007), eye of round (P < 0.0001), heel meat (P < 0.0001), and shank meat (P < 0.0001) (Table 4.4). Moreover, when expressed as a percentage of carcass weight, the percent yield increased for strip loin (P < 0.0001), peeled tender (P < 0.0001), top sirloin butt (P = 0.04), bottom sirloin tri-tip (P = 0.0001), peeled knuckle (P = 0.001), top inside round (P = 0.0004), bottom round flat (P = 0.01), eye of round (P < 0.0001), heel meat (P = 0.0002), and shank meat (P = 0.0001) with ZH inclusion into the diet (Table 4.4).

In the present study, an increase in wholesale carcass lean was documented and is similar to results reported by Hilton (2009) where calf-fed Holstein steers had a 1% increase in red meat yield when supplemented with ZH. As reported by Shook et al. (2009), total bone as well as percent bone decreased in beef steers supplemented with ZH for 20 d; however; in the same study there was no impact of ZH supplementation on total fat trim or percent fat trim which is in contrast to the present study. Additionally, Shook et al. (2009) reported that ZH supplementation had no impact on 50/50, 80/20, or 90/10 trim levels, which is similar to the response seen in the current study with calf-fed Holstein steers. Similar to results summarized by Hilton (2009), as well as Shook et al. (2009), subprimals from the forequarter showed less of a response to ZH supplementation as compared to subprimals of the hindquarter. In the present study, only one subprimal of major significance in the forequarter responded to ZH as compared to seven major subprimals that responded to ZH from the hindquarter. As previously reported by Smith et al. (1995) type II muscle fibers show a greater response to beta-agonist stimulation, and therefore, muscles with a greater amount of type II fibers will have a greater response to beta-agonist supplementation. Kirchofer et al. (2002) previously reported that the muscles from the chuck have a wide variety of muscle fiber types, moreover, they documented that the muscles from the round are mostly comprised of type II (white)

muscle fibers. With a greater amount of type II fibers present in the round, it can be concluded that beta-agonist would have a greater influence on the muscles of the rounds as compared to the muscles of the chuck.

# Steak Characteristics and Shear Force

*14 d Aged Bottom Round Steaks.* Bottom round steaks aged for 14 d postmortem had a greater cook loss (P = 0.004) due to ZH supplementation; however, when this weight was expressed as a percentage of the original frozen steak weight, there were no differences in percent cook loss (P = 0.73) between ZH and control steaks (Table 4.5). Moreover, enhanced steaks tended to have reduced (P = 0.06) cook loss compared to non-enhanced steaks; however, there was no difference (P = 0.35) in enhanced vs. nonenhanced steaks when cook loss was expressed as a percentage of original frozen steak weight (Table 4.5). Finally, ZH inclusion into the diet had no impact (P = 0.21) on shear force values of bottom round steaks aged for 14 d; however, steaks that were enhanced tended to be more tender (P = 0.08) than steaks that were not enhanced (Table 4.5). However, regardless of treatment, mean WBS values exceeded the toughness threshold of  $\geq 4.90$  kg established by Miller et al. (2001).

21 d Aged Bottom Round Steaks. Bottom round steaks aged for 21 d had greater thaw loss (P = 0.003) and cook loss (P = 0.001) due to ZH supplementation; however, when thaw loss was expressed as a percentage of frozen steak weight, there were no differences (P = 0.39) between ZH and control steaks (Table 4.5). Steaks that were enhanced had less (P = 0.006) thaw loss when expressed as a percentage of steak weight; however, enhanced steak had a greater chill loss (P = 0.03) when compared to nonenhanced steaks (Table 4.5). While there was no interaction between diet and

enhancement for most steak characteristics, there was a significant interaction between diet and enhancement for percent cook loss (P = 0.002) (Table 4.6). Steaks from ZH supplemented animals responded greatest to enhancement as percent cook loss was lowest for these steaks, while percent cook loss was the greatest for non-enhanced steaks from ZH supplemented animals (Table 4.6). A significant interaction between diet and enhancement was also seen for 21 d shear force value (P = 0.04). While the interaction was significant, means were not statistically different (Table 4.6). Numerically, nonenhanced steaks from ZH fed animals had the greatest shear values while the lowest shear values were seen in non-enhanced steaks from control animals. However, the improvements due to enhancement were not enough to reduce WBS values below the tough threshold of  $\geq$  4.5 kg established by Miller et al. (2001) as mean WBS values, regardless of treatment, exceeded this value.

*14 d Aged Strip Loin Steaks*. Strip loin steaks aged for 14 d from ZH supplemented animals had more thaw loss (P = 0.01) and cook loss (P = 0.02) when compared to steaks from control animals (Table 4.7). When expressed as a percentage of original frozen steak weight, diet had no impact on percent thaw loss (P = 0.98) or percent cook loss (P = 0.32) (Table 4.7). Furthermore, percent cook loss (P = 0.05) was improved when steaks were enhanced (Table 4.7). Lastly, steaks from control animals were more tender (P < 0.001) when compared to steaks from ZH fed animals and enhanced steaks had lower shear force values (P < 0.001) when compared to non-enhanced steaks (Table 4.7). However, mean WBS values for steaks, regardless of treatment, were below the tenderness threshold of  $\leq 4.50$  kg established by Miller et al. (2001).

21 d Aged Strip Loin Steak. Strip loin steaks aged for 21 d tended (P = 0.06) to have less thaw loss when expressed as a percentage of original frozen steak weight due to ZH supplementation into the diet (Table 4.7). Additionally, steaks from ZH fed animals had greater cook loss (P < 0.0001), yet when expressed as a percentage of original steak weight, there was no impact (P = 0.31) due to ZH (Table 4.7). Moreover, percent chill loss (P = 0.06) tended to be greater in control steaks when compared to ZH steaks (Table 4.7). Thaw loss (P = 0.005) was greatest in non-enhanced steaks and resulted in nonenhanced steaks having a greater percent thaw loss (P = 0.0002) when compared to enhanced steaks (Table 4.7). Additionally, percent cook loss (P < 0.0001) was improved with steak enhancement (Table 4.7). Finally, steaks from control animals were more tender (P < 0.0001) than non-enhanced steaks (Table 4.7). Regardless of treatment, mean WBS values remained below  $\leq 4.50$  kg, which was established by Miller et al. (2001) as the upper limit for being considered tender.

14 d Aged Top Round Steaks. Chill loss (P = 0.03) and percent chill loss (P = 0.03) were greater in enhanced steaks as compared to non-enhanced steaks (Table 4.8). A significant interaction between diet and enhancement (P = 0.0002) was seen for thaw loss (Table 4.9). Steaks from control animals that were enhanced had less thaw loss than all other treatment groups (Table 4.9). Additionally, the diet by enhancement interaction was also significant for percent thaw loss (P = 0.003) (Table 4.9). Very similar to the amount thaw loss, enhanced steaks from control animals had the lowest percentage of thaw loss when compared to all others (Table 4.9). Moreover, the diet by enhancement interaction significantly (P = 0.04) impacted percent cook loss. Steaks from ZH supplemented animals that were enhanced had the lowest percentage cook loss when compared to all others (Table 4.9). Finally, the interaction between diet and enhancement remained significant (P = 0.008) for WBS values. Warner-Bratzler shear values were greatest in ZH steaks that had not been enhanced and were lowest in enhanced steaks from both ZH supplemented and control animals (Table 4.9).

21 d Aged Top Round Steaks. Steaks from ZH supplemented animals had a greater amount of thaw loss (P = 0.01) which resulted in an increase in the amount of weight lost due to thawing (P = 0.04) when expressed as a percentage of the original frozen steak weight (Table 4.8). Additionally, percent cook loss (P = 0.006) was decreased with ZH supplementation and percent cook loss (P = 0.008) was improved with enhancement (Table 4.8). Finally, steaks from ZH supplemented animals tended to be tougher (P = 0.10) when compared to steaks from control animals and steaks that were enhanced were more tender (P < 0.0001) when compared to non-enhanced steaks (Table 4.8). Additionally, steaks that were enhanced had the only mean WBS value that was below the  $\leq 4.50$  kg threshold determined by Miller et al. (2001) which distinguished tender from intermediately tough.

As expected, tenderness decreased in steaks from animals supplemented with ZH. This is similar to many studies where ZH supplementation has resulted in increased WBS values as well as reduced consumer scores for tenderness in strip loin steaks (Brooks and Miller, 2009; Shook et al., 2009). In the present study, while mean WBS values for strip loin steaks were below the tenderness threshold, 20.3% of the 14 d aged steak and 6.5% of the 21 d aged steaks exceeded the  $\leq$  4.50 kg threshold. Of the 14 d aged steaks that exceed the  $\leq$  4.50 threshold, 50% were from animals supplemented with ZH while 85.7%

of the 21 d aged steaks exceeding  $\leq 4.50$  kg were from ZH fed animals. Zilpaterol is known to cause muscle hypertrophy which correlates to an increase in the diameter of muscle fibers (Mills, 2002). This increase in muscle fiber diameter can lead to a subsequent decrease in tenderness from both an objective and subjective standpoint. Moreover, Mills (2002) indicates that  $\beta$ -AA alter muscle metabolism to favor a greater proportion of fast-twitch muscle due to altered protein metabolism and increased blood flow to skeletal muscles. This increase in fast-twitch muscle fibers could also lead to decreases in tenderness as Seideman and Theer (1986) documented a positive correlation of percent white fiber number and area with sensory tenderness ratings. Additionally, the improvements in tenderness seen when steaks were enhanced is similar to results shown by Robbins et al. (2002) where both objective and subjective tenderness measurements indicated that enhancement improved the tenderness of beef steaks and roasts. However, Lawrence et al. (2004) evaluated enhancement on loin muscles with a phosphate and salt solution or a calcium lactate plus beef broth solution or carrageenan with rosemary extract solution and found there to be no difference in WBS between any of the treatments. Steaks tested on the same day that they were enhanced had improved WBS values when compared to non-enhanced controls which indicates that enhancement has an immediate effect on tenderness through either a dilution effect or through a physical disruption of the muscle via the injection needles (Grobbel et al., 2008). In the present study, when combining the total number of strip loin steaks aged for 14 and 21 d, 29 steaks out of 216 exceeded the tender threshold of  $\leq$  4.50 kg, however, only 2 of those steaks were from enhanced strip loins.

Cannon et al. (1993) reported improvements in cook loss when using a phosphate enhancement which is in agreement with the current study where cook loss was improved with enhancement. Phosphates increase the pH of meat and allow for greater retention of water due to a shift away from the isoelectric point. In contrast to enhancement, ZH supplementation had an opposite effect on cook loss yet, this increase in loss is not seen when cook loss is expressed as a percentage of the initial frozen steak weight. This is perhaps due to the steaks from ZH fed animals being larger and heavier than those from control animals and therefore a greater amount of moisture stands to be lost.

In conclusion, ZH, when fed for the last 20 d of the finishing phase at 8.3 mg/kg (DM basis), had a positive impact on carcass cutability in calf-fed Holstein steers, especially noting the significant increase in lean muscle accretion in the hindquarter of the animal. Furthermore, ZH supplementation during the final portion of the finishing phase had a negative impact on tenderness; however, this tenderness difference does not adversely effect the classification (tender or tough) given to each steak. Moreover, this study revealed that enhancement has a positive effect on tenderness, as well as cook loss. In future studies, the interaction of new technologies used for improving beef tenderness postmortem should be evaluated to determine their effects on improving beef tenderness in animals supplemented with  $\beta$ -AA. Learning to counteract the negative impacts on tenderness seen when using  $\beta$ -AA will allow the beef industry to take full advantage of the improvements in beef production from utilizing these products in the finishing diets of beef cattle.

|             | Р  | hase I<br>ZH <sup>2</sup> |    | se II<br>H <sup>2</sup> |
|-------------|----|---------------------------|----|-------------------------|
| Yield Grade | 0  | 8.3 mg/kg                 | 0  | 8.3 mg/kg               |
| < 1.99      | 1  | 4                         | 4  | 6                       |
| 2.0 - 2.49  | 4  | 11                        | 5  | 6                       |
| 2.5 - 2.99  | 7  | 6                         | 4  | 5                       |
| 3.0 - 3.49  | 5  | 8                         | 6  | 5                       |
| 3.5 – 3.99  | 7  | 0                         | 4  | 3                       |
| > 4.0       | 1  | 0                         | 0  | 0                       |
| Total       | 25 | 29                        | 23 | 25                      |

**Table 4.1.** Number of carcasses selected per treatment within each phase<sup>1</sup> based on yield grade.

<sup>1</sup>Phase 1 = Control harvested on 10-7-08; ZH harvested on 10-8-08. Phase 2 = Control harvested on 10-21-08; ZH harvested on 10-22-08. <sup>2</sup>ZH = Zilpaterol hydrochloride

| Trait                                   | 0      | 8.3mg/kg | Pr > F   | SEM <sup>3</sup> |
|---|--------|----------|----------|------------------|
| Total Side Weight, kg                   | 171.55 | 173.26   | 0.27     | 1.10             |
| Wholesale Carcass Lean, kg <sup>3</sup> | 88.55  | 93.01    | < 0.0001 | 0.78             |
| Wholesale Carcass Lean, %               | 51.53  | 53.57    | < 0.0001 | 0.08             |
| 50/50 Trim, kg                          | 11.80  | 11.35    | 0.55     | 0.54             |
| 50/50 Trim, %                           | 6.87   | 6.53     | 0.41     | 0.14             |
| 80/20 Trim, kg                          | 3.02   | 3.42     | 0.18     | 0.22             |
| 80/20 Trim, %                           | 1.76   | 1.98     | 0.13     | 0.06             |
| 90/10 Trim kg                           | 12.09  | 12.63    | 0.24     | 0.33             |
| 90/10 Trim, %                           | 7.04   | 7.30     | 0.32     | 0.09             |
| Kidney Knob Fat, kg                     | 6.48   | 6.04     | 0.07     | 0.17             |
| Kidney Knob Fat, %                      | 3.77   | 3.48     | 0.03     | 0.05             |
| Total Fat Trim, kg                      | 13.10  | 11.49    | 0.0014   | 0.35             |
| Total Fat Trim, %                       | 7.62   | 6.62     | 0.0003   | 0.09             |
| Total Bone, kg                          | 36.70  | 35.53    | 0.0036   | 0.28             |
| Total Bone, %                           | 21.40  | 20.52    | < 0.0001 | 0.06             |

**Table 4.2.** Effects of zilpaterol hydrochloride inclusion into the diet on the amount and percentage<sup>1</sup> of carcass cutout of calf-fed Holstein steers (n = 102).

<sup>1</sup>% Listed as a percentage of cold side weight. <sup>2</sup>ZH = Zilpaterol hydrochloride <sup>3</sup> total side weight minus 50/50 trim, 80/20 trim, 90/10 trim, kidney knob fat, total fat trim, and total bone.

**Table 4.3.** Effects of ZH inclusion into the diet prior to slaughter on the amount and percentage<sup>1</sup> of various wholesale beef cuts from the forequarter of calf-fed Holstein steers (n =102).

|                                     |         | $ZH^2$    |        |      |
|-------------------------------------|---------|-----------|--------|------|
| Item                                | 0 mg/kg | 8.3 mg/kg | Pr > F | SEM  |
| Shoulder Clod, trimmed, kg          | 8.70    | 9.30      | 0.0002 | 0.11 |
| Shoulder Clod trimmed, %            | 2.30    | 2.44      | 0.0001 | 0.02 |
| Chuck Shoulder Tender, kg           | 0.42    | 0.44      | 0.26   | 0.01 |
| Chuck Shoulder Tender, %            | 0.11    | 0.12      | 0.42   | 0.01 |
| Chuck Roll, kg                      | 8.06    | 8.12      | 0.70   | 0.12 |
| Chuck Roll, %                       | 2.13    | 2.13      | 0.90   | 0.03 |
| Chuck Mock Tender, kg               | 1.49    | 1.55      | 0.14   | 0.03 |
| Chuck Mock Tender, %                | 0.39    | 0.40      | 0.22   | 0.01 |
| Chuck Short Ribs, kg                | 1.57    | 1.62      | 0.45   | 0.05 |
| Chuck Short Ribs, %                 | 0.41    | 0.42      | 0.53   | 0.01 |
| Pectoral Meat, trimmed to blue, kg  | 0.62    | 0.66      | 0.13   | 0.02 |
| Pectoral Meat, trimmed to blue, %   | 0.16    | 0.17      | 0.33   | 0.01 |
| Rib Blade Meat, kg                  | 1.34    | 1.43      | 0.04   | 0.03 |
| Rib Blade Meat, %                   | 0.35    | 0.38      | 0.07   | 0.01 |
| Ribeye Roll, kg                     | 5.19    | 5.24      | 0.57   | 0.06 |
| Ribeye Roll, %                      | 1.37    | 1.37      | 0.98   | 0.01 |
| Rib Back Ribs, kg                   | 1.47    | 1.38      | 0.04   | 0.03 |
| Rib Back Ribs, %                    | 0.39    | 0.36      | 0.02   | 0.01 |
| Plate, kg                           | 8.49    | 8.69      | 0.41   | 0.18 |
| Plate, %                            | 2.24    | 2.27      | 0.59   | 0.04 |
| Brisket Whole, bnls packer trim, kg | 4.48    | 4.67      | 0.14   | 0.09 |
| Brisket Whole, bnls packer trim, %  | 1.18    | 1.22      | 0.20   | 0.02 |
| Inside Skirt, kg                    | 1.08    | 1.20      | 0.0002 | 0.02 |
| Inside Skirt, %                     | 0.29    | 0.31      | 0.0006 | 0.01 |
| Outside Skirt, kg                   | 0.68    | 0.68      | 0.74   | 0.02 |
| Outside Skirt, %                    | 0.18    | 0.18      | 0.70   | 0.01 |

<sup>1</sup>% Listed as a percentage of cold side weight. <sup>2</sup>ZH = Zilpaterol hydrochloride

|                                      |         | $ZH^2$    |          |      |
|--------------------------------------|---------|-----------|----------|------|
| Item                                 | 0 mg/kg | 8.3 mg/kg | Pr > F   | SEM  |
| Strip Loin, kg                       | 4.09    | 4.50      | < 0.001  | 0.05 |
| Strip Loin, %                        | 1.08    | 1.18      | < 0.0001 | 0.01 |
| Peeled Tender, side muscle on, kg    | 2.54    | 2.83      | < 0.0001 | 0.03 |
| Peeled Tender, side muscle on, %     | 0.68    | 0.74      | < 0.0001 | 0.01 |
| Top Sirloin Butt, kg                 | 5.49    | 5.77      | 0.01     | 0.07 |
| Top Sirloin Butt, %                  | 1.46    | 1.51      | 0.04     | 0.02 |
| Bottom Sirloin Ball-Tip, denuded, kg | 0.49    | 0.53      | 0.45     | 0.03 |
| Bottom Sirloin Ball-Tip, denuded, %  | 0.13    | 0.14      | 0.53     | 0.01 |
| Bottom Sirloin Tri-Tip, denuded, kg  | 0.86    | 0.95      | < 0.0001 | 0.01 |
| Bottom Sirloin Tri-Tip, denuded, %   | 0.23    | 0.25      | 0.0001   | 0.01 |
| Bottom Sirloin Flap, denuded, kg     | 1.50    | 1.51      | 0.89     | 0.03 |
| Bottom Sirloin Flap, denuded, %      | 0.40    | 0.39      | 0.82     | 0.01 |
| Knuckle, peeled, kg                  | 5.21    | 5.55      | < 0.0001 | 0.06 |
| Knuckle, peeled, %                   | 1.38    | 1.45      | 0.001    | 0.01 |
| Top Inside Round, kg                 | 9.28    | 9.99      | < 0.0001 | 0.11 |
| Top Inside Round, %                  | 2.46    | 2.62      | 0.0004   | 0.03 |
| Bottom Round Flat, kg                | 6.16    | 6.53      | 0.0070   | 0.10 |
| Bottom Round Flat, %                 | 1.63    | 1.71      | 0.01     | 0.02 |
| Eye of Round, kg                     | 2.15    | 2.39      | < 0.0001 | 0.02 |
| Eye of Round, %                      | 0.57    | 0.63      | < 0.0001 | 0.01 |
| Heel Meat, kg                        | 2.20    | 2.36      | < 0.0001 | 0.02 |
| Heel Meat, %                         | 0.58    | 0.62      | 0.0002   | 0.01 |
| Shank Meat, kg                       | 2.40    | 2.59      | < 0.0001 | 0.03 |
| Shank Meat, %                        | 0.64    | 0.68      | 0.0001   | 0.01 |
| Flank Steak, kg                      | 0.90    | 0.92      | 0.41     | 0.02 |
| Flank Steak, %                       | 0.24    | 0.24      | 0.58     | 0.01 |
| Elephant Ear, kg                     | 1.25    | 1.19      | 0.38     | 0.05 |
| Elephant Ear, %                      | 0.33    | 0.31      | 0.28     | 0.01 |

**Table 4.4.** Effects of ZH inclusion into the diet prior to slaughter on the amount and percentage<sup>1</sup> of various wholesale beef cuts from the hindquarter of calf-fed Holstein steers (n = 102).

<sup>1</sup>% Listed as a percentage of cold side weight. <sup>2</sup>ZH = Zilpaterol hydrochloride

|                            | $ZH^1$ |          | Er     | hancement | 2      |        |                  |
|----------------------------|--------|----------|--------|-----------|--------|--------|------------------|
| Trait                      | 0      | 8.3mg/kg | Pr > F | Е         | NE     | Pr > F | SEM <sup>3</sup> |
| 14 d aging                 |        |          |        |           |        |        |                  |
| Thaw Loss, g               | 29.85  | 30.50    | 0.75   | 29.46     | 30.88  | 0.49   | 1.46             |
| Thaw Loss, % <sup>4</sup>  | 6.86   | 6.72     | 0.76   | 6.55      | 7.03   | 0.32   | 0.34             |
| Cook Loss, g               | 99.14  | 111.17   | 0.004  | 101.20    | 109.11 | 0.06   | 2.93             |
| Cook Loss, % <sup>4</sup>  | 22.84  | 24.34    | 0.09   | 22.39     | 24.78  | 0.008  | 0.63             |
| Chill Loss, g              | 9.42   | 9.94     | 0.58   | 10.46     | 8.90   | 0.11   | 1.63             |
| Chill Loss, % <sup>4</sup> | 2.13   | 2.04     | 0.80   | 2.26      | 2.04   | 0.27   | 0.31             |
| WBS, kg                    | 7.18   | 7.59     | 0.21   | 7.09      | 7.69   | 0.08   | 0.69             |
| 21 d aging                 |        |          |        |           |        |        |                  |
| Thaw Loss, g               | 34.89  | 38.82    | 0.003  | 36.20     | 37.52  | 0.31   | 0.94             |
| Thaw Loss, % <sup>4</sup>  | 8.02   | 8.24     | 0.39   | 7.77      | 8.50   | 0.006  | 0.19             |
| Cook Loss, g               | 94.51  | 100.92   | 0.001  | 97.22     | 98.20  | 0.61   | 1.40             |
| Chill Loss, g              | 7.74   | 8.22     | 0.49   | 8.71      | 7.24   | 0.03   | 0.59             |
| Chill Loss, % <sup>4</sup> | 1.77   | 1.72     | 0.78   | 1.86      | 1.64   | 0.13   | 0.12             |

**Table 4.5.** Effects of zilpaterol hydrochloride inclusion into the diet and enhancement on cook
 characteristics and tenderness of bottom round steaks (n = 54).

 $\frac{\text{CHIII LOSS, 70}}{^{1}\text{ZH} = \text{Zilpaterol hydrochloride}}$   $^{2}\text{E} = \text{Enhanced, NE} = \text{Non-enhanced}$   $^{3}\text{Standard Error of the Mean for ZH/Enhancement}$   $^{4}\text{Listed as a percentage of frozen steak weight.}$ 

|                           |                     | $ZH^{2}$           |            |      |
|---------------------------|---------------------|--------------------|------------|------|
| Trait                     | 0                   | 8.3mg/kg           | $Pr > F^3$ | SEM  |
| Cook Loss, % <sup>4</sup> |                     |                    |            |      |
| Е                         | 21.61 <sup>b</sup>  | 20.12 <sup>c</sup> | 0.002      | 0.37 |
| NE                        | 21.94 <sup>ab</sup> | 22.81 <sup>a</sup> |            |      |
| WBS, kg <sup>*</sup>      |                     |                    |            |      |
| E                         | 7.23                | 6.74               | 0.04       | 1.14 |
| NE                        | 6.65                | 7.48               |            |      |
|                           |                     |                    |            |      |

**Table 4.6.** Interaction of zilpaterol hydrochloride inclusion into the diet and enhancement<sup>1</sup> on cook characteristics and tenderness of bottom round steaks aged for 21 days (n = 54).

<sup>a, b, c</sup> Within a main effect means with different superscripts differ (P < 0.05).

<sup>1</sup> E = Enhanced, NE = Non-enhanced

<sup>2</sup>ZH = Zilpaterol hydrochloride
<sup>3</sup> Probability of an interaction between ZH and enhancement.
<sup>4</sup> Listed as a percentage of frozen steak weight.
\* Using LSMEANS separation technique there was no significant difference in means although the model indicated a significant interaction between ZH  $\times$  enhancement (P < 0.05).

|                            | $ZH^{2}$ |          | Enhancement <sup>1</sup> |       |       |          |                  |
|----------------------------|----------|----------|--------------------------|-------|-------|----------|------------------|
| Trait                      | 0        | 8.3mg/kg | Pr > F                   | E     | NE    | Pr > F   | SEM <sup>3</sup> |
| 14 d aging                 |          | 00       |                          |       |       |          |                  |
| Thaw Loss, g               | 20.00    | 21.64    | 0.01                     | 20.61 | 21.04 | 0.50     | 0.53             |
| Thaw Loss, % <sup>4</sup>  | 6.76     | 6.93     | 0.98                     | 6.60  | 6.93  | 0.14     | 0.16             |
| Cook Loss, g               | 69.68    | 75.50    | 0.02                     | 72.21 | 72.90 | 0.77     | 7.76             |
| Cook Loss, % <sup>4</sup>  | 23.14    | 23.81    | 0.32                     | 22.78 | 24.17 | 0.05     | 0.56             |
| Chill Loss, g              | 4.77     | 4.20     | 0.36                     | 4.59  | 4.38  | 0.74     | 0.44             |
| Chill Loss, % <sup>4</sup> | 1.57     | 1.31     | 0.18                     | 1.45  | 1.42  | 0.88     | 0.14             |
| WBS, kg                    | 2.85     | 3.73     | < 0.001                  | 2.73  | 3.86  | < 0.001  | 0.12             |
| 21 d aging                 |          |          |                          |       |       |          |                  |
| Thaw Loss, g               | 21.00    | 19.28    | 0.51                     | 19.58 | 21.88 | 0.005    | 0.56             |
| Thaw Loss, % <sup>4</sup>  | 7.24     | 6.74     | 0.06                     | 6.47  | 7.52  | 0.0002   | 0.30             |
| Cook Loss, g               | 58.31    | 64.60    | < 0.0001                 | 60.34 | 62.63 | 0.12     | 1.25             |
| Cook Loss, % <sup>4</sup>  | 20.43    | 20.77    | 0.31                     | 19.60 | 21.60 | < 0.0001 | 0.24             |
| Chill Loss, g              | 5.04     | 4.96     | 0.83                     | 5.13  | 4.89  | 0.48     | 0.42             |
| Chill Loss, % <sup>4</sup> | 1.78     | 1.57     | 0.06                     | 1.68  | 1.67  | 0.93     | 0.11             |
| WBS, kg                    | 2.82     | 3.39     | < 0.0001                 | 2.64  | 3.58  | < 0.0001 | 0.10             |

**Table 4.7.** Effects of zilpaterol hydrochloride inclusion into the diet and enhancement on cook
 characteristics and tenderness of strip loin steaks (n = 54).

 $\frac{2.82}{1} = \frac{2.82}{5.5}$   $\frac{1}{1} = Enhanced, NE = Non-enhanced$   $\frac{1}{2} ZH = Zilpaterol hydrochloride$   $\frac{3}{5} Standard Error of the Mean for ZH/Enhancement$   $\frac{4}{1} Listed as a percentage of frozen steak weight.$ 

|                            | $ZH^2$ |          |        | E      | Enhancemer | nt <sup>1</sup> |                  |
|----------------------------|--------|----------|--------|--------|------------|-----------------|------------------|
| Trait                      | 0      | 8.3mg/kg | Pr > F | Е      | NE         | Pr > F          | SEM <sup>3</sup> |
| 14 d aging                 |        |          |        |        |            |                 |                  |
| Cook Loss, g               | 124.87 | 131.46   | 0.06   | 127.24 | 129.09     | 0.59            | 4.27             |
| Chill Loss, g              | 6.80   | 7.53     | 0.29   | 8.06   | 6.27       | 0.03            | 0.59             |
| Chill Loss, % <sup>4</sup> | 1.38   | 1.38     | 0.99   | 1.52   | 1.24       | 0.03            | 0.09             |
| 21 d aging                 |        |          |        |        |            |                 |                  |
| Thaw Loss, g               | 41.82  | 48.43    | 0.01   | 45.12  | 45.13      | 0.99            | 1.86             |
| Thaw Loss, % <sup>4</sup>  | 8.58   | 9.56     | 0.04   | 8.99   | 9.15       | 0.74            | 0.45             |
| Cook Loss, g               | 130.62 | 127.08   | 0.41   | 127.79 | 129.91     | 0.62            | 3.07             |
| Cook Loss, % <sup>4</sup>  | 26.61  | 25.12    | 0.006  | 25.15  | 26.58      | 0.008           | 0.38             |
| Chill Loss, g              | 8.70   | 9.76     | 0.37   | 9.32   | 9.13       | 0.87            | 0.86             |
| Chill Loss, % <sup>4</sup> | 1.78   | 1.97     | 0.48   | 1.86   | 1.89       | 0.93            | 0.19             |
| WBS, kg                    | 4.39   | 4.67     | 0.10   | 3.95   | 5.11       | < 0.0001        | 0.13             |

Table 4.8. Effects of zilpaterol hydrochloride inclusion into the diet and enhancement on cook characteristics and tenderness of top round steaks (n = 54).

<sup>1</sup> E = Enhanced, NE = Non-enhanced <sup>2</sup>ZH = Zilpaterol hydrochloride <sup>3</sup>Standard Error of the Mean for ZH/Enhancement <sup>4</sup>Listed as a percentage of frozen steak weight.

|  |                       | $ZH^2$                    |            |      |
|--|-----------------------|---------------------------|------------|------|
| Trait                                  | 0                     | 8.3mg/kg                  | $Pr > F^3$ | SEM  |
| Thaw Loss, g                           |                       |                           |            |      |
| E                                      | 38.03 <sup>b</sup>    | 59.53 <sup>a</sup>        | 0.0002     | 2.85 |
| NE                                     | 53.74 <sup>a</sup>    | 55.62 <sup>a</sup>        |            |      |
| Thaw Loss, % <sup>4</sup>              |                       |                           |            |      |
| Е                                      | 7.81 <sup>b</sup>     | $10.70^{a}$               | 0.003      | 0.72 |
| NE                                     | $10.84^{a}$           | $10.95^{a}$               |            |      |
| Cook Loss, % <sup>4</sup>              |                       |                           |            |      |
| Е                                      | $24.97^{a}$           | $23.56^{b}$               | 0.04       | 0.40 |
| NE                                     | 25.68 <sup>a</sup>    | 25.93 <sup>a</sup>        |            |      |
| WBS, kg                                |                       |                           |            |      |
| Ē                                      | $4.05^{\circ}$        | $4.23^{\circ}$            | 0.008      | 0.26 |
| NE                                     | $5.07^{\rm b}$        | $6.22^{a}$                |            |      |
| <sup>a, b, c</sup> Within a main effec | ct means with differe | nt superscripts differ (P | < 0.05).   |      |
| $^{1}$ E = Enhanced, NE = N            |                       |                           |            |      |
| ${}^{2}ZH = Zilpaterol hydrod$         |                       |                           |            |      |
| <sup>3</sup> Probability of an intera  | action between ZH and | nd enhancement            |            |      |
| <sup>4</sup> Listed as a percentage    | of frozen steak weigh | nt.                       |            |      |

**Table 4.9.** Interaction of zilpaterol hydrochloride inclusion into the diet and enhancement<sup>1</sup> on cook characteristics and tenderness of top round steaks aged for 14 days (n = 54).

#### CHAPTER V

## CONCLUSION

Results from these studies indicate that ZH, when fed to beef steers as well as calf-fed Holstein steers at 8.3mg/kg (DM basis) during the final 20 d of the feeding period, can improve the percentage of red meat yield recovered during fabrication. Numerous primals, mostly from the hindquarter of the animal, have increased weight due to ZH supplementation. Moreover, feeding ZH can reduce the amount of fat and bone waste collected during fabrication. The difference in response to ZH seen within location of the carcass is mostly likely due to the proportion of type II muscle fibers to type I muscle fibers. Type II muscle fibers have a greater response to ZH and are more abundant in the round of the animal when compared to the chuck, and for these reasons a greater increase in muscle is seen in the hindquarter. This increase in lean can lead to greater profits for beef processors per pound of carcass. Additionally, proximate analysis data indicate that ZH supplementation can result in a greater amount of protein in beef trimming recovered during fabrication. This increase in protein is accompanied by a decrease in moisture and fat within the trimming. Increased protein in lean trimming can result in improved profits for beef processors as lean trimmings have a higher market value as compared to fat trimmings. Although ZH decreased tenderness in bottom round, strip loin, and top round steaks, all steaks from the strip loin and top round, regardless of dietary treatment, were below the threshold for being considered tender. Additionally, all steaks from the bottom round were above the threshold for being considered tough, regardless of dietary treatment. Decreased tenderness is a common

negative associated with the use of  $\beta$ -agonist in animal production; however, in this study, the effects on tenderness, while significant, did not alter the acceptability of the final product. While the impact on tenderness should be considered, more emphasis should be placed on impact of  $\beta$ -AA on subjective tenderness rather than objective tenderness. While objective test may determine a change in tenderness, this change may not necessarily be large enough for average consumer to detect.

In this study, enhancement improved tenderness in all three steaks as well as improved the amount of weight lost during cooking. Enhancement technology has been utilized in the poultry and swine industry for a number of years to increase the water holding activity, moisture, flavor and overall acceptability to consumers; however, this technology is only being used in roughly 14% of the fresh beef sold at retail (National Cattlemen's Beef Association, 2007). With the improvement enhancement can bring in tenderness and moisture retention during cooking, enhancement stands to allow retailers of lower quality beef an opportunity to increase the quality and consumer acceptability of their product. Allowing lower quality beef to perform more closely to high quality beef could result in large profit increases for beef processors and retailers. Moreover, enhancement may allow beef producers to utilize technologies, such as  $\beta$ -agonist, more often with little hesitation due to the potential negative impacts on tenderness so often associated with  $\beta$ -AA.

In conclusion, beta-agonists had a positive effect on the amount of wholesale lean recovered from beef carcass from both beef and calf-fed Holsteins as well as reduced waste from bone and fat. Additionally, tenderness was significantly decreased due to beta-agonist; however, this change was not enough to classify the steaks as tough.

Furthermore, enhancement improved the tenderness as well as the cook loss in beef steaks. Therefore, the combined utilization of  $\beta$ -agonist and enhancement can allow the beef industry to more efficiently produce beef with little concern for the negative impacts that could result due to beta-agonist supplementation.

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## VITA

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Pages in Study: 87

Candidate for the Degree of Doctor of Philosophy

Major Field: Food Science

Scope and Method of Study: Beta-agonists are compounds similar to the natural catecholamines found in the body and are currently being used to improve the efficiency of gain and lean production in livestock. Zilpaterol hydrochloride (ZH) (Intervet/Schering Plough, Millsboro, DE) is the most recently approved beta-agonist for use in the U.S. beef cattle industry. Zilpaterol hydrochloride is marketed as a compound that will improve the growth efficiency, lean production, and carcass weight of fed cattle. The objectives of this study were to determine the impact of ZH on carcass cutability and tenderness of beef steers as well as calf-fed Holstein steers. Beef carcasses (n = 127) and calf-fed Holstein carcasses (n = 102) were selected from cattle fed either 8.3 mg/kg/d (DM basis) or 0 mg/kg of ZH during the final 20 d of the finishing period. Carcasses were fabricated into subprimals according to the National Meat Processors Association (NAMP). All steaks were randomly assigned to either 14 or 21 d aging period prior to WBS determination. Strip loins from beef steers and strip loins, bottom rounds, and top rounds from the calf-fed Holstein steers were collected for Warner-Bratzler shear force analysis (WBS). Subprimals collected from calf-fed Holstein steers were assigned to control or enhancement treatment. Thaw loss, cook loss, and chill loss were recorded during the cooking of steaks for WBS determination.

Findings and Conclusions: Wholesale carcass lean was increased (P < 0.001) from both beef and calf-fed Holsteins supplemented with ZH. Furthermore, in both sets of steers, subprimals from the hindquarter seemed to have a greater response to ZH treatment than subprimals from the forequarter. Zilpaterol hydrochloride also decreased (P < 0.01) the amount of total bone and fat trim in calf-fed Holsteins and decreased (P = 0.05) total bone in beef steers. Tenderness was decreased (P < 0.05) in all steaks tested from ZH supplemented carcasses when compared to control carcasses. However, enhancement improved (P < 0.05) tenderness in strip loin, bottom round, and top round steaks when compared to non-enhanced steaks. Additionally, enhanced strip loin and top round steaks. These results indicated that ZH is very effective at improving lean carcass yields in both beef and calf-fed Holstein steers. Furthermore, ZH has a negative impact on tenderness, but, enhancement improved beef tenderness and may be used to address tenderness issue in animals fed ZH.

ADVISER'S APPROVAL: Deb VanOverbeke