

PERFORMANCE, CARCASS, MEAT SENSORY AND ENDOCRINE  
TRAITS OF YOUNG BULLS AND STEERS IMPLANTED  
WITH TRENBOLONE ACETATE AND ZERANOL

by

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## DEDICATION

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This thesis is dedicated to my parents, Dean and Priscilla Johnson. It is through their love and support that I have been able to achieve this and previous goals. Also I would like to acknowledge my grandparents, Anna Arett along with Milton and Evelyn Johnson for standing behind me in pursuing this degree.

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## TABLE OF CONTENTS

<u>Chapter</u>	<u>Page</u>
DEDICATION . . . . .	ii
ACKNOWLEDGEMENTS . . . . .	iii
LIST OF TABLES . . . . .	vi
LIST OF FIGURES. . . . .	vii
I GENERAL INTRODUCTION. . . . .	1
II GENERAL REVIEW OF LITERATURE. . . . .	4
Bulls versus Steers . . . . .	4
Live Performance . . . . .	4
Pre-Slaughter Management . . . . .	5
Slaughter and Fabrication . . . . .	6
Carcass Cutability . . . . .	8
Carcass Quality. . . . .	9
Palatability. . . . .	10
Hormones Related to Postnatal Growth . . . . .	12
Modes of Receptivity. . . . .	12
Growth Hormone, Somatomedins and Insulin . . . . .	14
Thyroid Hormones . . . . .	16
Glucocorticoids. . . . .	17
Androgens . . . . .	18
Estrogens. . . . .	19
Anabolic Growth Promotants. . . . .	21
Response by Different Sexes. . . . .	21
Effects of Anabolic Agents on Bull and Steer. . . . .	23
Performance, Carcass and Palatability Traits	
Literature Cited. . . . .	29
III PERFORMANCE, CARCASS, MEAT SENSORY AND ENDOCRINE TRAITS OF YOUNG BULLS AND STEERS IMPLANTED WITH TRENBOLONE ACETATE AND ZERANOL. . . . .	41
Abstract . . . . .	41
Introduction. . . . .	43

	Materials and Methods. . . . .	45
	Management and Live Animal Measurements . . . . .	45
	Blood Collection and Analyses . . . . .	47
	Slaughter, Carcass and Meat Sensory Traits . . . . .	48
	Statistical Analyses . . . . .	49
	Results and Discussion. . . . .	50
	Animal Performance. . . . .	50
	Animal Measurements. . . . .	50
	Hormonal Concentrations. . . . .	54
	Carcass Characteristics. . . . .	59
	Longissimus Sensory Traits . . . . .	63
	Conclusions. . . . .	66
	Literature Cited. . . . .	67
IV	Appendices . . . . .	71
V	Abstract . . . . .	80

## LIST OF TABLES

<u>Table</u>		<u>Page</u>
1	GROWTH PROMOTING IMPLANTS . . . . .	24
2	DIET COMPOSITION FOR CONTROL BULLS AND TRENBOLONE ACETATE PLUS ZERANOL IMPLANTED BULLS AND STEERS.	46
3	WEIGHTS AND PERFORMANCE CHARACTERISTICS OF CONTROL BULLS AND TRENBOLONE ACETATE PLUS ZERANOL IMPLANTED BULLS AND STEERS. . . . .	51
4	HIP HEIGHTS AND MASCULINITY CHARACTERISTICS OF CONTROL BULLS AND TRENBOLONE ACETATE PLUS ZERANOL IMPLANTED BULLS AND STEERS. . . . .	52
5	CARCASS CHARACTERISTICS OF CONTROL BULLS AND TRENBOLONE ACETATE PLUS ZERANOL IMPLANTED BULLS AND STEERS. . . . .	60
6	LONGISSIMUS QUALITY CHARACTERISTICS OF CONTROL BULLS AND TRENBOLONE ACETATE PLUS ZERANOL IMPLANTED BULLS AND STEERS. . . . .	64
7	TASTE PANEL EVALUATIONS AND WARNER-BRATZLER SHEAR VALUES OF LONGISSIMUS MUSCLE OF CONTROL BULLS AND TRENBOLONE ACETATE PLUS ZERANOL IMPLANTED BULLS AND STEERS.. . . .	65

LIST OF FIGURES

<u>Figure</u>		<u>Page</u>
1	Serum testosterone concentrations of control bulls and implanted bulls. . . . .	56
2	Serum estradiol-17 $\beta$ concentrations of control bulls and implanted bulls and steers. . . . .	58

## Chapter I

### General Introduction

Results of a National Consumer Retail Beef Study (Cross et al., 1986) suggest that many consumers prefer leaner beef cuts with less trimmable fat. Beef is perceived by many consumers to contain excess fat and cholesterol and to be limited or avoided in the diet. Therefore, the beef industry currently is undergoing a facelift in an attempt to improve the image of beef and to increase consumer demand for beef. Some packers and most retailers are cooperatively trimming excess subcutaneous fat from of meat cuts. An effort currently is underway to investigate the feasibility of hot-fat trimming. However, the industry must realize the great inefficiency of producing excess animal fat to later be trimmed off and rendered. Production alternatives which address the heightened demand for lean beef currently are being examined. Among these is the utilization of intact males for their performance and cutability advantages over that of steers.

Extensive reviews by Field (1971) and Seideman et al. (1982) clearly demonstrate that young bulls have higher average daily gains (ADG), consume less feed per unit of gain (FE) and produce leaner carcasses than steers at similar ages or weights. However, utilization of young bulls under present production, grading, and processing systems has met with resistance from feeders, packers, processors, retailers and, indirectly, consumers (Dikeman, 1984). This resistance primarily deals with bulls' aggressive behavior during finishing and pre-slaughter phases which ultimately may influence their meat quality (Mies, 1982; Oltjen, 1982; Seideman et al.,



1982). Furthermore, Smith (1982) described difficulties pertaining to hide removal and excessive carcass weights. And, finally, that bulls might be stressed more easily than steers (Field, 1971; Price and Tennessen, 1981) which may result in a lower USDA quality grade, meat that has a higher ultimate pH (hence, a darker, firmer, drier lean) and meat that is generally less tender and juicy than steer beef (Seideman et al., 1982).

Concentrations of endogenous androgen and estrogen in bulls allows for near maximal expression of their genetic growth potential (Schanbacher, 1984), and this may partially explain why implants have not been utilized or perfected for use in young bulls. Baker and Arthaud (1972) reviewed the use of estrogenic compounds in bulls and found no consistent improvement in performance in studies up to that time. However, zeranol (Z) has been shown to be effective in reducing testicle size (Staigmiller et al., 1985; Vanderwert et al., 1985; Silcox et al., 1986; Unruh et al., 1986a), in improving performance (Kirk and Cooper, 1983; Greathouse et al., 1983) and increasing carcass desirability (Greathouse et al., 1983; Unruh et al., 1986b) in young bulls. Furthermore, trenbolone acetate (TBA), a synthetic testosterone analogue that is approximately 10 to 50 times more active than testosterone (Neumann, 1976), can promote a positive, nitrogen balance when combined with estrogenic compounds in bulls, steers and heifers, which results in higher ADG and FE (Grandadam et al., 1975; Galbraith, 1982; Fabry et al., 1983). However, Silcox et al. (1986) concluded that there appeared to be no beneficial effects from implanting bulls with Z and TBA.

There is little known about performance, masculinity, carcass, meat sensory, and endocrine traits of young bulls implanted near birth with Z and TBA until approximately 5 mo of age and sequentially reimplanting with Z

alone until slaughter. Therefore, the objectives of this study were to determine the effects of this implant scheme for young bulls compared with non-implanted, control bulls (CB) and with steers implanted with Z plus TBA near birth and at planned intervals until slaughter on growth, performance, carcass, meat sensory characteristics and concentrations of testosterone (T) and estradiol  $17_{\beta}(E_2)$ .

## Chapter II

### Review of Literature

#### Bulls versus Steers

Live Animal Performance Studies comparing young bulls with steers have been conducted by numerous researchers. Good agreement exists in the literature which documents increased live weight gain and feed conversion benefits that bulls possess over steers (Champagne et al., 1969; Hedrick et al., 1969; Arthaud et al., 1977; Brethour, 1982; Hodge, 1982; Gregory and Ford, 1983; Dyer et al., 1986). Field (1971) and Seideman et al. (1982) reviewed research which compared young bulls with steers and concluded that bulls had a 17% advantage in average daily gain (ADG) and were 13% more feed efficient (FE) when fed to a weight endpoint. Bidart et al. (1970) stated that bulls produced 20% more protein per day per unit of digestible energy consumed than steers. Gortsema et al. (1974) and Galbraith et al. (1978) have suggested that testicular androgens may be partially responsible for the increased performance by acting in concert with nitrogen metabolism.

At all ages (12, 15, 18 and 24 mo of age), bulls had higher ADG, superior FE, and produced leaner carcasses than steers (Arthaud et al., 1977). Furthermore, Champagne et al. (1969) evaluated time of castration and found that bulls had superior ADG and FE than their counterparts castrated at 2, 7, and 9 mo of age. In addition, Hedrick et al. (1969) reported that bulls had heavier slaughter weights and(or) higher ADG and

FE than steers or heifers when slaughtered at a similar number of days on feed or constant weight. However, Crouse et al. (1985) reported that the 15% improvement in FE and 14% improvement in metabolizable energy per unit of gain for Angus and Simmental bulls would be nullified if the data were adjusted to live animal weights associated with a constant rib fat percentage (33.5%). Those authors stated that differences in energy intake between bulls and castrates accounted for the decreased efficiency of bulls compared to steers when fed to a constant rib-fat percentage. Ntunde et al. (1977) reported that Holstein-Friesian bulls must be fed a minimum of 20 d longer than steers to achieve a similar fat thickness. Therefore, the documented performance advantages for feeding young bulls over steers would more likely lend this practice to an accelerated, lean-beef production system rather than feeding to achieve the same compositional endpoint as steers, such as the Choice quality grade.

Pre-Slaughter Management Bulls' inherent aggressive behavior, partially accounted for by the androgens produced by the testes (Craig, 1981), may lead to stress more easily than in steers (Field, 1971). This agonistic behavior can detrimentally influence meat quality by increasing the potential for a condition known as "dark cutters", which is characterized by dark, firm, and dry (DFD) lean which has an ultimate pH greater than 6.0 and its color ranges from dark-red to purplish-black.

Management practices to reduce aggressive behavior such as not mixing strange bulls together during backgrounding, feeding and transport along with slaughtering them quickly upon arrival at the packing plant can help alleviate problems associated with dark lean (Dikeman et al., 1985).

Price and Tennessen (1981), Gregory and Ford (1983) and Brown et al. (1986) reported that bulls subjected to minimal stress prior to slaughter had meat with an acceptable color. However, Price and Tennessen (1981) investigated the effects of mixing strange bulls before slaughter on the incidence of DFD and reported 73% of the mixed bulls were dark cutters compared to 2 % for unmixed bulls. In a similar type of study, Jones et al. (1986) evaluated the effects of mixing and shipping on the incidence of DFD in bulls and steers. They found that animals subjected to moderate stress (mixed, held off feed and water overnite, transported 160 Km, and slaughtered within 24 hr), regardless of sex, had darker lean with a higher pH than unmixed counterparts which were transported 4 km and slaughtered within 4 hr after leaving the feedlot.

Since young bulls may have a higher predisposition to be dark cutters than steers (11 to 15% compared to 1 to 5%; Oltjen, 1982), reducing pre-slaughter stress of young bulls is essential for helping to insure normal pH, cherry-red appearing muscle.

Slaughter and Fabrication Slaughtering young bulls can pose certain difficulties not typically encountered in steers or heifers. Smith (1982) listed difficulties which include: a) bullocks are less manageable in holding pens, b) bullocks are more difficult to stun, c) bullock heads must often be manually skinned, d) bullock hides are difficult to pull which might tear the carcass or break hock tendons, e) increased bone density of bullocks may cause problems with saws and hock cutters, f) standard rail switches cannot support the weights of heavy bullock carcasses, g) the wide, thick, heavy bullock hides can cause breakdowns of hide-fleshing equipment and may

need to be passed through the fleshing machine two or three times, and h) bullock hides may require additional curing time and their hides are usually worth less money. These difficulties associated with hide removal are quite a disadvantage for bullocks (Seideman et al., 1982). Jones et al. (1986) reported that the hide, as a percentage of live weight, was 8.6% for bulls compared with 7.6% for steers, and that bull hides were 10 kg heavier than those from steers.

Two factors that can influence the usefulness of bulls in a commercial, boxed-beef operation are carcass weight and fat cover. The meat industry places constraints on carcasses that are too large (over 384 kg) and those with an inadequate fat cover (less than .5 cm) (Cross, 1982). Bowling (1982) stated that adequately finished carcasses are acceptable up to 410 kg while inadequately finished carcasses present problems at 385 kg. Therefore, small-framed bulls may be fed to heavier weights than steers without excess fattening; however, large-framed bulls may not have a sufficient fat cover at acceptable carcass weights.

Carcass masculinity characteristics may determine whether the carcass will be merchandised in boxed-beef form or carcass form at a price considerably less than USDA Good-grade steers. Binger (1982) listed undesirable characteristics of bulls which include size of the pizzle eye, excess fullness and thickness of neck, and the appearance of the jump muscle, round and ribeye. These factors may be interrelated to meat color and texture.

Therefore, these implications could impede the production of young bulls for beef, partially due to packer resistance. However, considering the variability which exists in market cattle at present (carcass weight, fat

thickness, propensity to marble), slaughtering bulls may be just as acceptable to some packers as steers.

Carcass Cutability Allen (1982) summarized a composite of research trials and found bulls to be superior to steers in carcass cutability. The advantages are mainly noted in trimness and larger ribeye area per unit of carcass weight when compared with steers. He also stated that bulls were 14 kg heavier, 5.8 mm trimmer over the longissimus muscle, yielded 5.1% more separable lean and had .2% more carcass bone than steers at the same age. Field (1971) reviewed several studies and reported very similar findings. He reported similar dressing percentages for bulls and steers (59.7% vs. 59.6%, respectively) whereas bulls had a 2.6% advantage in boneless chuck, rib, loin and round using the equation of Murphey et al. (1960) over steers. However, Champagne et al. (1969), Jacobs et al. (1977a), Greathouse et al. (1985), Hopkinson et al (1985) and Jones et al. (1986) found differences of over 9.0% in edible portion yield using actual cut-out data. In addition, Jacobs et al. (1977a) indicated that bull carcasses yield 5.5% more boxed beef and have 17% less cutting losses than steers. Since small variation exists in percentage of carcass bone between bulls and steers, bull carcasses have a higher muscle to bone ratio than steer carcasses (Berg and Butterfield, 1968; Greathouse et al., 1985).

The carcass weight distribution in the major wholesale cuts is of economic importance. Bulls exhibit a greater propensity to develop muscles of the forequarter which tend to decrease the value distribution of the "middle-meat" portions (Mukhoty and Berg, 1973; Kay and Houseman, 1975).

Vanderwert et al. (1985) stated that bull carcasses had a higher percentage of chuck and lower percentage of loin than steers.

Therefore, bulls have definite advantages over steers by yielding more separable lean, having less subcutaneous adipose tissue, and having a higher muscle to bone ratio.

Carcass Quality Reduced marketability of bullock beef, which has been implicated as receiving lower USDA quality grades than steer beef (Bailey et al., 1966; Field, 1971; Binger, 1982; Seideman et al., 1982), is a serious consideration, particularly when bullock beef usually is also darker colored and coarser-textured. Cross and Allen (1982) and Smith and Merkel (1982) reviewed 16 and 21 studies, respectively, and reported that bullocks had an average marbling score of "slight-typical" and a mean USDA quality grade of average Good. In comparison, their steer mates had marbling scores in the upper portion of "small" and lower portion of "modest" and had a mean USDA quality grade of low Choice.

Another factor influencing the USDA quality grade is carcass maturity. Differences in lean and skeletal maturity have shown that bulls are more advanced at the same chronological age than steers (Glimp et al., 1971; Reagan et al., 1971; Crouse et al., 1985). Arthaud et al. (1977) reported that carcass maturity was similar for bulls and steers when slaughtered at 12 mo of age. However, from 15 to 24 mo, bull carcasses were more mature physiologically than steers, but not enough to significantly affect USDA quality grade.

The limitations of bulls in their propensity to marble comparable to steers remains a serious drawback towards enticing feeders and packers to



move towards utilization of bulls for meat production. Smaller genotypes would appear to work best for this type of market, whereas genotypes with larger size and more muscling do not appear to work well for this marketing system (Dikeman et al., 1985).

Meat Palatability and Retail Acceptance Price, appearance and palatability ultimately influence consumer buying decisions. Cross and Allen (1982) stated that part of the price disparity between bull and steer carcasses stems from the belief that bullock beef has a lower consumer acceptance at the retail level because of differences in color, texture and marbling than steer beef. Palatability characteristics of steaks from young bull carcasses have been found to be generally acceptable, but rated slightly less desirable than those from steers (Field, 1971; Seideman et al., 1982).

Modern consumers prefer lean beef (Breidenstein, 1982). Studies conducted by Jacobs et al. (1977b) and Berry et al. (1978) concluded that leanness was the most important criteria for consumers in making purchase selections. In addition, Baron et al. (1978) found leanness and texture to be equally important to consumers. Jacobs et al. (1977b) reported that nearly 50% of those interviewed regarded marbling as the least important factor affecting meat selection when also considering color and leanness. Therefore, bullock beef may actually have a tremendous advantage at the retail level, because of its high lean-to-fat ratio.

In addition to the heightened demand for leaner beef, consumers desire beef to be tender, flavorful, and juicy. Smith and Merkel (1982) have suggested that beef from young bulls is usually less tender than beef from steers. Of these attributes, Breidenstein and Carpenter (1983) considered

tenderness to be the most important palatability attribute. Cross (1982) investigated the palatability of beef from bulls and steers from 6 to 24 mo of age whereas Dikeman et al. (1986) summarized the effects that high and low energy diets had on longissimus characteristics of bulls and steers slaughtered from 9 to 24 mo of age. Cross (1982) concluded that bullock beef was rated lower in palatability and was more variable than beef from castrates, while Dikeman et al. (1986) reported that Warner-Bratzler Shear (WBS) values were similar for bulls and steers fed a low energy diet, whereas steers fed a high energy diet had lower WBS values than bulls fed either diet.

Possible explanations for the observed reduction in tenderness of meat from bulls relative to steers may be partially due to the increased incidences of cold-shortening in bulls (Riley et al., 1983). Bull carcasses may lack sufficient fat thickness to properly insulate the carcass during the stringent chilling conditions which exist in modern, commercial packing plants. Additionally, the amount and solubility of collagen can influence tenderness (Boccard et al., 1979; Cross et al., 1984; Judge et al., 1984; Klastrup et al., 1984; Burson et al., 1986). Boccard et al. (1979) suggested that variations between bulls and steers may be linked to sexual development and the associated increase in circulatory androgens. Goll et al. (1964) stated that the cross-linking of collagen occurs with increased age. Therefore, Judge et al. (1984) speculated that the lower proportions of heat-soluble collagen of bullock beef may be partially explained by an accelerated rate of collagen maturation compared with steers. Finally, Hall (1976) reported that synthesis and tensile strength of collagen may be increased by anabolic steroids such as testosterone.

Although differences exist in the appearance and palatability of beef from young bulls and steers, most studies agree that bullock beef is very acceptable. Furthermore, Cross (1982) added that future consumers will likely have a "lower threshold of acceptability than those from the past." Once some palatability threshold is met, leanness might be relatively more important than palatability. Therefore, the utilization of young bulls for meat production might increase in the future.

### Endocrine Relationships to Growth

One of the objectives in animal science is to increase our understanding of the basic biological mechanisms that regulate skeletal-muscle and adipose tissue growth. It is necessary to study principles which enhance our understanding of how the cell membrane allows cells to communicate, recognize and respond to their environment, and to regulate nutrient entry and exit. By increasing our understanding of the relationships between the endocrine system and cellular mechanisms of growth, it may be feasible to enhance either the rate, extent or efficiency of animal growth (Etherton, 1982).

Modes of Receptivity Since skeletal muscle development is more pronounced in males than in females, it is assumed that androgenic hormones are responsible for this myotropic or anabolic action (Snochowski et al., 1981). Androgenic agents have been shown to increase protein accretion in muscle by exerting an anabolic effect on protein metabolism (Vernon and Buttery, 1978). Because the protein proportion in muscle fibers can be altered by

administering anabolic steroid hormones, cellular mechanisms that regulate the rate of protein deposition must be present (Snochowski et al., 1981).

The first step in the regulation of cellular metabolism by steroid hormones appears to be binding to a specific receptor on the plasma membrane (Roth, 1973; Cuatrecasas, 1974). They penetrate into the target cell and bind with stereospecificity to high affinity receptors. The receptors in the cytosol are different from acceptor sites in the nucleus, and the translocation of the hormone from one subcellular site to the other requires an activation step. There is an induction of mRNA and protein synthesis, and physiological expression of induced protein.

Butt (1976) reported that T and E<sub>2</sub>, the gonadal steroids, bind to a β-globulin, plasma protein while a specific α-globulin protein shows a higher affinity for cortisol (C). Vernon et al. (1980) stated that insulin (I) receptor sites were present on isolated sheep adipocytes and Olefsky et al. (1976) and Marchand-Brustel et al. (1978) found insulin receptors in skeletal muscle. Furthermore, Butt (1976) indicated that binding is a reversible process and may involve hydrogen bonding, hydrophobic bonding and dipole interactions. The binding of steroids to plasma proteins plays an important physiological role, since it is only the unbound fraction which is exchangeable with the intracellular and extravascular compartments, and has biological activity.

Subtle differences exist in the reporting of hormone binding characteristics of androgens, estrogens, glucocorticoids (GLC) and I which have all been isolated from skeletal tissue (Knudsen and Max, 1980; Vernon et al., 1980; Snochowski et al., 1981). Dube et al. (1976) and Dionne et al. (1979) found that the estrogen receptor is different from the androgen

receptor. However, Michel and Baulieu (1980) reported that  $E_2$  binds to the androgen receptor but with 5 to 10 times lower affinity. However, these authors concluded that at high enough concentrations,  $E_2$  could act as an androgen. Additionally, Mayer and Rosen (1977) and Buttery et al. (1978) stated that the anabolic activity of T and  $E_2$  might result from their displacement with cortisol from the GLC receptors, respectively. This would impede the GLC protein-catabolic activity. However, Michel and Baulieu, (1983) reported that the GLC receptors were different from androgen receptors and will not bind androgens.

These differences may become apparent when considering that  $E_2$  binds with the androgen receptor; however, synthetic estrogenic compounds such as DES do not.  $E_2$ , a naturally occurring hormone, acts by having both a direct and an indirect effect on protein synthesis, whereas synthetic estrogenic agents appear to act indirectly only (Michel and Baulieu, 1983) by increasing growth hormone (GH) secretion which can promote growth, feed conversion and carcass protein accretion (Preston, 1975).

Growth Hormone and Insulin Many hormones act synergistically and antagonistically in the regulation of growth and tissue development (Gray et al., 1986). Among these, GH and I appear to act synergistically and both are essential for growth (Etherton and Kensinger, 1984; Muir, 1985). The endogenous release of GH from the pituitary is under control of GH-release factor (GRF) and GH-release inhibiting factor (somatostatin) from the hypothalamus (Guilleman et al., 1982). GH is thought to mediate its action by stimulating the secretion of somatomedins (SM). These SM, in turn, have been shown to increase myoblast proliferation and differentiation of

myotubes in myoblast cultures, while GH addition to these cultures has generally not promoted growth or differentiation (Florini et al., 1977; Ewton and Florini, 1980; Florini, 1985). Furthermore, Allen et al. (1983) evaluated the effects that GH and T had on actin formation in myoblast cultures from adult muscle-satellite cells. They reported that neither GH or T were the active agents in the serum that stimulated actin formation and that SM was the most likely stimulatory agent.

It appears that I, at concentrations of .2 to  $2.0 \times 10^{-6}$  M, can mimic SM actions and enhance myoblast proliferation (Florini, 1985). However, at physiological concentrations of I ( $10^{-10}$  to  $10^{-9}$  M), Airhart et al. (1982) reported that I stimulates protein synthesis in chick muscle in vitro. Modes of I action have been proposed which suggest that the growth-promoting activity of I may be attributed to the cross-reactivity of I with somatomedin receptors and does not involve the insulin receptor (Baxter et al., 1980; King et al., 1980; Florini and Ewton, 1981). Insulin also has been found to decrease the rate of proteolysis when added to isolated diaphragm muscle (Goldberg et al., 1980) and chick myoblast cell cultures (Libby and Goldberg, 1981). Nevertheless, Roeder and Hossner (1986) reported that SM exhibited a greater effect on protein synthesis and degradation than I. Furthermore, those authors concluded that SM play a primary role in protein turnover in muscle.

A positive relationship exists between GH concentration and carcass muscle, while a negative relationship exists between GH and adipose tissue of ruminants (Trenkle and Topel, 1978; Wagner and Veenhuizen, 1978). Furthermore, Wagner and Veenhuizen (1978) administered exogenous GH to lambs and found an improvement in ADG and FE while Moseley et al. (1982)

infused steers and Moseley et al. (1986) infused bulls with GRF and found that this stimulated nitrogen accretion. A recent study investigated the effects of actively immunizing animals against somatostatin. Results from a trial conducted by Lawrence et al. (1986) showed a 17.6% improvement in ADG and a benefit of 12.7% for FE in immunized, crossbred steers compared to their controls. Research in this area will likely intensify to further investigate the role that SM and I, along with other hormones, have in regulating growth and development.

Thyroid Hormones Minimal levels of  $T_3$  (triiodothyronine) and  $T_4$  (thyroxine) are essential for normal growth and development. These iodinated hormones secreted by the thyroid are generally thought to have anabolic actions (Florini, 1985) in addition to stimulating oxidative metabolism of cells (Mosier, 1981).

Bray (1964) stated that at physiological levels, the thyroid hormones have an important influence in regulating growth and appetite. However, high concentrations of  $T_3$  and  $T_4$  cause induction of some degradative enzymes and enhance protein catabolism (Burman et al., 1979; Goldberg et al., 1980). Additionally, Muir and Wien (1983) concluded that decreased gain, feed conversion and carcass protein likely resulted from high levels of circulating  $T_3$  and  $T_4$  in lambs infused with exogenous thyrotrophin-releasing hormone.

It appears that thyroid hormones may have a relationship to GH somewhat similar to the manner in which I can influence GH receptor and SM levels (Furlanetto et al., 1979; Davis et al., 1984). These findings suggest that  $T_3$  and  $T_4$  may ultimately be involved with the intricate

balance of growth-promoting hormones which must be precisely regulated for normal tissue growth (Beitz, 1985).

Glucocorticoids Cortisol is a GLC which influences growth. Daughaday et al. (1975) indicated that C acts on skeletal muscle causing an increase in protein catabolism and decreased protein synthesis which can increase amino acid availability to the liver for gluconeogenesis. Nevertheless, Florini (1985) stated that GLC have a role in the maintenance of normal muscle. In that review, Florini cited some studies which showed that GLC stimulate proliferation and differentiation in rat myoblasts and others that report an increase in protein degradation.

In cattle studies which compared the effects of castration and sex on plasma C concentration, bulls had lower levels than heifers (Henricks et al., 1984) or steers (Tennessee et al., 1984). Furthermore, Purchas et al. (1971, 1980) and Trenkle and Topel (1978) demonstrated that plasma C was negatively correlated with growth rate in heifers and steers. A more recent study by Henricks et al. (1984) reported that no relationship existed between plasma C and weight gains in crossbred beef bulls.

The action of GLC may be influenced by I and T. Bassett (1968) and Lewis and Goldspink (1982) demonstrated that high plasma C concentrations are accompanied by elevated I levels and that the presence of I was necessary for C to have its antilipolytic effects. This may partially explain the positive relationship between increasing plasma C concentrations and carcass lipid found by Trenkle and Topel (1978).

Johnson et al. (1982) reported that an antagonistic relationship exists between plasma C and T concentrations in bulls administered



adrenocorticotropin, and Henricks et al. (1984) noted a similar trend. Furthermore, trenbolone acetate (TBA), a synthetic androgen with 10 to 50 times the potency of T is quite effective in reducing plasma C levels when administered to rats and sheep (Thomas and Rodway, 1982a). Finally, Thomas and Rodway (1982b) and Bukoski et al. (1986) conducted basic studies which investigated the effects of TBA or T on C production in rat and porcine adrenal gland preparations, respectively. Both studies concluded that either TBA or T depressed C secretion. This suggests that T and TBA may exert some of their anabolic influence through the suppression of adrenal activity (Thomas and Rodway, 1982a).

Further research in this area may more clearly elucidate the role of C in growth and development in meat animals, particularly the hormonal relationships with I and T, and competition for receptor sites.

Androgens Florini (1985) reviewed several studies that investigated the effects of androgens on skeletal muscle growth. He concluded that T, produced from the testes and adrenal cortex (Rudd, 1976), acts as an anabolic hormone through a direct interaction with a cytoplasmic receptor. This may lead to the migration of the hormone-receptor complex, stimulation of RNA synthesis and protein accumulation.

These implications become apparent when considering the fact that androgens have definite anabolic activity. Positive correlations between plasma T and growth rate have been observed in many studies (Secchiari et al., 1976; Sitartz et al., 1976; Sundby and Velle, 1983; Gray et al., 1986). Since circulating hormone concentrations may fluctuate in a somewhat random manner (Henricks et al., 1984), a technique used by Sundby and

Velle (1983) to evaluate bulls' ability to respond to a hormonal challenge (human chorionic gonadotropin) was employed. Those workers measured plasma T in 5- to 11-mo old bulls and discovered that by dividing bulls into high and low T groups, that growth was higher for bulls in the high T group.

Testosterone levels in bulls increased from birth to puberty (Rawlings et al., 1972; Lunstra et al., 1978; Renaville et al., 1983). In post-pubertal bulls, McCarthy et al. (1979) and Schanbacher (1984) reported that T elevation is preceded by a peak release of lutenizing hormone (LH). Furthermore, Fabry et al. (1983) demonstrated that one or two T spikes followed each LH surge with a delay of 40 to 80 min, and that T average basal concentrations of .7 ng/ml rose to a peak of 9.0 ng/ml following LH surge.

Additionally, androgens may have indirect effects on growth by enhancing the production of GH (Galbraith et al., 1978), by affecting the binding inhibition of C to their receptors (Mayer and Rosen, 1975) and peripheral aromatization to estrogens (Knudsen and Max, 1980). These observations suggest possible alternative modes of action in addition to direct cytosol interaction.

Estrogens Estradiol is produced from the aromatization of androgens (Knudsen and Max, 1980) and is secreted from the testis in bulls (Morris, 1976). The effects of endogenous E<sub>2</sub> on muscle growth seem more obscure than those of androgens, and less direct and more indirect growth influences have been characterized for E<sub>2</sub> than for androgens (Michel and Baulieu, 1983).

Neither Powers and Florini (1975), nor Knudsen and Max (1980) were able to demonstrate an increase in protein content in cultured muscle cells when  $E_2$  was administered. However, Preston (1975) suggested that exogenous estrogenic treatments are believed to indirectly influence growth by enhancing the release of GRF, which causes a release of GH. Trenkle (1983) reported that GH concentration in blood plasma is increased following treatment with  $E_2$ . Also, increased nitrogen retention, greater protein deposition, decreased excretion of urea, and often increased I levels are noted with administration of exogenous  $E_2$  (Trenkle, 1983). However, Baker and Arthaud (1972) found that bulls did not respond with as much weight gain as steers when given similar levels of exogenous  $E_2$ . Therefore, inconsistent improvements for young bulls treated with  $E_2$  agents were observed.

The previous sections have revealed insight on relationships between muscle growth and specific hormones. The sections are by no means all inclusive of the controlling mechanisms thought to regulate the endocrine system. At present, the modes of action of anabolic agents are receiving considerable attention. Presumably, the results from these and future studies will enhance our understanding of the hormonal relationships involved in growth and composition of young bulls and steers.

## Exogenous Anabolic Agents in Cattle Production

Response by Different Sexes A major objective of animal production is to improve the efficiency of lean-tissue growth. From the preceding section, it was observed that some of the hormones reviewed have distinct effects on growth. Therefore, certain estrogenic-like or androgenic-like agents administered in the form of a subcutaneous implant at the base of the ear, have generally demonstrated positive effects on protein accretion and growth for steers and heifers in several studies (Unruh, 1986c). Heitzman (1978) investigated growth of steers and heifers implanted either singly or in combination, with estrogenic-like and androgenic-like agents. It was concluded by Heitzman (1978) that heifers responded maximally to androgens, while a combination of androgen and estrogen was a more potent growth stimulator for steers. However, growth improvements reported for implanted steers and heifers, particularly with estrogenic-like agents, have not been consistently demonstrated for bulls (Baker and Arthaud, 1972; Unruh, 1986c). This was attributed partly to the endogenous concentrations of androgens and estrogens in bulls which may already allow for near-maximal growth (Schanbacher, 1984; Unruh, 1986c). Galbraith (1982) speculated that estrogenic compounds stimulate growth only in young bulls or castrates that have low concentrations of endogenous sex hormones. Therefore, administering estrogenic-like compounds at dose levels commonly used would have little supplementary effect in stimulating growth rate above that which is inherent by the bulls' endogenous secretion of androgens and estrogens.

Modes of Action for TBA and Z Trenbolone Acetate (3-oxo-17 $\beta$ -hydroxy-4,9,11-estratriene acetate) is deacetylated in the blood to its active component trenbolone, which is very similar in structure to T (Pottier et al., 1975; Vernon and Buttery, 1978). The primary action reported for TBA has been associated with protein metabolism. Greater nitrogen retention has been demonstrated by Chan et al. (1975) and Vernon and Buttery (1978), who also reported a reduction in both protein synthesis and a proportionately greater decrease in protein degradation. Therefore, the net effect was protein accretion.

Zeranol, an estrogenic, resorcylic-acid lactone is not classified as an estrogen. Nevertheless, Z has been observed to have estrogenic-like activity (Beverly, 1984). This estrogenic-like activity might be accounted for by an increase in production of androgens from the adrenal cortex, increased thyroid hormone activity, elevated GH secretion and a direct effect on the target tissue (Trenkle and Burroughs, 1978).

Trenkle (1983) reported that there does not seem to be a single mechanism of action that can explain the increase in growth and protein deposition from various anabolic agents. Furthermore, Trenkle (1983) concluded that the effects of estrogens and androgens on accumulation of muscle proteins appear to be independent, but additive, as current evidence indicates that the androgens primarily act directly on muscle to increase protein while the estrogens may act more on the hypothalamus and anterior pituitary to stimulate GH secretion.

Commercial Implants for Cattle There are numerous implants available for cattle producers (table 1).

This list of commercial implants includes compounds which have a mandatory withdrawal period. Those that include Z and TBA are not to be administered to cattle the final 65 days prior to slaughter. If used properly, these implants can enhance growth efficiency in cattle.

The following section of this review will primarily focus on effects of implanting young bulls and steers with Z plus TBA. Brown (1983), Unruh (1984) and Gray (1985) provide excellent reviews on the effects of zeranol on bulls and steers. Likewise, Bouffault and Willemart (1983) have extensively reviewed the response of cattle to TBA implants.

Effects on Growth. Growth benefits have been reported in male veal calves implanted with 140 mg TBA plus 20 mg  $E_2$  from 40 to 170 kg (Grandadam et al., 1975; Vanderwal et al., 1975; Bouffault and Willemart, 1983). VanderWal et al. (1975) demonstrated that the maximum growth response was observed for the implant combination of TBA +  $E_2$  when compared with  $E_2$  alone, or combined with T or progesterone. Bouffault and Willemart (1983) reviewed 11 studies involving veal calves implanted with Z and TBA and reported a 10 to 17% improvement in ADG.

Enhanced growth in older bulls may be expected to occur only following implantation with anabolic compounds more potent in terms of quantity present and growth-promoting activity than those produced endogenously (Galbraith, 1982). Grandadam et al. (1975) reported that implanting Salers bulls at 15 mo of age with 20 mg  $E_2$  plus 200 mg TBA resulted in a 28% improvement in ADG over the final 50 d prior to slaughter. In addition, Galbraith (1982)

TABLE 1. Growth promoting implants

Implant	Active Compound	Dose mg	Use	Activity days	Withdrawal time
Ralgro <sup>®a</sup>	Zeranol	36	Heifers and steers-birth to slaughter	90	65
Compudose <sup>®a</sup>	Estradiol-17 $\beta$	24	Steers-birth to slaughter	200	0
Synovex-S <sup>®a</sup> (Steer-oid <sup>®</sup> ) <sup>a</sup>	Estradiol benzoate Progesterone	20 200	Steers over 400 pounds	90	0
Synovex-H <sup>®a</sup> (Heifer-oid <sup>®</sup> ) <sup>a</sup>	Estradiol benzoate Progesterone	20 200	Heifers over 400 pounds	90	0
Synovex-C <sup>®a</sup>	Estradiol benzoate Testosterone	10 100	Nursing steer and heifer calves	-	-
Finaplix <sup>®b</sup>	Trenbolone acetate	140	Feedlot steers	90	60
Forplix <sup>®b</sup>	Trenbolone acetate Zeranol	140 36	Used for all classes of cattle	90	65
Revalor <sup>®b</sup>	Trenbolone acetate Estradiol-17 $\beta$	140 20	Used for all classes of cattle	90	60

<sup>a</sup>Adapted from Mader (1983).

<sup>b</sup>Adapted from Schanbacher (1984).

demonstrated that implanting British-Friesian bulls with 300 mg TBA plus 45 mg hexesterol enhanced ADG 22% over the final 70 d period and increased FE by 17%. Fabry et al. (1983) reported responses of similar magnitude in double-muscled Belgian, White-Blue bulls implanted at 8 mo of age with 36 mg Z plus 140 mg TBA. However, Silcox et al. (1986) and Fisher et al. (1986b) were unable to demonstrate lasting advantages in ADG or FE in bulls either previously implanted with 36 mg Z and then reimplanted with 200 mg TBA alone, or with 200 mg TBA plus 36 mg Z or with 36 mg Z plus 140 mg TBA at 44 and 300 d, respectively.

The effects of implanting steers with TBA plus estrogenic agents on ADG and FE have been more intensively studied than for bulls and the results show a clear-cut improvement for steers implanted with TBA and Z. Heitzman (1978) demonstrated substantial increases in both ADG and FE for steers implanted with TBA plus  $E_2$ . Furthermore, several researchers have shown marked improvements for these traits in subsequent studies utilizing TBA plus estrogenic agents (Roche et al., 1978a and b; Galbraith and Geraghty, 1982; Galbraith et al., 1983; Lobley et al., 1985; Steen, 1985; Fisher et al., 1986a and b). The magnitude of improvement for ADG ranged from only slightly (Steen, 1985) to over 40% in trials conducted by Galbraith and Geraghty (1982). Heitzman (1978) also reported a 27% increase in FE.

Subtle differences exist in the literature for the various estrogenic agents and the dose levels used to compliment the TBA implant. Furthermore, variation in age, breed, time of castration and weight at time of implantation may have influenced the effectiveness of these compounds. Nevertheless, researchers are in good agreement that TBA, in association with estrogenic



agents, does enhance growth, and reimplanting stimulates additional growth, but to a lesser extent than the first implant (Lobley et al., 1985).

Effects on Certain Hormones and Blood Metabolites. Implanting bulls with TBA + hexestrol has not been shown to alter mean concentrations of plasma glucose, free fatty acids, C, GH and I (Galbraith, 1982). Similar results for GH and I were reported for steers by Heitzman et al. (1977) and Trenkle (1983). However, Thomas and Rodway (1982a) found that implanting ewe lambs resulted in lower C levels. Galbraith (1982) also stated that T was reduced substantially following implantation. The work of both Fabry et al. (1983) and Silcox et al. (1986) support this observation for bulls implanted with TBA plus Z. Neumann (1976) attributed this reduction in T to the strong androgenic and estrogenic activity of the implant which may interfere with normal hypothalamic feedback mechanisms. Furthermore, Galbraith et al. (1978) have shown that plasma urea was reduced in implanted bulls and steers, and that these reductions have been associated with increases in weight gain and protein content. Additionally, Lobley et al. (1985) demonstrated a decrease in urinary N<sup>+</sup>-methylhistidine elimination, which might suggest a reduction in protein degradation. Therefore, the net affect of TBA and Z may be to increase protein accretion.

These observations are consistent with our current understanding of some of the complex interactions which occur when implanting bulls and steers with a combination of androgenic and estrogenic agents. Nevertheless, studies which address the metabolic pathways which more clearly elucidate the rates at which these interactive reactions take place will certainly enhance our knowledge about the mechanisms of action of these compounds.

Effects on Masculinity Traits. Evaluating traits relating to masculinity has been conducted to a limited extent. Silcox et al. (1986) found no differences in testis weight in bulls implanted with TBA plus Z following a 112-d finishing period. However, scrotal circumferences were larger for non-implanted, control bulls than for implanted bulls.

Fisher et al. (1986a) observed no effect of anabolic agents on the characteristic muscle weight distribution in bulls, particularly the neck muscles which are relatively hypertrophic because of androgen stimulation. The implanted steers in that trial did not develop a muscle distribution similar to bulls. However, in a separate study, implanted steers had heavier hides and penis' were more developed than non-implanted steers (Fisher et al., 1986b).

Further studies are necessary to characterize the effects of these implants on both live animal and carcass masculinity traits in relationship to their effects on endogenous T production (Silcox et al., 1986).

Effects on Carcass and Meat Traits. Implanting with TBA plus estrogenic agents has been shown to increase carcass weight in bulls (Grandadam et al., 1975; Galbraith, 1982; Fisher et al., 1986a) and steers (Steen, 1985; Fisher et al., 1986a and b). Furthermore, these implants combined have had little or no influence on dressing percentages in bulls (Grandadam et al., 1975; Galbraith, 1982; Bouffault and Willemart, 1983; Fisher et al., 1986a and b) or steers (Bouffault and Willemart, 1983; Fisher et al., 1986b).

Kidney, pelvic and heart fat was estimated to be .8 kg more for bulls implanted with TBA and Z (Silcox et al., 1986) than controls, whereas Steen (1985) reported that implanted steers (IS) had slightly less kidney knob than

non-implanted steers. Also, Silcox et al. (1986) reported similar rib eye areas for control and IB. Likewise, Steen (1985) observed no differences for rib eye area per kg carcass weight for control and IS. Furthermore, Silcox et al. (1986) found no differences between IB and CB for subcutaneous fat thicknesses, marbling, yield grades or quality grades. However, Fisher et al. (1986b) stated that fat deposition, both subcutaneous and intermuscular, was increased in IB while these adipose tissue depot sites were decreased for IS compared to non-implanted steers. Fisher et al. (1986b) hypothesized that if  $E_2$  is the more active ingredient in combined implants, it would appear that it has a mild effect on protein anabolism in steers and a lipogenic effect in bulls.

Longissimus-muscle chemical composition showed that IS had marginally higher concentrations of water and protein and less fat than controls; while IB tended to have higher concentrations of lipid and lower concentrations of water and protein (Fisher et al., 1986b).

Studies which evaluate meat sensory properties of bulls and steers implanted with TBA plus estrogenic agents routinely have not been conducted. Bouffault and Willemart (1983) mentioned a study by Maghuin-Rogister et al. (1982) which reported no differences in collagen or hydroxyproline levels for implanted versus non-implanted animals, and that shear-force values were similar for IS and IB, and cooking losses were identical for IB and CB.

Therefore, there appears to be a void in the literature on the effects of TBA plus estrogenic agents on carcass and meat sensory traits of implanted bulls and steers. Studies are needed which address these considerations to provide information relating this practice of implanting bulls and steers to the palatability of the meat produced from their carcasses.

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### Chapter III

## PERFORMANCE, CARCASS, MEAT SENSORY AND ENDOCRINE TRAITS OF YOUNG BULLS AND STEERS IMPLANTED WITH TRENBOLONE ACETATE AND ZERANOL.

### ABSTRACT

Twenty-five Polled Hereford calves were assigned randomly to one of three treatments at birth. Eight bull calves were nonimplanted controls (CB). Nine bull calves were implanted with 140 mg trenbolone acetate (TBA) and 36 mg of zeranol (Z) at about 1 mo of age and reimplanted with both implants 10 wk later. When the nine implanted bulls (IB) were about 21 wk of age, TBA implants were removed. The IB then were reimplanted with Z alone every 10 wk until slaughter. The remaining eight bull calves were castrated (IS) at about 3 wk of age and implanted with TBA and Z every 10 wk until slaughter. Five calves had to be removed from the study because of chronic bloat, and (or) blindness or a mistake in castration. Calves were weaned about 7 mo of age and fed a high concentrate diet for up to 210 d. Blood samples were collected biweekly starting at 5 wk of age and analyzed for estradiol  $17\beta$  ( $E_2$ ) and testosterone (T). Weaning weight, daily gain and feed efficiency were similar for all treatments, whereas hip heights at 12 mo and masculinity scores at 13 mo tended ( $P=.09$ ) to be lowest for IS. Scrotal circumferences were lower ( $P<.05$ ) for IB than for CB at both 8 and 13 mo. Slaughter weight was lowest ( $P=.07$ ) for CB. There were no differences in carcass traits due to treatment. CB had heavier ( $P<.05$ ) testicular weights and more ( $P=.07$ )



longissimus muscle detectable connective tissue as determined by a trained sensory panel. Implanting bulls with TBA and Z suppressed ( $P < .05$ ) concentrations of serum T from 7.5 to 11 mo. However, IB had higher ( $P < .05$ ) serum T levels from 12 to 13.6 mo than CB. Serum concentrations of  $E_2$  were similar among treatments. Implanting steers resulted in the optimum combination of performance, carcass and meat sensory traits.

Key words: Bulls, Steers, Anabolic Implants, Performance, Carcass, Endocrine

## Introduction

Cattle production alternatives which address the heightened consumer demand for lean beef are currently being examined. Among these is the utilization of young bulls for meat production because of their performance, carcass leanness, and cutability advantages over that of steers (Field, 1971; Seideman et al., 1982). However, producing young bulls for meat under present feeding, grading and processing systems has met with resistance from various industry sectors (Dikeman, 1984). This resistance primarily stems from bulls' aggressive behavior, lower quality grades and (or) marbling scores and increased incidence of darker, coarser-textured lean than steers (Seideman et al., 1982).

Steroid implants developed to increase growth in steers and heifers have been examined in bulls as well. Baker and Arthaud (1972) concluded that estrogenic agents administered to bulls resulted in no consistent improvement in performance. This might be attributed to the high endogenous concentrations of androgens and estrogens in bulls that allow for near maximal expression of their genetic growth potential. This is the primary reason why implants have not been utilized in young bulls (Schanbacher, 1984). However, when bulls were implanted with Z near birth and subsequently reimplanted until slaughter, masculinity was suppressed (Ralston, 1978; Unruh et al., 1983, 1986a). This characteristic may be important as Riley et al. (1983) and Johnson et al. (1984) found that less masculine carcasses produced steaks that were rated more palatable and required less Warner-Bratzler (WBS) shear force than steaks from carcasses which were more masculine.

Trenbolone acetate, a recently approved growth implant for feedlot steers in the U.S., in combination with estrogenic agents, improved performance of male veal calves at the time when endogenous androgen and estrogen concentrations were low (Grandadam et al., 1975; VanderWal et al., 1975; Bouffault and Willemart, 1983). Enhanced growth in older bulls may be expected to occur only following implantation with anabolic compounds more potent in terms of quantity present and growth-promoting activity than those produced endogenously (Galbraith, 1982). Generally, positive growth responses from implanting with TBA plus an estrogenic agent have been observed in bulls (Grandadam et al., 1975; Galbraith, 1982; Fabry et al., 1983; Fisher et al., 1986a) and steers (Heitzman, 1978; Loblely et al., 1985; Steen, 1985). However, Fisher et al. (1986b) and Silcox et al. (1986) were unable to demonstrate favorable performance improvements for IB. Furthermore, there is an absence of information about the effects of TBA implants combined with estrogenic agents on masculinity, and meat sensory traits of young bulls and steers.

Therefore, our objectives were two-fold: the first objective was to determine the implant effects on performance, masculinity, carcass, meat sensory and endocrine traits of bulls and steers; the second objective was to evaluate the response of bulls to TBA plus Z early in life, followed by subsequent reimplanting with Z to depress masculine development.

## Materials and Methods

Management and Live Animal Measurements. Twenty-five Polled Hereford bulls, born in the spring of 1985, were obtained from the Kansas State University Cow-Calf Unit and were assigned randomly to one of three treatments shortly after birth. Eight calves remained as non-implanted, control bulls (CB). Nine bulls were implanted (IB) with 140 mg trenbolone acetate (TBA) and 36 mg zeranol (Z) at about 1 mo of age and reimplanted with both compounds 10 wk later. When these nine IB calves were about 21 wk of age, the TBA implant was removed and they were reimplanted with Z every 10 wk until slaughter. The remaining eight calves were castrated at about 3 wk of age and implanted (IS) with TBA and Z every 10 wk until slaughter. Five calves had to be removed from this trial for the following reasons: three calves became chronic bloaters, one calf became totally blind, and one calf was unintentionally castrated. This left 5 CB, 9 IB and 6 IS for our study. The performance data on these calves were not included in the analysis of this study.

Calves were left with their dams on native bluestem pasture until weaning in early October at an average age of 7 mo. Calves were pre-conditioned for 4 wk prior to arrival at the Kansas State University Beef Cattle Research Unit on October 28, 1985. Initial feedlot weights were taken following a 12 h fast from feed and water. Over a 4 wk period, calves were fed continually increasing proportions of concentrate up to 89% concentrate (DM basis, table 2). Calves remained on this diet until slaughter.

Cattle within a treatment were assigned to two pens (partially covered, concrete floor, 4.3 x 8.5 m) so that the average weights for the pair of pens

TABLE 2. DIET COMPOSITION<sup>a</sup> FOR CONTROL BULLS  
AND IMPLANTED BULLS AND STEERS

Ingredient	Percent of ration
Grain sorghum (IFN 4-20-893)	82.96
Sorghum silage (IFN 3-04-323)	11.04
Soybean meal (IFN 5-20-637)	4.05
Ground limestone (IFN 6-02-632)	.75
Sodium chloride (IFN 6-04-152)	.36
Urea (IFN 5-05-070)	.34
DICAL (IFN 6-01-090)	.27
Potassium chloride (IFN 6-03-755)	.12
Soybean oil (IFN 4-05-077)	.06
Rumensin 60	.02
Trace minerals	.02
Vitamin A (30,000 IU/gm)	.01

<sup>a</sup>Dry matter basis.

were approximately equal. Individual weights were obtained at 28-d intervals with calves held off feed and water for 12 h before weighing. In addition, feed consumption was measured for each pen for all 28-d intervals for feed efficiency (FE) and average daily gain (ADG) calculations over the feedlot phase.

Cattle were slaughtered at an estimated 60 to 70% Choice grade endpoint. Realtime ultrasound was used to assist us in evaluating live animal composition. Eleven calves were slaughtered after 180 d on feed, and the remaining nine calves were slaughtered 30 d later. Calves were slaughtered at an average age of 13.6 mo.

Scrotal circumferences were measured at 8 and 13 mo, while hip heights were measured at 12 mo of age. Furthermore, masculinity was scored prior to slaughter by a four-member panel. The scoring scale ranged from 1=steer to 5=very masculine. The masculinity characteristics evaluated were based on appearance of the head, neck, forearm, jump (gluteus medius) muscle and overall proportion, relative to traits typical of average bulls at that age (Appendix I).

Blood Collection and Analyses. Blood samples were obtained from each calf beginning at about 5 wk of age and at 14-d intervals until slaughter. Blood was collected between 7 and 9 a.m. via jugular venipuncture into three 15-ml tubes, refrigerated for 24 h at 5 C, centrifuged to separate serum, and stored frozen at -20 C for subsequent analysis for concentrations of T and E<sub>2</sub> by radioimmunoassay. Testosterone concentrations were determined in duplicate 100- $\mu$ l (bulls) or 200- $\mu$ l (steers) aliquots of serum by the technique (Appendix II) previously described by Pruitt (1983). Testosterone extraction efficiency

was 88.9% and intra-assay and inter-assay coefficients of variability were 8.1 and 15.6%, respectively. Estradiol concentrations were determined in duplicate 5-ml aliquots of sera by the procedure (Appendix III) reported by Skaggs et al. (1986). Estradiol extraction efficiency was 77.1% and intra-assay and inter-assay coefficients of variability were 17.3 and 30.8%, respectively.

Slaughter, Carcass and Meat Sensory Traits. Cattle were transported approximately 200 km to a commercial packing plant where they were held approximately 4 h before slaughter. Upon slaughter, carcasses were shrouded and chilled overnight.

Testicles were weighed during the slaughter process. After a 24 h chill, USDA (1976) yield and quality grade data, carcass masculinity scores and longissimus quality traits were evaluated (Appendix IV). Carcass masculinity scores were based on jump muscle and crest development, whereas the longissimus muscle was scored for lean color, texture, firmness and presence of heat ring.

A wholesale rib from each carcass was shipped to Kansas State University and aged for 7 d at 2 C. Two steaks, 2.54-cm thick were removed from the 11th to 12th rib-region, double-wrapped in freezer paper and stored at -20 C until evaluated. The most posterior steak was designated for sensory panel (SP) analysis, while the adjacent steak was removed for Warner-Bratzler shear (WBS) force determination using an INSTRON universal testing machine.

Steaks used for both SP evaluations and WBS determinations were thawed overnight at 5 C and oven broiled at 166 C to an internal temperature of 70 C (monitored with thermocouples). A mechanical coring device was used to obtain uniform, 1.27-cm diameter longissimus cores which were excised

perpendicular to the steak surface. These cores were either served warm to a five-member, trained SP (AMSA, 1978), or allowed to equilibrate to room temp for 2 h prior to WBS determination (Appendix V). The panel rated juiciness, flavor intensity, myofibrillar tenderness, overall tenderness and connective tissue amount based on an 8-point scale (Appendix V). Evaluations were rated for six or seven steaks per session (three sessions), which were randomly selected, proportionately to the number representing each treatment.

Statistical Analyses. Data were analyzed by analysis of variance and means were separated using multiple t-tests calculated by the General Linear Model (GLM) Procedure of the Statistical Analysis System (SAS, 1982). The model used to evaluate treatment effects for all traits, except for endocrine traits, included adjustments for age and size differences as date of birth and hip heights were used as covariates in the analysis. Differences between treatments for performance, carcass and meat sensory traits in which there was an overall treatment effect ( $P < .10$ ) were evaluated by Least Squares Means Procedures (SAS, 1982). Results were reported as least-squares means with appropriate standard errors (SE).

Serum hormone concentrations were analyzed by a split-plot variance with repeat measures. Treatments served as the main plot and 14 d bleeding periods were used as subplots. Main-effect means were separated by Fisher's LSD when the respective F-tests were significant ( $P < .05$ ) using appropriate error terms for split plot analysis (Steel and Torrie, 1960). Results were reported as means with appropriate SE.



## Results and Discussion

Animal Performance. Performance data are presented in table 3. Both IB and IS tended ( $P=.07$ ) to be heavier at slaughter time than CB. All groups started the feeding phase with similar weaning weights and were similar in feedlot ADG and FE.

Neither Fisher et al. (1986b) nor Silcox et al. (1986) reported differences in performance between bulls implanted with a combination of TBA plus Z or  $E_2$  compared with CB, whereas Fisher et al. (1986a) found that steers implanted with TBA plus hexestrol had higher ADG than either IB or CB during the finishing phase and tended to be heavier than CB at slaughter. Our results tend to support their observations. Numerous researchers have demonstrated that TBA plus an estrogenic agent improved performance in young bulls (Grandadam et al., 1975; Galbraith, 1982; Bouffault and Willemart, 1983; Fabry et al., 1983) and implanted steers (Heitzman, 1978; Galbraith and Geraghty, 1982; Bouffault and Willemart, 1983; Loblely et al., 1985; Steen, 1985). Performance traits of bulls implanted with Z near birth, and sequentially thereafter were unaffected (Calkins et al., 1986; Gray et al., 1986) or increased (Greathouse et al., 1983) compared with CB. This seems to agree with the conclusion of Baker and Arthaud (1972) that administration of estrogenic agents to bulls results in inconsistent effects on performance.

Animal Measurements. Hip heights and masculinity traits are presented in table 4. The IS tended ( $P=.09$ ) to have lower hip heights at 12 mo of age than either bull group. This tendency may have been due to random error in assigning calves to treatments at birth. Furthermore, IS tended ( $P<.10$ ) to be

Table 3. PERFORMANCE TRAITS OF CONTROL BULLS AND IMPLANTED BULLS AND STEERS

Trait	Treatments			SE
	Implanted		Control Bulls	
	Steers	Bulls		
Weaning wt., kg	214	218	205	9.20
Slaughter wt., kg	514 <sup>a</sup>	517 <sup>a</sup>	479 <sup>b</sup>	10.90
Average daily gain, kg	1.6	1.5	1.4	.05
Feed/Gain (DM basis)	5.2	5.4	5.2	.09

<sup>a,b</sup> Means in the same row with different superscript letters differ ( $P < .07$ ).

Table 4. HIP HEIGHTS AND MASCULINITY CHARACTERISTICS FOR CONTROL BULLS AND IMPLANTED BULLS AND STEERS

Trait	Treatments			SE
	Implanted		Control Bulls	
	Steers	Bulls		
Hip height at 12 mo., cm	114 <sup>c</sup>	118 <sup>cd</sup>	121 <sup>d</sup>	1.99
Masculinity score at 13 mo. <sup>1</sup>	2.3 <sup>c</sup>	3.0 <sup>d</sup>	3.2 <sup>d</sup>	.24
Scrotal circ. at 8 mo., cm	--	20.9 <sup>a</sup>	25.9 <sup>b</sup>	.71
Scrotal circ. at 13 mo., cm	--	34.5 <sup>a</sup>	38.8 <sup>b</sup>	1.04
Testicular wt., g	--	294 <sup>a</sup>	398 <sup>b</sup>	15.62

<sup>1</sup> Scores of 1 to 5: 2=slightly masculine, 3=moderately masculine, 4=masculine.

a,b Means in the same row with different superscript letters differ (P<.05).

c,d Means in the same row with different superscript letters differ (P<.10).

less masculine than either CB or IB, but our implant scheme for bulls did not suppress masculinity. Other studies involving bulls implanted with Z after weaning have reported that IB are similar to CB in masculinity appearance (Ford and Gregory, 1983; Price et al., 1983; Unruh et al., 1983). In contrast, implanting bulls with estrogenic agents from near birth until slaughter has reduced masculinity development (Ralston, 1978; Unruh et al., 1983, 1986a; Hopkins, 1986). Therefore, time of implanting and type of steroid implant used may be important factors in affecting masculinity development.

Scrotal circumferences (SC) were smaller ( $P < .05$ ) for IB at both 8 and 13 mo and testicle weights (TWT) were also lighter ( $P < .05$ ) than for CB. These results agree with previous studies in which bulls implanted with Z near birth had smaller SC and TWT (Corah et al., 1979; Unruh et al., 1983, 1986a; Staigmiller et al., 1985; Silcox et al., 1986) than CB. However, few differences in testicle development have been observed between CB and bulls implanted beginning after approximately 6 mo of age (Unruh et al., 1983; Staigmiller et al., 1985; Vanderwert et al., 1985). Furthermore, Unruh et al. (1986a) reported that development of these traits in Simmental bulls was delayed up to 15.7 mo in IB compared to CB. However, at 17.4 mo of age, IB had attained testicle development similar to CB. Silcox et al. (1986) reported that Angus bulls initially implanted with Z at 175 d and reimplanted at 231 d with either Z, TBA or a combination of the two resulted in smaller SC and non-significantly lighter TWT than non-implanted bulls. The variability in results caused by different ages at time of initial implantation may partially stem from the pubertal maturation of the hypothalamic-hypophyseal-gonadal axis of bulls (Amann and Schanbacher, 1983). Thus Silcox et al. (1986) concluded that Z implantation prior to 200 d of age may arrest pubertal

maturation of bulls by altering LH secretion, which influences T production which may influence testicular growth and masculinity development.

Hormonal Concentrations. Serum T levels are illustrated in figure 1. Since steers had very low levels of T (equal to the lower limit of assay sensitivity), their values were not included in figure 1. Implanting bulls with TBA and Z early in life, and reimplanting with Z alone after 5 mo of age resulted in very low T levels (near assay sensitivity) until 10 mo of age. However, T increased ( $P < .05$ ) substantially from 11 mo of age until slaughter at 13.6 mo, and was higher ( $P < .05$ ) than for CB from 12 to 13.6 mo of age. Nevertheless, CB had higher ( $P < .05$ ) T levels than IB from 7.5 to 11 mo of age. The observed suppression of T may be related to the age of the bulls at the time of first implantation. Silcox et al. (1986) found that bulls implanted at 100 or 150 d of age with Z (reimplanted at 56 d intervals) had reduced testicular growth and T compared to those initially implanted at 250 d, which were similar to non-implanted bulls. Our findings are in agreement with those previously reported for Z (Staigmiller et al., 1985; Gray et al., 1986), TBA plus Z (Fabry et al., 1983; Silcox et al., 1986) and  $E_2$  (Schanbacher et al., 1983; Hopkins, 1986). McCarthy et al. (1979) and Schanbacher et al. (1983) both reported that increased serum T concentration was the most pronounced hormonal change during sexual maturation in bulls. Therefore, it appears that these growth implants, if administered early in life, can delay the time of sexual maturation.

Serum  $E_2$  levels are illustrated in figure 2. There were no clear-cut differences among treatments. However, within treatment observations revealed evidence of periodic fluctuations for all treatments. Similarities did

Figure 1. Serum testosterone concentrations of control bulls (blue, O, - - -) and implanted bulls (red,  $\Delta$ , —). Values are reported as ng/ml with SE represented by treatment symbol above and below the mean for each biweekly bleeding period. Least significant difference (LSD) ( $P < .05$ ) within treatment = 1.92 ng/ml. Between treatment LSD ( $P < .05$ ) = 2.05 ng/ml.

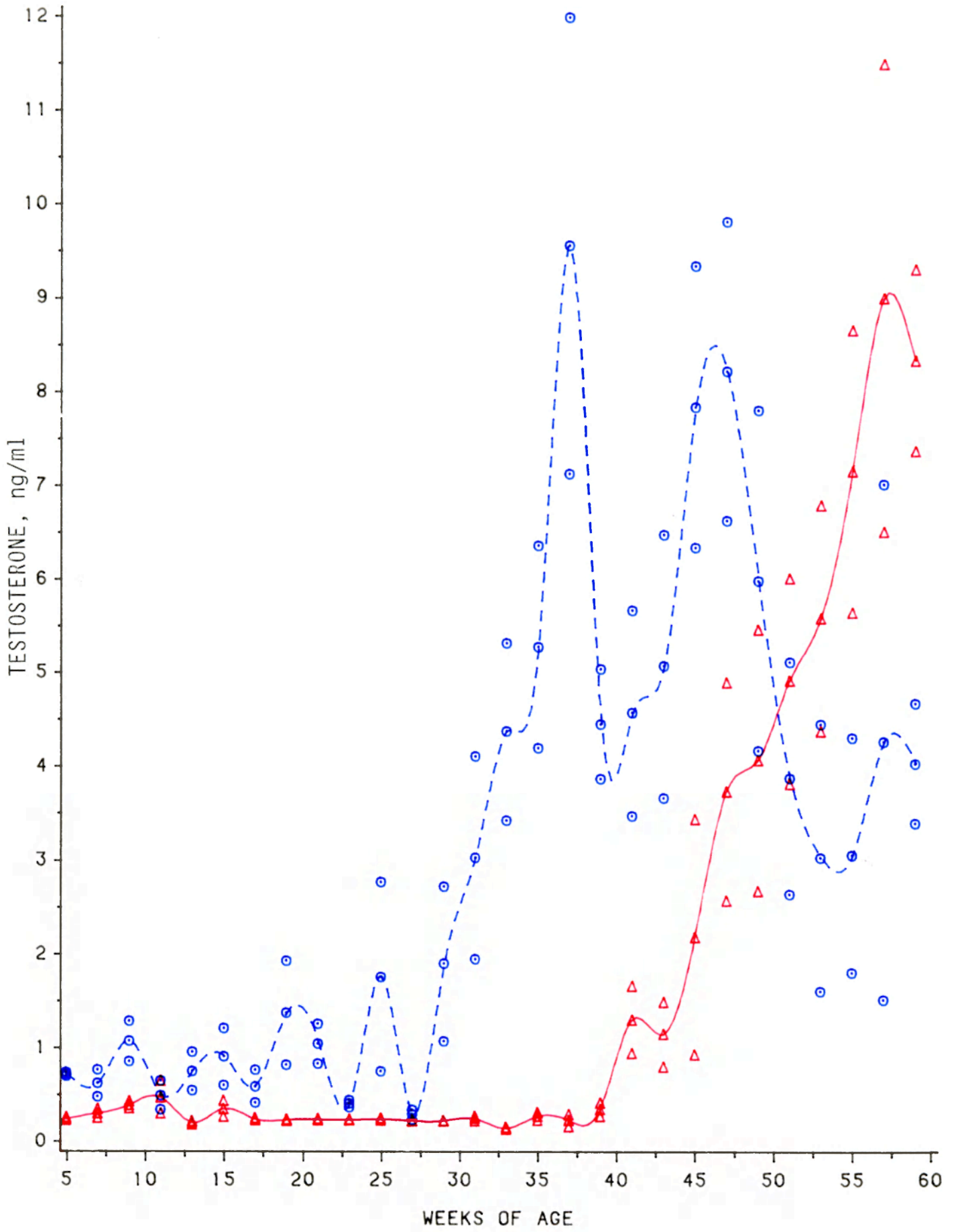
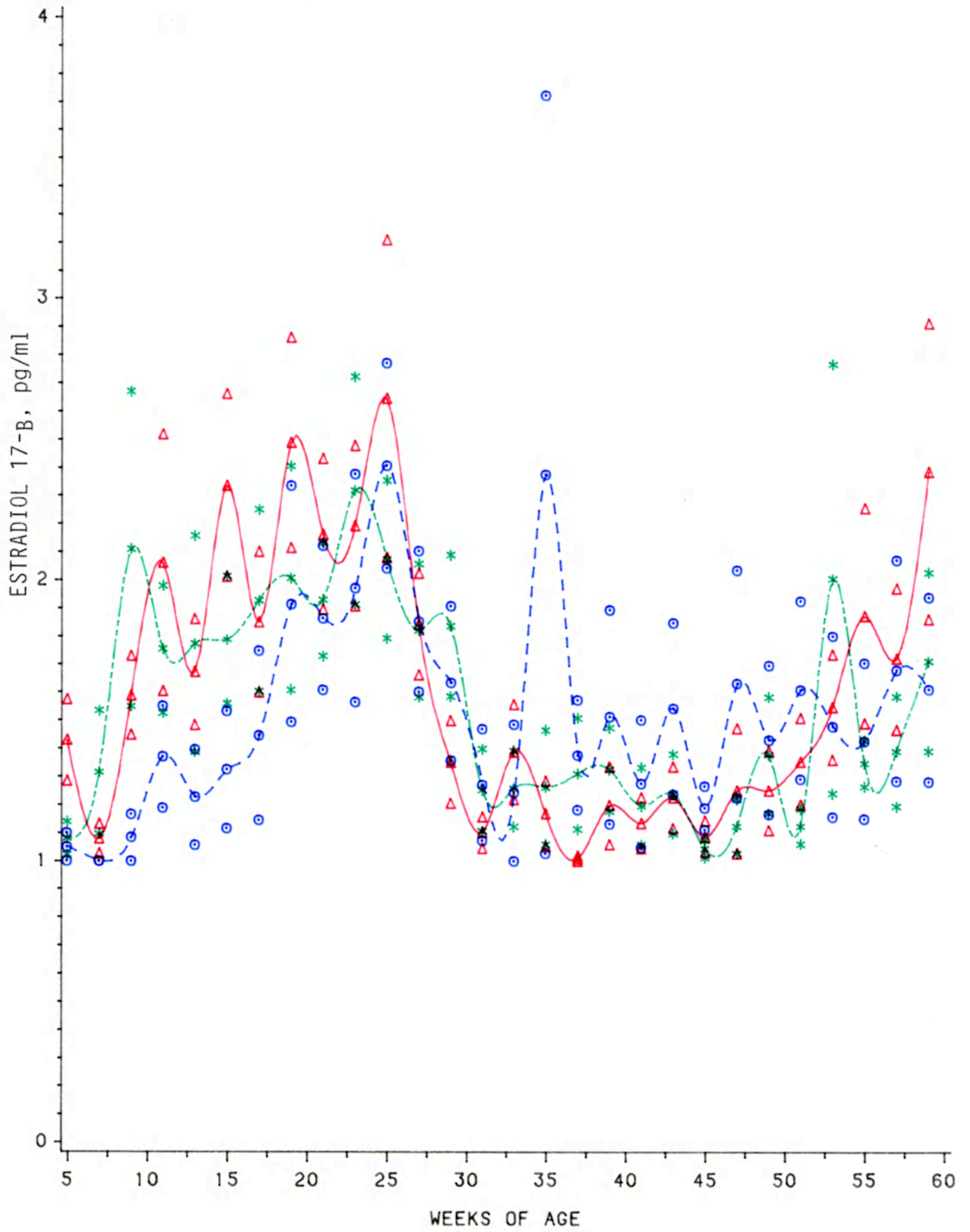


Figure 2. Serum estradiol concentrations of control bulls: (blue, O, - - - - ); implanted bulls: (red,  $\Delta$ , ———); implanted steers: (green, \*, — - - —). Values are reported as pg/ml with SE represented by treatment symbol above and below the mean for each biweekly period. LSD ( $P < .05$ ) within treatment = .69 pg/ml. Between treatment LSD ( $P < .05$ ) = .83 pg/ml.





exist for all treatments at certain time periods. Gray et al. (1986) found that bulls implanted with Z had lower  $E_2$  levels from 8.3 to 13 mo than non-implanted bulls but had higher levels from 14 to 15 mo of age. After 15 mo, both groups were similar until slaughter at 17.4 mo. In addition, Schanbacher et al. (1983) reported that non-implanted bulls had serum  $E_2$  levels over twice that of non-implanted steers at 12.5 mo of age. Our results seem to contradict these reports. Zeranol, a resorcylic acid-lactone known to have estrogenic activity, may have been recognized as  $E_2$  in quantities sufficient to result in feedback inhibition on  $E_2$  secretion. In support, Fabry et al. (1983) showed that TBA plus Z eliminated the pulsatile surges of luteinizing hormone (LH) in Belgian White-Blue bulls. Although,  $E_2$  was not measured in their study, it may be speculated that suppression of pulsatile LH secretion may reduce testicular  $E_2$  secretion similar to the way it suppressed T, which they did measure. Nevertheless, this reasoning cannot address why the CB in our study had such low levels of  $E_2$  ( $1.5 \pm .3$  pg/ml); whereas, Schanbacher et al. (1983) reported much higher values for non-implanted bulls and steers ( $9 \pm 2$  and  $4 \pm 1$  pg/ml, respectively); and Gray et al. (1986) stated that values for IB and CB were 6 to 8 pg/mo from 12 to 15 mo of age. The bulls used in our study were small-framed cattle and seemed to be very non-aggressive and exhibited low libido. Their low  $E_2$  levels may be attributed to behavioral characteristics.

Carcass Characteristics. Examination of carcass characteristics (table 5) shows that there were no treatment differences. Silcox et al. (1986) reported that hot carcass weights (HCW) for non-implanted bulls were similar to those of bulls implanted with Z, TBA, or Z plus TBA. Steen (1985) found that bulls

Table 5. CARCASS CHARACTERISTICS OF CONTROL BULLS AND IMPLANTED BULLS AND STEERS

Trait	Treatments			SE
	Implanted		Control Bulls	
	Steers	Bulls		
No. animals	6	9	5	-
Hot carcass wt., kg	306	311	291	7.20
Dressing percent	59.5	60.1	60.7	.61
Carcass maturity	A <sup>52</sup>	A <sup>53</sup>	A <sup>55</sup>	4.79
Marbling score	Small <sup>07</sup>	Small <sup>00</sup>	Slight <sup>82</sup>	.14
Fat thickness, cm	1.2	1.0	.9	.10
Ribeye area, cm <sup>2</sup>	81.3	83.2	85.2	3.72
Kidney knob, %	1.5	1.3	1.5	.15
Yield grade	2.5	2.3	1.9	.20
Jump muscle and crest score <sup>1</sup>	1.3	1.6	1.5	.14

<sup>1</sup> Scores of 1 to 6: 2=barely evident, 3=slightly prominent, 4=moderately prominent.

implanted with Z had heavier HCW than steers implanted with Z plus TBA, although dressing percentages were similar for both groups. Nevertheless, implanting bulls with TBA plus an estrogenic agent has been shown to increase HCW (Grandadam et al., 1975; Galbraith, 1982). In contrast, other studies have shown that implanting bulls with Z has not affected HCW (Vanderwert et al., 1985; Gray et al., 1986; Johnson et al., 1986).

The lack of differences in carcass maturity was somewhat expected, because calves were slaughtered at a young age (13.6 mo of age). Implanting bulls with Z did not alter overall carcass maturity relative to controls (Greathouse et al., 1983; Johnson et al., 1986; Unruh et al., 1986b) or to advance maturity in bulls more than in steers (Vanderwert et al., 1985). Furthermore, Unruh et al. (1986b) stated that the largest mean differences for carcass maturity advancement occurred from 13.8 to 15.7 mo of age in Simmental bulls when measurements began when calves were 12.0 mo of age and were concluded at 17.4 mo. Therefore, at typical slaughter ages of 14 to 17 mo of age, carcass maturity may change only slightly due to implant scheme.

Steers generally have higher marbling scores than bulls (Seideman et al., 1982; Gregory and Ford, 1983; Vanderwert et al., 1985; Calkins et al., 1986). When bulls were implanted with Z near puberty (Ford and Gregory, 1983; Gregory and Ford, 1983; Johnson et al., 1984; Johnson et al., 1986), implanting had no effect on marbling. Silcox et al. (1986) found that bulls implanted with Z or Z plus TBA were similar to CB in marbling. However, enhanced marbling was observed in studies when bulls were implanted near birth with Z, and reimplanted sequentially until slaughter (Calkins et al., 1986; Unruh et al., 1986b). Furthermore, Fisher et al. (1986b) reported that

bulls implanted with TBA plus  $E_2$  had a higher muscle lipid content than non-implanted bulls, whereas implanting steers with these compounds appeared to reduce muscle lipid content. Studies to investigate the intramuscular fat deposition response of TBA plus estrogenic agents, particularly in steers, will likely increase, since TBA was recently approved in the U.S.

Although IS had higher yield grade (YG) numbers than CB, these groups were not different ( $P > .10$ ). However, other studies have shown that bulls generally have a lower fat thickness (FTH), larger ribeye area (REA), and similar percentage of kidney, pelvic and heart (KPH) fat than steers which results in bulls having an advantage in YG (Crouse et al., 1985; Vanderwert et al., 1985). Steen (1985) reported that Z implanted bulls were trimmer than both non-implanted steers and steers implanted with Z plus TBA. Furthermore, Silcox et al. (1986) found that REA and FTH were similar for non-implanted bulls and bulls implanted with Z or Z plus TBA. In contrast, Calkins et al. (1986) and Unruh et al. (1986b) both reported increased FTH and higher numerical YG for bulls implanted near birth with Z than for CB. It appears that time of implantation influences carcass composition, particularly with regard to fat deposition in young bull carcasses.

Carcass masculinity, as measured by jamp muscle and crest development, was scored fairly low for all treatments. Consequently, no differences were attributable to sex or implant scheme. This might have been expected as calves were slaughtered at 13.6 mo of age and secondary-sex characteristics may not have been fully developed for these bulls. Arthaud et al. (1977) stated that at 15 to 18 mo of age the crest on bulls was moderately to prominently developed. Fisher et al. (1986a) found that there was no effect on the muscle weight distribution of Friesian bulls implanted with Z or TBA plus hexestrol.

However, muscles from the forequarter were developed to a greater extent than for steers implanted with Z or TBA plus hexesterol. However, young Simmental bulls implanted with Z or  $E_2$  near birth were less masculine than non-implanted bulls (Unruh et al., 1986a; and Hopkins, 1986, respectively). Furthermore, Hopkins (1986) found that IS were less masculine than IB or CB.

Longissimus quality traits (lean firmness, texture, color and absence of heat ring) were very desirable, regardless of sex or implant scheme, and no differences resulted from treatment (table 6). Unruh et al. (1986b) also found that lean firmness, texture and color were not different for IB and CB. The incidence of heat ring in our study was negligible. This was likely due to effects of adequate insulation (fat cover) over the ribeye muscle during chilling (Riley et al., 1983).

Longissimus Sensory Traits. Implanting bulls and steers near birth until slaughter had no significant effects on SP flavor intensity, juiciness, myofibrillar or overall tenderness and WBS values (table 7). However, connective tissue amount tended ( $P=.07$ ) to be higher for CB than for IB or IS. This may be linked to the early higher levels of T observed in CB, which may have hastened the collagen maturation process (Hall, 1976). Although IS and CB were not different ( $P>.10$ ), it appears that tenderness-related traits may have been slightly higher for IS. Previous studies report no differences for SP palatability traits when bulls were implanted with Z initially during the finishing phase (Gregory et al., 1983; Johnson et al., 1984; Johnson et al., 1986). However, implanted bulls were rated similar to IS for SP flavor intensity, connective tissue amount, myofibrillar tenderness and overall tenderness when implanted near birth with Z (Gray et al., 1984) or  $E_2$

Table 6. RIBEYE (LONGISSIMUS) QUALITY CHARACTERISTICS FOR CONTROL BULLS AND IMPLANTED BULLS AND STEERS

Trait	Treatments			SE
	Implanted		Control Bulls	
	Steers	Bulls		
Lean firmness <sup>1</sup>	6.4	5.8	5.9	.40
Lean texture <sup>2</sup>	5.6	4.9	4.3	.36
Lean color <sup>3</sup>	4.0	4.1	4.3	.34
Heat ring (dark coarse band) <sup>4</sup>	1.1	1.0	1.0	.05

<sup>1</sup> Scores of 1 to 8: 5=slightly firm, 7=firm.

<sup>2</sup> Scores of 1 to 8: 4=slightly coarse, 5=slightly fine.

<sup>3</sup> Scores of 1 to 9: 3=light cherry red, 4=cherry red.

<sup>4</sup> Scores of 1 to 5: 1=none, 2=slight.

Table 7. TASTE PANEL EVALUATIONS AND WARNER-BRATZLER SHEAR VALUES OF RIBEYE (LONGISSIMUS) STEAKS FOR CONTROL BULLS AND IMPLANTED BULLS AND STEERS

Trait	Treatments			SE
	Implanted Steers	Implanted Bulls	Control Bulls	
Flavor intensity <sup>1</sup>	6.3	6.3	6.1	.18
Juiciness <sup>1</sup>	6.4	6.3	6.1	.11
Connective tissue amount <sup>2</sup>	7.2 <sup>a</sup>	7.1 <sup>a</sup>	6.7 <sup>b</sup>	.12
Myofibrillar tenderness <sup>3</sup>	6.4	6.2	5.4	.32
Overall tenderness <sup>3</sup>	6.5	6.4	5.9	.30
Warner-Bratzler shear, kg.	3.3	3.4	4.0	.29

<sup>1</sup> 6=Slightly intense or slightly juicy, 7=very intense or very juicy.

<sup>2</sup> 6=Slight amount, 7=practically none.

<sup>3</sup> Scores of 1 to 8: 5=slightly tender, 6=moderately tender.

<sup>a,b</sup> Means in same row with different superscripts differ (P<.07).



(Hopkins, 1986). Based on our results, life-long implanting had no negative effects on SP traits for IB which were as desirable as IS. However, since no control group of steers was available for this trial, and implant studies involving TBA have not included SP evaluation, we cannot speculate on the effects that TBA plus Z has on the meat sensory traits of implanted steers compared with those non-implanted, or those implanted with compounds more commonly used in the U.S.

Conclusions. The results of our study indicate that implanting young bulls near birth with TBA plus Z and then Z alone after about 5 mo of age did not affect performance, except that IB tended ( $P=.07$ ) to be heavier at slaughter. Furthermore, IB had smaller ( $P<.05$ ) SC and lighter ( $P<.05$ ) TWT weights than CB and serum T was suppressed ( $P<.05$ ) from 7.5 to 11 mo of age, but was higher ( $P<.05$ ) from 12 to 13.6 mo of age. Implanting bulls also resulted in ribeye steaks that had less ( $P=.07$ ) detectable connective tissue. The implanting scheme of TBA and Z early in life and then Z alone, may counteract some of the problems associated with feeding bulls for meat production.

Implanting steers near birth and until slaughter with TBA plus Z resulted in performance traits similar to bulls, along with very desirable carcass characteristics and SP palatability ratings.

For small-framed cattle such as those used in our study, castration and implantation with TBA plus Z near birth may optimize performance, carcass and meat quality traits.

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Chapter IV

APPENDICES

## Appendix I

### Live Masculinity Score Descriptions<sup>a</sup>

1. Steer: no prominent facial features over the eye brow and jaw; narrow and long head in relation to body size; refined head crest and neck; smooth, little muscular development of crest, shoulder, rib and hindquarter; and fine hair coat texture, especially over the head.
2. Slightly masculine: slight prominence of eye brow and facial features; somewhat narrow and long head in relation to body size; somewhat refined head, crest, and neck; somewhat smooth and moderate muscular development; and moderately fine hair coat.
3. Moderately masculine: somewhat prominent brow and facial features; moderate relationship of length and width of head to body size; slight fullness of head, crest and neck; moderate muscular development of crest, shoulder, rib and hindquarter; and slightly coarse hair coat.
4. Masculine: prominent brow and facial features; moderately wide head in relation to length of head; full head, crest and neck; muscular through crest, shoulder, rib and hindquarter; and coarse hair texture.
5. Very masculine: very prominent eye brow and facial features; relatively wide head in relation to length; very full head, crest and neck; high degree of muscular development through the crest, shoulder, rib and hindquarter; and very coarse hair texture.

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<sup>a</sup> Scores made relative to the normal development of average young bulls at the observed age.

## Appendix II

### TESTOSTERONE RADIOIMMUNOASSAY

1. Thaw standards, serum samples, antiserum, and standard serum overnight in refrigerator (5°C).
  2. To estimate recovery of testosterone, during solvent extraction of verum, add 10  $\mu$ l  $^3$ H-1, 2, 6, 7-testosterone ( $^3$ H-T) in 95 % EtOH (~2,500 cpm/10 $\mu$ l) to six 16 x 100 mm disposable culture tubes (Tracer tubes). Add 10  $\mu$ l  $^3$ H-T to four scintillation vials (7 ml minivials) and set aside to air dry (total count tubes).
  3. Add appropriate dilution (50, 100, or 200  $\mu$ l) of serum in duplicate to 10 x 100 mm tubes and aliquot random serum dilutions to Tracer tubes (T).
  4. Extract serum with minimum of 10 volumes (10x serum dilution) ethyl acetate. Vortex for 5 min.
  5. Freeze extracts (~30 min) and decant solvent phase into 12 x 75 mm disposable culture tubes. Dry down extract under air in 40°C water bath. Decant tracer tubes directly into scintillation vials and set aside to air dry.
  6. Prepare standard curves of testosterone<sup>a</sup> by pipetting 0.1 ml of each standard concentration (1 ml total/tube) in duplicate 12 x 75 mm tubes which corresponds to 2000, 1500, 1000, 750, 500, 250, 100, 50, 25 and 10 pg/tube. NOTE: BE CERTAIN that standards in EtOH are at room temperature before pipeting volumes! Pipets are calibrated for accuracy when solutions are at room temperature (i.e., 22-26°C).
  7. Include Hot-Buffer tubes (HBT, 0.2 ml assay tracer only), nonspecific binding tubes (NSB; 0.2 ml phosphate-buffer saline-gelatin (PBSG) in place of antiserum and 0.2 ml assay tracer), and zero tubes (0, 0 standard-0.2 ml assay tracer and 0.2 ml antiserum).
  8. Add 0.2 ml testosterone antiserum<sup>b</sup>  
0.2 ml  $^3$ H-T<sup>c</sup>  
0.1 ml PBSG<sup>d</sup>  
to all samples 0 tubes, and standards.
  8. Add 0.2 ml  $^3$ H-T and 0.8 ml PBSG to HBT tubes.  
Add 0.2 ml  $^3$ H-T and 0.2 ml PBSG to NSB tubes.
  10. Vortex all tubes for 2-3 sec, incubate overnight (14-18 h) at 5°C covered with foil or plastic wrap.
  11. Pipet 0.5 ml ice-cold dextran-coated charcoal (DCC) solution<sup>e</sup> into all tubes except HBT tubes. Add DCC to approximately 144 tubes at a time, vortex (2-3 sec) in backwards order (i.e., first tubes to receive DCC are the last ones vortexed), and load into centrifuge to begin centrifugation within 10 min after beginning of addition of DCC.
  12. Centrifuge at 5°C for 10 min at 1,500 x g (2,400 rpm).
  13. Pipet 0.5 ml supernatant into scintillation vials. Do not contaminate supernatant by pipetting some off the DCC pellet. Add 5 ml scintillation fluid to each vial. Cap, label, and vortex for 1 min on shaker.
  14. Count for 4 min in liquid scintillation ( $\beta$ )-counter.
  15. Add scintillation fluid (5 ml) to tracer and total count tubes and handle as in 14.
-



<sup>a</sup> Testosterone (Sigma, T-1500) standards are made from a stock solution of 100 ng/ml 95% EtOH, dried down, and reconstituted in PBSG. They are stored frozen in 1 ml aliquots in concentrations of 10, 25, 50, 100, 250, 500, 750, 1,000, 1,500, and 2,500 pg/0.1 ml.

<sup>b</sup> Stock solution of testosterone antiserum is diluted to give 45% (40-50%) binding in B<sub>0</sub> or 0 tubes. Approximate dilution is 1:20,000.

<sup>c</sup> Stock solution of assay tracer is made by diluting <sup>3</sup>H-T in PBSG to achieve 16,000 cpm/0.2 ml assay buffer (PBSG).

<sup>d</sup> PBS is made by combining the following phosphate buffers to achieve pH 7.4.

(1) 0.01 M phosphate-buffered saline, monobasic "A"

To a 1000 ml volumetric flask, add:

1.38 g Na<sub>2</sub>PO<sub>4</sub>·H<sub>2</sub>O

8.7 g NaCl

0.1 g Thimerosal (antibacterial compound)

Add double-distilled deionized water to 1000 ml mark, invert to mix or add stirring bar. Store indefinitely at 5°C.

(2) 0.01 M phosphate-buffered saline, dibasic "B"

To a 1000 ml volumetric flask, add:

1.42 g Na<sub>2</sub>HPO<sub>4</sub>

8.7 g NaCl

0.1 g Thimerosal

Add double-distilled deionized water to 1000 ml mark, invert to mix or add stirring bar. Store indefinitely at 5°C.

PBSG is made by combining "A" and "B" buffers to pH 7.4 (approx. 6B:1A, v/v). Add 0.1% gelatin (Sigma, G-2500), heat and stir to dissolve. Do not heat over 30°C. Store at 5°C.

<sup>e</sup> Dextran-coated charcoal (DCC). Add 1.0% dextran T-70 (Pharmacia) to double-distilled deionized water and stir until dissolved. Add 0.5% charcoal (Norit, A, Fisher Scientific). Make-up fresh suspension for each assay. Store at 5°C until needed. When adding to assay tubes, stir continuously.

<sup>f</sup> Complete counting cocktail 3a70B (Research Products International Corp.)

### Appendix III

#### ESTRADIOL RADIOIMMUNOASSAY

1. Remove antiserum, serum samples, and standard serum from freezer and thaw overnight in refrigerator at 5°C. Standards can be removed a few hours before needed.
2. Add 10 µl <sup>3</sup>H-2, 4, 6, 7 estradiol (~ 2,500 cpm/10 µl) to six 20 x 150 mm tubes (random tracers) to four scintillation vials to estimate recovery of estradiol from solvent extraction.
3. Add serum (5 ml) to 20 x 150 mm tubes in duplicate and to random tracer tubes.
4. Extract serum with 2 volumes ethyl acetate (2x serum volume). Vortex for 5 min on shaker.
5. Freeze extraction tubes to separate solvent from aqueous phases (~ 60 min).
6. Decant solvent into 16 x 100 mm tubes. BE CAREFUL to not allow the aqueous pellet to thaw (i.e., remove only a dozen or so tubes from the freezer at one time). If thawing occurs, simply refreeze.
7. Dry down solvent in 40°C water under air. (This will require approx 60 to 90 min depending on the volume). Thaw pellet. Re-extract with 2x vol. of solvent. Vortex 1 min. Freeze and decant into same 16 x 100 mm tube as in 6.
8. When dry, rinse down inside of tubes with an additional 1 ml ethyl acetate. Vortex 10 sec and dry down again.
9. Rinse down tubes again with 1 ml ethyl acetate and then carefully decant into 12 x 75 mm disposable culture tubes and air-dry in water bath.
10. Decant tracer tubes directly into scintillation vials and set aside to air dry.
11. Remove standard stock estradiol<sup>a</sup> (1 ng/ml) from freezer or if already done so, be certain it is room temperature (22-26°C) before pipetting dilution from standard concentrations. Pipet in duplicate the following volumes for standards: 5, 10, 15, 30, 60, 120, 250, and 500 µl which corresponds to 5, 10, 15, 30, 60, 120, 250, and 500 pg/tube. Dry down in water bath under air as described above.
12. Include Hot Buffer tubes (HBT; 0.2 ml assay tracer), nonspecific binding tubes (0; 0 standards, 0.2 ml antiserum, and 0.2 ml assay tracer).
13. Make-up estradiol antiserum<sup>b</sup> in PBSG.

14. Add 0.2 ml antiserum to all sample tubes, 0 tubes, and to all standard tubes. Add equivalent amount of buffer to NSB tubes. Vortex and then incubate for 30 min at room temperature.
15. Dilute  $^3\text{H}$ -estradiol<sup>d</sup> in PBSG and add 0.2 ml to all samples, 0, and standard tubes. Vortex and incubate overnight (14 to 18 h) at 5°C covered with aluminum foil or plastic wrap. (Add 0.2 ml  $^3\text{H}$ -E<sub>2</sub> and 0.8 ml PBSG to HBT tubes and 0.2 ml  $^3\text{H}$ -E<sub>2</sub> and 0.3 ml PBSG to NSB tubes).
16. Pipet 0.5 ml ice-cold dextran-coated charcoal<sup>c</sup> (DCC) solution into all tubes except HBT tubes. Add to tubes in one direction and vortex (2-3 sec) in backwards order. Within 10 min of adding DCC to tubes, centrifugation step should begin.
17. Centrifuge at 1,500 x g (2,400 rpm) at 5°C for 10 min.
18. Pipet 0.5 ml supernatant fluid into 7-ml scintillation vials add 5 ml scintillation fluid, cap, label, shake for 1 min.
19. Add scintillation fluid<sup>1</sup> (5 ml) to tracer and total count tubes after they have dried down.
20. Count all vials for 4 min in liquid scintillation (β) counter.

<sup>a</sup> Estradiol-17β (Sigma, E-8875) was prepared by serially diluting 10 mg to a stock 1 ng/ml solution in 95% EtOH.

<sup>b</sup> Stock solution of estradiol antiserum is diluted to give 50% (45-55%) binding in β<sub>0</sub> or 0 tubes. Approx dilution 1:35,000.

<sup>c</sup> PBS is made by combining the following phosphate buffers to achieve pH 7.4.

1. 0.01 M phosphate-buffered saline, monobasic "A"  
To a 1000 ml volumetric flask, add:  
1.38 g Na<sub>2</sub>PO<sub>4</sub>·H<sub>2</sub>O  
8.7 g NaCl  
0.1 g Thimerosa (antibacterial compound)

Add double-distilled deionized water to 1000 ml mark, invert to mix or add stirring bar. Store indefinitely at 5°C.

2. 0.01 M phosphate-buffered saline, dibasic "B"  
To a 1000 ml volumetric flask, add:  
1.42 g Na<sub>2</sub>HPO<sub>4</sub>  
8.7 g NaCl  
0.1 g Thimerosal

Add double-distilled water to 1000 ml mark, invert or stir with stirring bar, and store indefinitely at 5°C.

PBSG is made by combining "A" and "B" buffers to pH 7.4 (approx 6B:1A, v/v). Add 0.1% gelatin (Sigma, G-2500), heat and stir to dissolve (do not heat above 30°C). Store at 5°C for no more than 2 weeks.

<sup>d</sup> Stock stock solution of assay tracer is made by diluting <sup>3</sup>H-2, 4, 6, 7-estradiol (Amersham TRK.322) to PBSG to achieve 16 to 18,000 cpm/0.2 ml.

<sup>e</sup> Dextran-coated charcoal (DCC). Add 1% dextran T-70 (Pharmacia) to double-distilled deionized water and stir until dissolved. Add 1.5% charcoal (Norit A, Fisher Scientific) and stir until dissolved. Make-up fresh suspension for each assay. Store at 5°C until needed. When adding to assay tubes, stir continuously.

<sup>f</sup> Complete counting cocktail 3a70B (Research Products International Corp.).

## Appendix IV

### Carcass Evaluation Score Descriptions

#### Jump Muscle and Crest

- 1=none
- 2=barely evident
- 3=slightly prominent
- 4=moderately prominent
- 5=prominent
- 6=very prominent

#### Longissimus Quality

##### Lean Firmness

- 1=very soft
- 2=soft
- 3=moderately soft
- 4=slightly soft
- 5=slightly firm
- 6=moderately firm
- 7=firm
- 8=very firm

##### Lean Color

- 1=pale red
- 2=very light cherry red
- 3=light cherry red
- 4=cherry red
- 5=slightly dark red
- 6=moderately dark red
- 7=dark red
- 8=very dark red
- 9=black

##### Lean Texture

- 1=very coarse
- 2=coarse
- 3=moderately coarse
- 4=slightly coarse
- 5=slightly fine
- 6=moderately fine
- 7=fine
- 8=very fine

##### Heat Ring (Dark Coarse Band)

- 1=none
- 2=slight
- 3=moderate
- 4=severe
- 5=extremely severe

## Appendix V

### Sensory Panel Evaluation Score Descriptions

#### Flavor intensity

1=extremely bland  
2=very bland  
3=moderately bland  
4=slightly bland  
5=slightly intense  
6=moderately intense  
7=very intense  
8=extremely intense

#### Juiciness

1=extremely dry  
2=very dry  
3=moderately dry  
4=slightly dry  
5=slightly juicy  
6=moderately juicy  
7=very juicy  
8=extremely juicy

#### Connective tissue amount

1=abundant  
2=moderately abundant  
3=slightly abundant  
4=moderate  
5=slight  
6=traces  
7=practically none  
8=none

#### Myofibrillar and overall tenderness

1=extremely tough  
2=very tough  
3=moderately tough  
4=slightly tough  
5=slightly tender  
6=moderately tender  
7=very tender  
8=extremely tender

### INSTRON 4201 WBS SHEAR FORCE DETERMINATION PROCEDURES

#### Item

Cross Head  
Chart  
Load Range  
Load Cell

#### Technical Setting

250 mm/min  
200 mm/min  
25%  
50 kg

PERFORMANCE, CARCASS, MEAT SENSORY AND ENDOCRINE  
TRAITS OF YOUNG BULLS AND STEERS IMPLANTED  
WITH TRENBOLONE ACETATE AND ZERANOL

by

Robert D. Johnson

B.S., Purdue University,  
West Lafayette, IN 1985

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AN ABSTRACT OF A MASTER'S THESIS

Submitted in partial fulfillment of the  
requirements for the degree

MASTER OF SCIENCE

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KANSAS STATE UNIVERSITY  
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1987

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88-AD

Twenty-five Polled Hereford calves were assigned randomly to one of three treatments at birth. Eight bull calves were nonimplanted controls (CB). Nine bull calves were implanted with 140 mg trenbolone acetate (TBA) and 36 mg of zeranol (Z) at about 1 mo of age and reimplanted with both hormones 10 wk later. When the nine implanted bulls (IB) were about 21 wk of age, TBA implants were removed. The IB then were reimplanted with Z alone every 10 wk until slaughter. The remaining eight bull calves were castrated at about 3 wk of age and implanted (IS) with TBA and Z every 10 wk until slaughter. Five calves had to be removed from the study because of chronic bloat, and (or) blindness or a mistake in castration. Calves were weaned about 7 mo of age and fed a high concentrate diet for up to 210 d. Blood samples were collected biweekly starting at 5 wk of age and analyzed for estradiol  $17\beta$  ( $E_2$ ) and testosterone (T). Weaning weight, daily gain and feed efficiency were similar for all treatments, whereas hip heights at 12 mo and masculinity scores at 13 mo tended ( $P=.09$ ) to be lowest for IS. Scrotal circumferences were lower ( $P<.05$ ) for IB than for CB at both 8 and 13 mo. Slaughter weight was lowest ( $P=.07$ ) for CB. There were no differences in carcass traits due to treatment. CB had heavier ( $P<.05$ ) testicular weights and more ( $P=.07$ ) longissimus muscle detectable connective tissue as determined by a trained sensory panel. Implanting bulls with TBA and Z suppressed ( $P<.05$ ) concentrations of serum T from 7.5 to 11 mo. However, IB had higher ( $P<.05$ ) serum T levels from 12 to 13.6 mo than CB. Serum concentrations of  $E_2$  were similar among treatments. Implanting steers resulted in the optimum combination of performance, carcass and meat sensory traits.

Key words: Bulls, Steers, Anabolic Implants, Performance, Carcass, Endocrine.