Acta Veterinaria (Beograd), Vol. 50. No. 2-3, 137-146, 2000.

UDK 619:636.2.087.8

A TRENBOLONE ACETATE/ESTRADIOL COMBINATION IN FEEDLOT SIMMENTAL BULLS: MEAT QUALITY AND WITHDRAWAL TIME OF TRENBOLONE

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(Received 2. March 2000)

Anabolic implants are used to improve growth rate and feed efficiency of cattle in feedlots thus resulting in economic benefits. One of the growth promoters used in raising animals for slaughter is yhe androgen - trenbolone acetate (TBA). It is most frequently applied alone or in combination with an estrogen such as estradiol (E)

The aim of this work was to examine the combined effect of trenbolone acetate and estradiol on meat quality in young bulls of the Simmental breed. The residues of trenbolone in faeces and urine, and time of their excretion from the organism were determined also.

On the basis of the results obtained it can be concluded that the combined use of trenbolone acetate and estradiol in feedlot Simmental bulls increased the dressing percentage by 6,5% and decreased fat by 27,5% as well as total pigment.The withdrawal time of trenbolone was two months after its application.

Key words: Anabolics, implantation, meat quality, residues, withdrawal time

INTRODUCTION

The application of growth promoters in meat animals kept in feedlots began from the desire to achieve economic profits. Anabolics accelerate growth in such way that they improve feed efficiency. Namely, they increase the rate of feed conversion and improve the meat : fat ratio. However, in addition to a positive role in the stimulation of growth, the use of anabolics can have a deleterious effect on beef quality. This effect is especially reflected on marbling and tenderness of beef, although there are opposing findings (Trenkle, 1990; Foutz et al., 1990; Huck et al., 1991; Thonney et al., 1991; Apple et al., 1991; Perry et al., 1991; Moran et al., 1991; Bartle et al., 1992; Hayden et al., 1992; Herschler et al., 1995; Gerken et al., 1995; Preston et al., 1995; Johnson et al., 1996; Saičić and Spirić, 1997; Saičić et al., 1998).

Opinions on the use of anabolics as growth stimulators differ very much. In countries with large meat production (USA, Australia)from castrated cattle, the use

of some hormone preparations is permitted, whereas in countries such as members of the European Union, the use of anabolics is banned by Council Directive, 96/22/EC, issued on April 1996, except for therapeutic purposes.

One of the growth promoters used in raising animals for slaughter is the androgen - trenbolone acetate (TBA), whose active component is trenbolone (TBOH). Numerous examinations of the activity of trenbolone acetate, alone or in combination with other hormones, have been performed in steers.

Thus, Cecava and Hancock (1994) found that 17-estradiol, alone or in combination with trenbolone acetate, after the introduction of feed with a high protein content, in steers of the Angus breed increased nitrogen retention and growth.

Similar activity of trenbolone acetate and estradiol was noted in Holstein and Simmental steers.. Namely, average daily gain and feed efficiency were increased by anabolics, although no effects on the carcass conformation and dressing percentage were observed. Daily gains of protein and fat in treated steers were increased by 23% (Perry et al., 1991).

Herschler et al. (1995) examined the influence of trenbolone acetate alone, combinations of estradiol and trenbolone acetate in two different ratios (1:5 and 1:10) and estradiol alone, on feedlot steers and heifers. Animals treated with E:TBA (1:10) showed better growth and feed conversion in relation to the group treated with E:TBA (1:5) during a period of 145 days. The doses of 28 mg E and 200 mg TBA were close to the optimum for stimulation of growth and feed conversion in both steers and heifers. However, the authors emphasized that growth in the steers treated with TBA alone was far lower than in steers treated with combined anabolics.

Similar investigations in steers of the Brangus breed were performed by Gerken et al. (1995). After application of TBA alone, TBA + E and E alone, steers were fed with concentrates for 112 days and afterwards slaughtered. In comparison with the control group, treated steers had higher average daily growth and carcass mass. The authors concluded that treatment of the steers with anabolics did not have an essential influence on meat tenderness and intramuscular fat.

According to Johnson et al. (1996), the average daily growth was increased by 16% and feed utilization was improved by 13% in steers treated with TBA and E, 143 days after implantation. Moreover, carcasses of treated steers had a lower percentage of fatty tissue on the kidneys, pelvis and heart, increased body water and somewhat wider Longissimus dorsi muscles in relation to untreated steers.

Many investigations were performed with the aim to develop new methods or to improve current methods for determination of trenbolone residues (Jansen et al., 1989; De Boer, 1991;Daeseleire et al., 1991; Bagnati and Fanelli, 1991; Lagana and Marino, 1991: Daeseleire et al., 1992; Hewitt et al., 1993). A routine immunoassay (IA) "screening" method which is quick, cheap and sensitive is used

most frequently. Samples with positive findings are further examined by confirmative methods such as TLC (thin - layer chromatography), HPTLC (high performance thin layer chromatography), HPLC (high performance liquid chromatography), RIA (radioimmunoassay), GC/MS (gas chromatography/mass spectrometry) and the combination RIA/HPLC. The lowest detectable limit (LDL) of these methods is in the range of 0,02 - 5 μg/kg (μg/L). As a target sample for the examination of residues, muscle, organs (liver, kidney), fat, bile, urine, faeces, plasma and serum can be used (Commission of the European Communities, 1994).

Since the effect of anabolics, to a great extent depends on the species of animal, as well as on some other parameters like sex, age and diet, etc., we decided to examine the combined effect of trenbolone acetate and 17β-estradiol on meat quality in feedlot bulls of the Simmental breed, which is customary in these areas.

Determination of the trenbolone residues, as the main metabolite in the urine and faeces of five animals, as well as in muscle tissue at slaughter, was included in our investigation as a measure aimed at public health protection.

MATERIAL AND METHODS

The examinations were performed on a private cattle farm in Ečka. Ten Simmental bull calves, eight months old, were assigned randomly to two groups. The first five were used as the control group (no implant), and the other five were treated with Revalor S. Revalor S (24 mg of 17β-estradiol and 120 mg of trenbolone acetate) was surgically implanted under the skin, behind the ear. The bulls were weighed and placed in the pen. During fattening they were fed a grower diet, consisting of hay, corn silage and ground cereals (concentrate) containing 12% protein. Sixty days after implantation, since the analysis of faeces and urine showed that the trenbolone had been excreted, reimplantation was done. The samples of faeces and urine were always taken at the same time of day. Trenbolone was not detected in faeces and urine 120 days from the first implantation. Bulls of both groups were weighed and slaughtered. The carcass weights were recorded. Pelvis, heart and kidneys were examined in for fat content. Samples of Musculus longissimus dorsi, from the first to the third vertebra, were taken for the following analyses: pH (ISO, 1974), water, fat and protein (AOAC, 1995), collagen (ISO, 1994), water binding capacity - WBC (Grau and Hamm, 1957), weight loss by cooking (Honikel, 1987) and total pigments (Mohler, 1966). Sensory evaluation of meat tenderness was carried out on the 7th day in cooked samples (70°C) by six trained persons, using a scale from 1 (very tough) to 10 (very tender).

Muscle tissue of treated slaughtered animals was examined for trenbolone residues using the HPLC method developed by Laitem et al. (1978) and the Laboratory for residue analysis (RIVM - Bilthoven, Holland).The procedure was modified in our laboratory to achieve the necessary analytical requirements. The method involves enzymatic hydrolysis of the sample, to release the bound trenbolone. With different solvents, the trenbolone was extracted from the medium and after purification passed through an HPLC column. The experimentally established detection limit was 5 μg/kg. In order to determine the recovery percentage, blank and spiked samples were analysed in parallel with the samples from treated animals. The recovery was approximately 85%.

The results obtained were statistically evaluated and significant differences established by the Studentžs test.

RESULTS AND DISCUSSION

Table 1 shows the average daily gain (ADG) of the bulls and the dressing percentage. A (21%) higher ADG was observed in implanted bulls in relation to not implanted ones (control group). Treated bulls had higher weights and better dressing percentage (6,5%) and the differences were statistically significant ((P<0.05). These results are in accordance with the investigations of Preston et al. (1995), Herschler et al. (1995), Gerken et al. (1995), Johnson et al. (1996) and others.

Parameters of meat quality are presented in Table 2. Values for pH (24h post mortem) between the control group and treated group did not show significant differences. Also, the weight loss by cooking did not differ essentially between the mentioned groups (P>0,05). However, the percentage of released water (free water), as a criterion for water binding capacity, differed significantly (P<0.05), being higher in treated bulls. Implant treatment did not essentially influence meat tenderness and the results for collagen content confirmed these findings (P>0.05).

According to the results of Smulders et al. (1991), the use of hormones as growth promoters in animal raising, reduces the heme-iron content of muscles and may thus affect muscle colour. Our results confirm these data. The content of total pigments in M. longissimus dorsi of the treated bulls, differed significantly (P<0.05) from the control group.

Concerning basic chemical composition, differences in the contents of water and protein were not significant (P>0,05) between the two groups. However, a lower mean fat content (27,5%) was found in the treated animals. This difference is significant (P<0.05). Moreover examination of pelvis, heart and kidneys indicated grater fatness in control bulls.

The quantities of trenbolone residues found in faeces and urine are presented in Figures 1. and 2. The residues of anabolics and the elimination rate

Table 1. Average daily gain (kg) and dressing percentage

Investigated parameters	Control group		Treated group		Significant differences
	x	Sd	x	Sd	Student's test
Body weight before implantation, kg	392.2	7.71	386.0	13.32	NS
Body weight before slaughter, kg	488.8	8.37	503.0	8.39	*
Carcass weight,kg	292.8	7.93	321.0	12.25	*
Dressing percentage	59.88	0.74	63.80	1.15	*
Average daily gain, kg	0.81		0.98		NS

NS – $P > 0.05$; * – $P < 0.05$

Table 2. Charasteristic of beef

Investigated parameters	Control group		Treated group		Significant differences
	x	Sd	x	Sd	Student's test
pH (24 pm)	5.70	0.01	5.69	0.01	NS
Water binding capacity (% of free water)	29.65	0.61	31.18	0.51	*
Weight loss after cooking, %	24.99	0.08	25.47	0.49	NS
Mean scores for tenderness of cooked beef (1 – very tought; 10 – very tender)	7.67	0.08	7.56	0.08	NS
Collagen content, %	0.486	0.02	0.496	0.04	NS
Total pigment, mg/kg	145.12	3.35	138.46	2.70	*
Basic chemical composition					
Water, %	74.10	0.25	75.17	0.68	NS
Fat,%	3.05	0.29	2.21	0.61	*
Protein,%	21.52	0.67	21.72	21.72	NS

NS – P > 0.05; * – P < 0.05

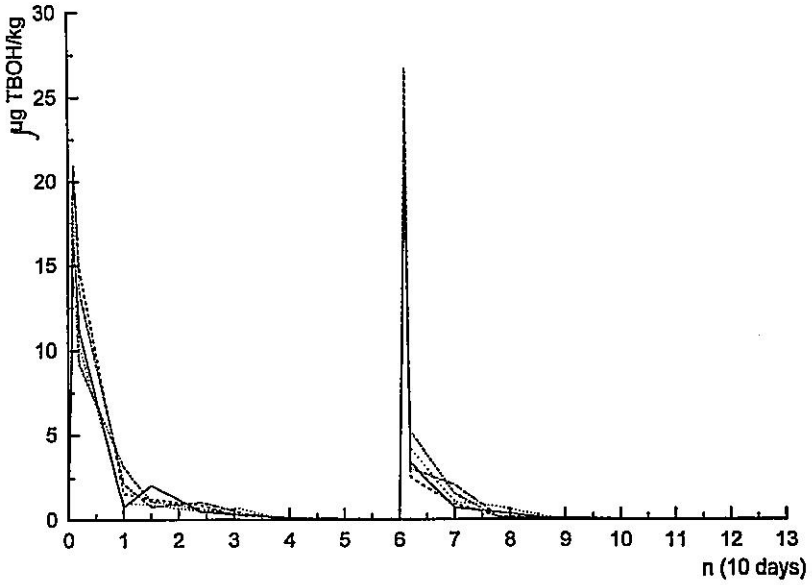

Figure 1. Residues of TBOH in the urine of 5 bulls implanted with 24 mg 17β-estradiol and 120 mg TBA

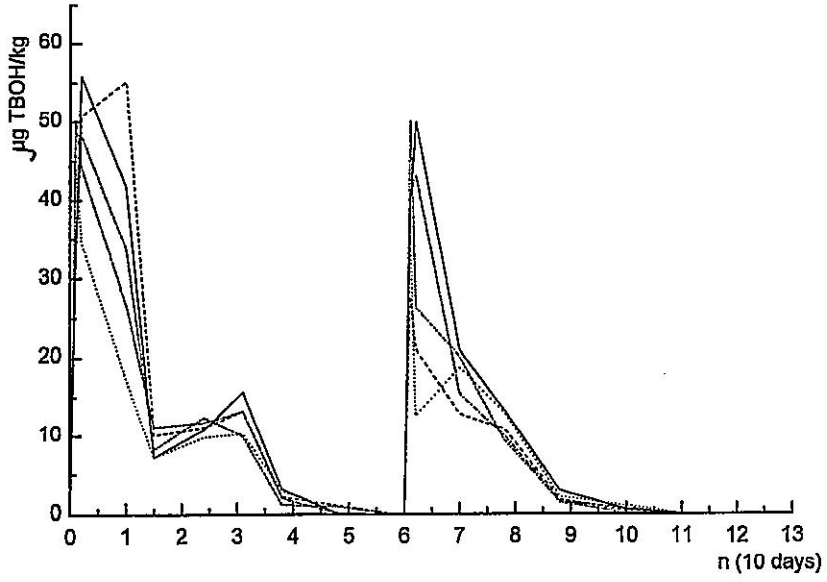

Figure 2. Residues of TBOH in the faeces of 5 bulls implanted with 24 mg 17β-estradiol and 120 mg TBA

from the body depend on the route of administration, the composition of the implant, animal type and the time expired after drug application (Karg et al., 1984).

It is evident there was a great difference of trenbolone concentration in urine and faeces. Namely, higher concentrations of trenbolone were observed in samples of faeces than in urine. Similar results were obtained by other investigators (Karg et al., 1984; Henricks, 1981). The maximum quantities of trenbolone residues in urine were found 24h after the application of Revalor S. In the faeces, maximum concentrations were detected 48h after pellet application. These data are in accordance with the findings of Karg et al. (1984). Moreover, a sudden decrease of trenbolone concentration in urine 15 days after implantation and in faeces 40 days after implantation was also demonstrated. As can be seen in Figures 1 and 2, complete excretion of trenbolone from the treated Simmental bulls occurred by two months after the application of Revalor S. Trenbolone residues were not detected in muscle tissue, which indicates that trenbolone excretion from the organism occurred prior to slaughter.

Acknowledgment

This work was supported by The Serbian Ministry of Science and Tecnology, Grant No 12M15.

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KOMBINOVANO DELOVANJE TRENBOLON ACETATA/ESTRADIOLA KOD BIKOVA U TOVU RASE SIMMENTAL : KVALITET MESA I VREME IZLUČIVANJA TRENBOLON ACETATA

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SADRŽAJ

Anabolici se koriste da poboljšaju rast i iskorišćenje hrane kod stoke u tovu, čime se postiže ekonomska dobit. Jedan od promotera rasta koji se koristi u uzgoju stoke za klanje je androgen - trenbolon acetat. On se najčešće primenjuje sam ili u kombinaciji sa estrogenom - estradiolom.

Cilj ovog rada je bio da se ispita zajedničko delovanje trenbolon acetata i estradiola na kvalitet mesa kod bikova simentalske rase u tovu. Ovim radom je obuhvaćeno i ispitivanje ostataka trenbolona u fecesu i urinu posle aplikacije hormona, kao i utvrđivanje vremena njegove ekskrecije iz organizma.

Na bazi dobijenih rezultata, može se zaključiti da je kombinovano delovanje trenbolon acetata i estradiola kod simentalskih bikova u tovu imalo uticaja na povećanje randmana za 6,5% i smanjenje masti za 27,5%. Do ekskrecije trenbolona došlo je dva meseca posle njegove primene.