1 Preface

Extracts of the thesis presented here, have been written to be submitted for publication in *The Veterinary Journal*.

Castration of lambs – Comparison of different castration techniques in lambs older than 10 weeks of age with regard to animal welfare

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3 Summary

Seventy male lambs older than 10 weeks of age were control handled or castrated by Burdizzo-, rubber ring- or surgical method to assess the acute and long-term effects of castration. For local anaesthesia, either lidocaine or bupivacaine was used.

Elevated levels in serum cortisol concentrations, in the proportion of abnormal postures and in the frequency of immediate behavioural responses to castration indicated that surgically castrated lambs were most distressed. The results of the lambs of Burdizzo- and rubber ring groups in many respects were similar to those of lambs of the control group. Due to a faster wound healing, Burdizzo castration seems to be preferable when compared to the rubber ring technique. Between 1.5 to 9 hours after castration, signs of pain and distress were at a lower level in lambs anaesthetised with bupivacaine compared to lambs treated with lidocaine. It is concluded that local anaesthesia with bupivacaine, followed by the Burdizzo method is the preferable technique for the castration of lambs older than 10 weeks of age.

Keywords: Lamb; castration; local anaesthetics; behaviour; cortisol

4 Zusammenfassung

Lämmer im Alter von über 10 Wochen wurden während und nach der Kastration beobachtet, um Kurz- und Langzeiteffekte von drei verschiedenen Kastrationsmethoden mit 2 verschiedenen Lokalanästhetika zu untersuchen.
Siebzig Lämmer wurden entweder mit der Burdizzo-Zange, dem Gummiring oder chirurgisch kastriert oder sie gehörten einer Kontrollgruppe an. Bei jedem Tier wurde 10 Minuten vor der Kastration entweder Lidocain oder Bupivacain in beide Samenstränge und subcutan in den Skrotumhals injziert.
Während den ersten 9 Stunden nach der Kastration wurde in vorgegebenen Intervallen die Serum Kortisol Konzentration gemessen. Verhalten, Körperhaltung und klinische Parameter wurden über einen Zeitraum von mindestens einem Monat oder bis zur Abheilung der Kastrationswunden beobachtet und notiert.
Die Unterschiede der Serum Kortisol Konzentrationen, Körperhaltung und Schmerzantwort auf Palpation zwischen der Gruppe Chirurgie und allen anderen Gruppen zeigten, dass chirurgisch kastrierte Lämmer mehr Schmerzen und Stress hatten. Die Resultate der Burdizzo- und Gummiringgruppen waren oft ähnlich verglichen zur Kontrollgruppe. Die Burdizzo-Kastration ist die Methode der Wahl, weil die Wundheilung im Vergleich zu der Gummiringgruppe viel schneller und ohne Komplikationen vonstatten ging. Wenn Bupivacain im Vergleich zu Lidocain als Lokalanästhetikum gebraucht wurde, waren die Anzeichen für Schmerz und Stress während den ersten 90 Minuten bis 9 Stunden nach der Kastration bei der Bupivacain-Anästhesie geringer. Daraus kann geschlossen werden, dass die „schonendste“ Kastrationsmethode bei Lämmern älter als 10 Wochen, die Lokalanästhesie mit Bupivacain gefolgt von der Kastration mit der Burdizzo-Zange ist.

5 Introduction

In extensive animal husbandry, male lambs reach puberty before slaughter. Consequently, castration of males in the first days or weeks after birth is often a standard management procedure to prevent discomposure in the flock due to sexual behaviour of young males, unwanted gestation and indiscriminate breeding (Archer, 2004; Baird and Wolfe, 1998; Capucille et al., 2002).

Surgical castration as well as bloodless methods such as rubber ring, classical Burdizzo, and combined rubber ring and Burdizzo method are used to accomplish routine castration of male lambs (Homeyr, 1987; Molony et al., 1993). The rubber ring causes ischemia, leading to tissue death and sloughing, whereas Burdizzo castration destroys a narrow segment of each spermatic cord, the appropriate nerves and blood vessels and parts of the scrotal tissue, leading to ischemia and testicular atrophy. The scrotum thereby remains intact (Dinnis et al., 1997a; Lester et al., 1996). The rationale of the combined method is that crushing the nerves of the spermatic cord may reduce afferent neural transmission from ischemic tissue situated distal to the crush and ring. If the clamp is applied proximal to the ring and across the entire width of the scrotum, this method is reported to result in less signs of acute pain than the rubber ring method alone (Dinnis et al., 1997b; Kent et al., 2001; Kent et al., 1993). Basically it is assumed, that in Switzerland the combined method with rubber ring and Burdizzo is not well established in practise, therefore, it will not be assessed in this study.

Castration-related responses indicating pain and distress in lambs include changes in behaviour and serum cortisol concentration, and the occurrence of abnormal postures. This has been well established by several authors investigating post castration-distress over the first few hours (Dinnis et al.,

1997a; Kent et al., 1993; Lester et al., 1996; Mellor and Murray, 1989; Molony et al., 2002; Molony et al., 1993; Thornton and Waterman-Pearson, 2002). In these studies, observations were made on lambs between a few days and 9 weeks of age, and most authors evaluated the first day after castration only. There are few reports of long-term effects of castration (Kent et al., 2000; Mellema et al., in revision; Molony, 1995; Thornton and Waterman-Pearson, 1999). With regard to animal welfare it is, however, important to collect information about long-term effects of castration on pain and distress, in order to be able to make recommendations for suitable castration methods of young male lambs.

Male lambs are preferably castrated in the first few days to weeks of their life, because animal handling is easier and postoperative complications are fewer (Baird and Wolfe, 1998). Sometimes, however, it is inevitable to castrate animals older than 10 weeks of age. Reasons for castrating at an older age include castration of animals of different age groups at the same time for economical reasons, and castration of slowly growing individuals that did not reach slaughter weight before occurrence of puberty. For these cases, as far as the authors' are aware of, no results of studies on the effect of different castration methods on indicators of pain and distress are currently available in the literature.

Local anaesthesia seems to be one of the most important precautions for reduction of pain. For this reason, in Switzerland since September 1, 2001, the castration of male ruminants is no longer allowed without anaesthesia. Many studies confirm that after local anaesthesia of testes, lambs show less behaviour of pain and distress (Dinnis et al., 1997a; Kent et al., 1998; Mellema et al., in revision; Molony et al., 1997; Sutherland et al., 1999; Thornton and Waterman-Pearson, 1999; Wood et al., 1991). In these studies, lidocaine is the most frequently used local anaesthetic. In some studies, however,

bupivacaine was shown to result in a better and longer-lasting analgesia in lambs (Graham et al., 1997; Miller, 2000; Molony et al., 1997). The effective period of lidocaine is 60-90 minutes, while bupivacaine is metabolized more slowly than lidocaine and has an effective period of 3-4 hours.

The current study was designed to evaluate the most "gentle" castration method when combined with an effective analgesia in lambs older than 10 weeks of age. The methods compared were elastic rubber ring, Burdizzo and surgical castration. All methods were performed with either lidocaine or bupivacaine for local anesthesia. In addition, a control group with lidocaine anesthesia and mock (castration) handling was included as a baseline.

6 Animals, materials and methods

6.1 Animals and Housing

Seventy male White Swiss Mountain lambs or lambs of crossings with this breed (at least 50% White Swiss Mountain blood) were investigated during and after castration, using the standard methods rubber ring, Burdizzo clamp or surgery. The lambs were treated at 10 to 24 weeks of age, with a bodyweight of 25 to 46 kg (mean 37 kg, SD +/- 4.5). They were housed indoor in straw bedded pens in groups of 30 to 40 lambs (Fig. 1). The day before castration (day -1), clinical examination was performed, and only lambs not exhibiting any sign of disease were included in the study.

Figure 1: Pen with about 30 lambs

Lambs were weighed, marked, and an indwelling catheter (Venflon™, 16GA, Becton Dickinson, Helsingborg, Sweden) was introduced into one of the jugular veins to reduce stress during blood sampling. The scrotal wool of lambs of the surgery group was removed with fine electric clippers. From day -1 until blood sampling was finished at 9 hours after castration, the lambs were kept pairwise in pens measuring 1 x 2 m. Afterwards, they were put back to their group mates.

6.2 Study design and treatments

The lambs were randomly allocated to one of seven treatment groups by a second person drawing lots from a "pot". The pot included one lot of each group. Each time a lot was drawn, it was removed from the pot and not replaced until seven lambs were assigned to the different treatment groups. Thus, these groups were completely randomized regarding age and body weight. The treatment groups included rubber ring castration with lidocaine (RR/li) or with bupivacaine (RR/bu), Burdizzo castration with lidocaine (B/li) or

bupivacaine (B/bu), surgical castration with lidocaine (S/li) or bupivacaine (S/bu), and control handling with lidocaine (H/li).

The syringes for local anaesthesia were prepared by a second person. The operator, therefore, was blinded as to the use of lidocaine or bupivacaine for local anaesthesia.

Before treatment, all animals received a tetanus prophylaxis of 1500 I.U. of Tetanus-Serum (Veterinaria AG, Zurich, Switzerland). In each case, the castrations or control handlings were performed between 7 and 8 o'clock in the morning.

Analgesic treatments

Lidocaine:

4 mg/kg lidocaine (2%) (Biokema AG, Crissier, Switzerland) was diluted with physiologic sodium chloride solution (NaCl) to a final volume of 15 mL. Local disinfection of the skin with diluted povidone iodine solution (Betadine, Provet AG, Lyssach, Switzerland) was performed before anaesthesia. For RR and B castration, the anaesthetic was injected into both spermatic cords and subcutaneously around the scrotal neck (Fig. 2). The scrotal neck was massaged for a few seconds to disperse the local anaesthetic. For S castration, 5 mL of the diluted lidocaine solution were injected subcutaneously in the distal third of the scrotum and 5 mL into each spermatic cord.

Figure 2: Injection of local anaesthetic

Bupivacaine:

1,5 mg/kg bupivacaine (0.5%) (G. Streuli & Co. AG, Uznach, Switzerland) was diluted with NaCl to a final volume of 15 mL. The methods of disinfection and injection were identical to those used with lidocaine.

The interval between the injection of the anaesthetic (li and bu) and the beginning of the castration procedure was between 10 and 15 minutes.

Castration methods

Control handling:

The operator manipulated the testes for 1 to 2 minutes, in order to simulate the handling associated with castration.

Rubber ring castration with lidocaine or bupivacaine:

A constricting rubber ring (Provet AG, Lyssach, Switzerland) was applied to the neck of the scrotum (Fig. 3), using an elastrator (Provet AG, Lyssach, Switzerland) after ensuring that both testes were situated distal and the teats proximal to the ring.

Figure 3: Rubber ring castration

Burdizzo castration with lidocaine or bupivacaine:

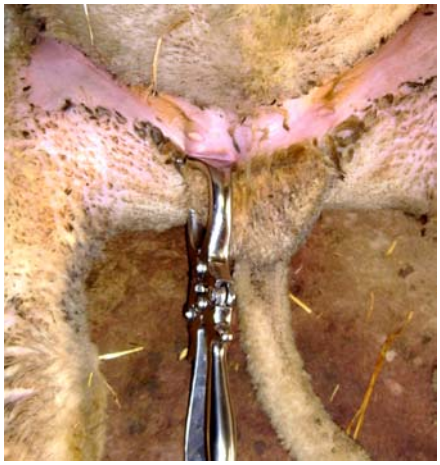

Figure 4: Burdizzo castration

Each spermatic cord and associated scrotal tissue was crushed twice at a distance of about 0,5 cm for 30 seconds each with a Burdizzo clamp (23 cm, Provet AG, Lyssach, Switzerland). The second application of the clamp lay distal to the first. Care was taken to ensure that the clamp crush lines did not overlap and the septum scroti remained intact (Fig. 4).

Surgical castration with lidocaine or bupivacaine:

Half an hour prior to castration, 10 mg/kg of oxytetrazykline (Oxysentin 100; Novartis AG, Basel, Switzerland) was administered intravenously. Twenty minutes before castration, 0.15 mg/kg xylazine (Rompun®, Provet AG, Lyssach,

Switzerland) was applied in the supraspinatus muscle. Ten minutes prior to castration, the local anaesthesia was performed as described above. For castration, the animal was placed with his hindquarters on clean cloths, and the scrotum was cleaned alternately with povidone iodine liquid soap (Betadine liquid soap, Provet AG, Lyssach, Switzerland) and alcohol (3 times for 1 minute each cycle). With a sterile pair of scissors, the distal third of the scrotum was resected. Both testicles were then exposed, preserving the common vaginal tunic (Fig. 5). Using a mash emasculator (Provet, Lyssach, Switzerland), each spermatic cord was crushed above the testicles. While the emasculator remained in place for 2 minutes, the cord was ligated (chromcatgut 7, Provet, Lyssach, Switzerland) proximal to the emasculator. The testis was resected distal to the crushing line.

Figure 5: Crushing the spermatic cord

The sedation of the lamb was antagonised with 2 mg/kg of the alpha$_2$-adrenergic antagonist tolazolinum hydrochloricum (Benzazolin®, Streuli AG, Uznach, Switzerland) administered intravenously.

6.3 Time need for castration

The time for castration or manipulation was measured, including the time for fixation and disinfection.

6.4 Blood sampling and serum cortisol assays

Blood samples (9 mL) were collected in a blood sampling system for serum preparation (Monovette®, Sarstedt, Nümbrecht, Germany). They were taken from each lamb prior to local anaesthetic administration or injection of oxysentin in the surgery group, immediately after anaesthesia, and at 20 and 40 minutes and 1, 1.5, 2, 2.5, 3, 3.5, 4, 6 and 9 hours after castration or control

handling. The blood samples were centrifuged at 2376 g for 10 minutes within 1 hour after blood sampling. The serum was separated and stored at –20°C until cortisol analysis. The cortisol concentrations were determined using a competitive immunoassay (Immulite®, Labor Laupeneck, Bern, Switzerland). The area under the curve (AUC) was calculated for cortisol values between -20 minutes to 9 hours.

6.5 Recording of behavioural responses

The immediate behavioural response of each lamb throughout the duration of the castration (or control handling) was assessed and categorized as no response (code = 0), moderate response (= 1) and severe response (= 2). Rare bleating, kicking with hind legs and cramping was judged as moderate response. Severe response was noted when the lamb was bleating or struggling with the whole body throughout the entire procedure.

Observations were made from outside the pen without disturbing the lambs. The observation periods lasted for 10 minutes each. A first recording was done on day –1 in the morning to describe the individual baseline value. On the day of castration (day 0), observations were made prior to each blood sampling. On days 1, 2, 3, 4, 5, 6, 9, 12, 15, 18, 21, 24, 27, and 30, one recording each was done in the morning. If the castration wounds had not healed at one month after castration (remaining signs of infection, swelling or discharge), the lamb was additionally observed once within the time periods (days) 36-38, 43-45, 50-52 and 58-60.

During the 10-minute observation periods, a recording of the lambs posture was performed every 2 minutes if no change in posture occurred, otherwise the change was noted. The food intake was noted in an analogous manner. All occurrences of the other behavioural parameters were recorded.

Behaviour and postures were recorded according to Molony et al. (2002) as defined in Table 1.

Table 1: Description of behaviours, postures and other observations.

Behaviour:	
Foot stamping/ kicking	A limb is lifted and forcefully placed on the ground
Easing quarters	A limb is moved in a less forceful manner than with stamping
Standing up / lying down	A lamb stands up and lays down, each unit scored included both the act of rising and lying down
Head turning	Movement of the head beyond the shoulder
Vocalisation	Vocal sound
Postures:	
Normal lying	Ventral recumbence, the lamb laying on its sternum and abdomen with all four legs tucked in
Abnormal ventral lying	The lamb laying on its sternum with the hind legs extended or ventral recumbence with keeping the scrotal region off the ground (dog sitting)
Abnormal lateral lying	The lamb lying on its side with one ore both forelegs and both hind legs stretched out laterally
Normal standing/ walking	Standing and walking with no apparent abnormalities
Abnormal standing / walking	Standing or walking unsteadily, swaying, arched back, hind limbs apart and positioned further back than normal
Statue standing	Standing still for more then 10 s
Tail stretching	Holding tail in unusual stretched posture, not counted while defecation and urination
Others:	
Eating	Eating or rumination

The combined index "total behaviour" was calculated as the sum of foot stamping/kicking, easing quarters, standing up/ laying down, head turning and vocalisation. The "proportion of abnormal postures" was calculated as the number of recorded abnormal postures (abnormal lying, abnormal standing/ walking, statue standing, tail stretching) relative to the total recorded postures.

6.6 Assessment of the scrotal region and bodyweight measurement

The local effects of the different castration methods were assessed by palpation and visual assessment of the scrotal region. Pain was recorded by response to palpation of the scrotal neck and testis. The response to local palpation was categorized as no reaction (code = 0), wincing (= 1), and struggling with attempts to escape (= 2).

The maximum scrotal circumference was measured and swelling, signs of inflammation, exudation, state of necrosis, and drop off of the testis were noted. The scrotal condition was rated as follows: intact scrotum, testes disintegrating, scrotum drying, beginning to fall off, scrotum dry and hard, scrotum absent with eschar, lesion healed, atrophic testes, moderate swelling of the scrotum, severe swelling/ inflammation, or secretion.

On the day of castration, the lambs were captured for a short time for visual inspection and for palpation after the behavioural observation periods at 40 minutes and at 1, 2, 3, 4, 6 and 9 hours after castration. Afterwards, this was done after each observation period. In surgically castrated lambs, the response to local palpation and scrotal condition was noted for the first time at 9 hours after castration to reduce additional contamination of the wound immediately after surgery.

The bodyweight was measured on days 2, 6, 12, 21, 30 after castration and at the day of slaughter.

6.7 Slaughter and histological examination of the scrotal tissue

Lambs were slaughtered at an average bodyweight (± SD) of 45.3 ± 1.0 kg. Testes from lambs of the Burdizzo and control groups were collected after culling and preserved in formalin (4%). The samples were embedded in paraffin wax, and slides were stained with haematoxylin and eosin for histological analysis.

6.8 Statistical Analysis

Data were stored in Microsoft Excel spreadsheets (www.microsoft.com) and analysed using the commercial statistic program NCSS® (2001, Kaysville, Utah, USA, www.ncss.com). Pre-treatment values of cortisol concentration, bodyweight and scrotal circumferences were subtracted from the subsequent sample values to correct for individual animal variation in the baseline levels. The immediate pain response to castration (0,1,2) was compared among groups by chi-square tests. The Kruskal-Wallis ANOVA on Ranks with Bonferroni Correction for Multiple Comparisons was used to assess the association between the treatment groups and the following factors: time needed for castration, cortisol response, AUC of the cortisol response over defined time intervals, proportion of abnormal postures, total behaviour, frequency of eating, daily weight gain, scrotal circumference and the time until healing. The response to local palpation was compared by Poisson regression with the frequency counts as the outcome and treatment group (baseline = control group) as independent variables. To assess the differences between lidocaine and bupivacaine, separate analyses were run for each castration method. The overall level for statistical significance was set at $p = 0.05$.

7 Results

7.1 Time needed for castration

The time needed for castration of lambs of the S groups (13.9 ± 2.34 min) was significantly longer ($p<0.01$) than that of all other groups (Figure 6). The castration of lambs of the B groups (5.4 min ± 0.82) lasted significantly longer ($p<0.01$) than the castration of lambs of the RR groups (0.7 min ± 0.26).

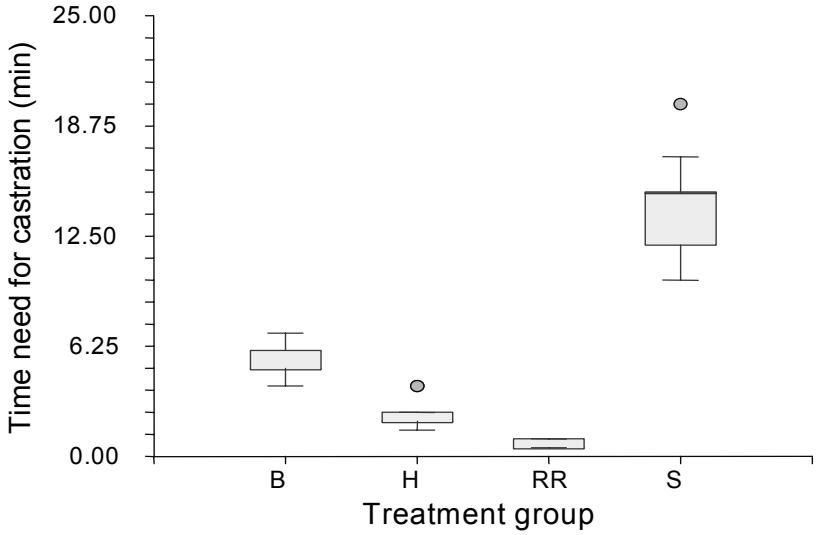

Figure 6: Box-plot presentation of time needed for castration of lambs of the control- (H), Burdizzo- (B), rubber ring- (RR) and surgery- (S) groups.

7.2 Immediate expression of pain during castration

Expression of pain during castration was significantly influenced by the castration method. Lambs of the S and B groups showed significantly more pain response during castration ($p<0.01$) than lambs of the RR and H groups (Table 2). The type of anaesthetic drug did not have any effect on the pain response during castration.

Table 2: Distribution of experimental lambs over the categories used to score the immediate behavioural response to castration. Percentage values for each castration method are given in parenthesis.
B = Burdizzo, H = control handling, RR = rubber ring, S = surgical castration

	Castration method			
Response category	**B**	**H**	**RR**	**S**
No response (0)	2 (10)	8 (80)	15 (75)	2 (10)
Moderate response (1)	13 (65)	2 (20)	3 (15)	15 (75)
Severe response (2)	5 (25)	0 (0)	2 (10)	3 (15)
Total	20 (100)	10 (100)	20 (100)	20 (100)

7.3 Cortisol

Cortisol concentrations over the first 9 hours after castration of all 7 treatment groups are depicted in Figure 7. Lambs of treatment groups S showed significantly ($p<0.01$) higher cortisol levels throughout the day of castration as compared to all other treatment groups. Nine hours after castration (last blood sampling), the baseline value was not yet reached. Among the treatment groups B, RR and H, significant differences of cortisol concentrations were not evident.

Within lambs castrated with the Burdizzo method, cortisol concentrations after li anaesthesia were significantly higher ($p<0.05$) at 1.5 and 2.5 hours and tended to be higher ($p=0.058$) at 3 hours after castration than after bu anaesthesia. At 6 hours after castration, cortisol concentrations of B lambs reached baseline values. With regard to RR lambs, the cortisol concentrations of RR/li lambs tended to be higher ($p=0.075$) than those of RR/bu lambs at 3 hours after castration, and the cortisol concentrations were significantly higher ($p=0.012$) in RR/li lambs after 4 hours. Within the S treatment, no

significant differences in cortisol concentrations were found between lambs of groups S/li and S/bu.

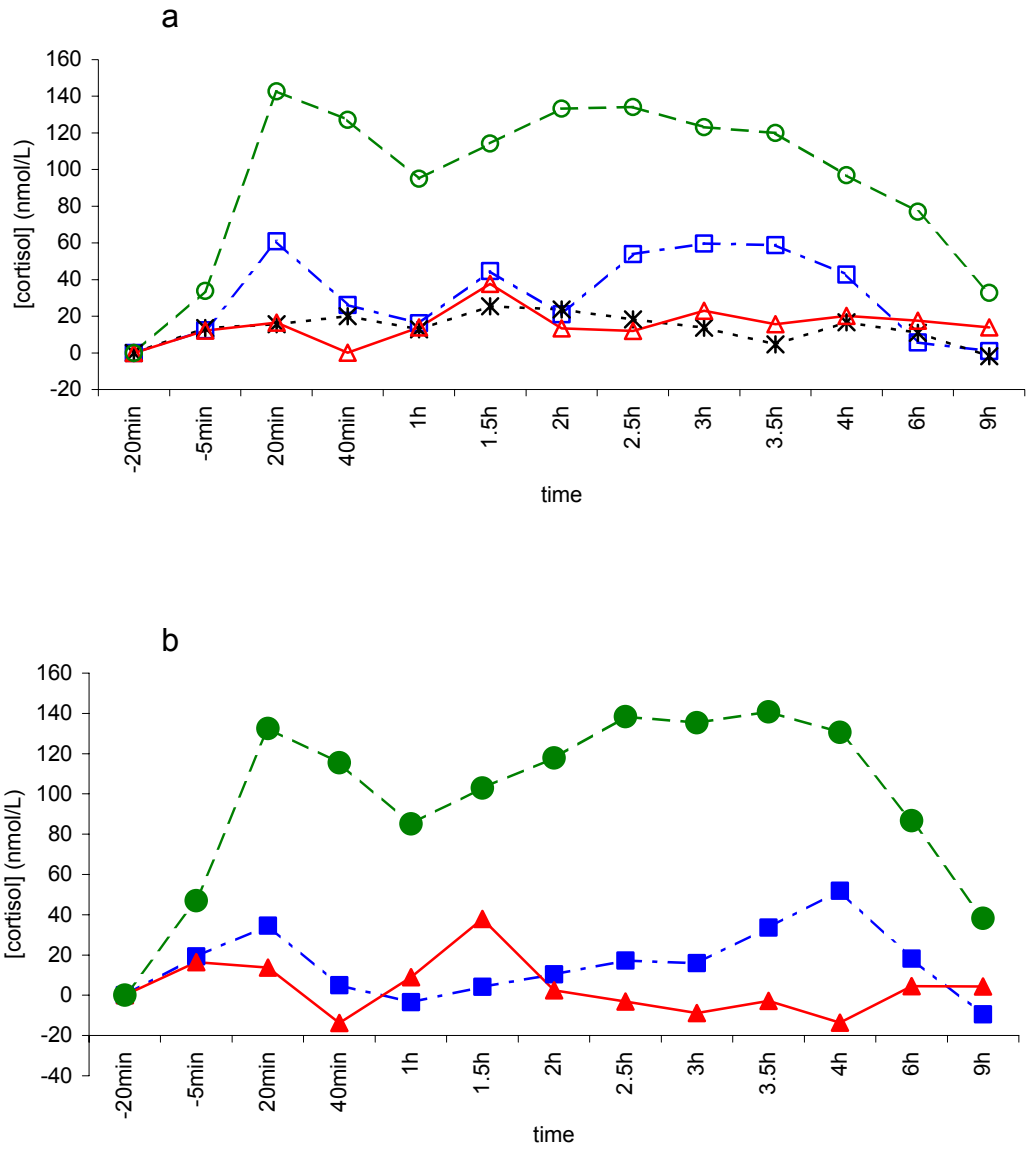

Figure 7:

Mean plasma cortisol concentrations from 20 min before until 9 hours after treatment, in response to three different castration methods and control handling anaesthetised with bupivacaine (a) and lidocaine (b). min = minutes, h = hour(s). Burdizzo castration with lidocaine □, rubber ring castration with lidocaine △, surgical castration with lidocaine ○, control handling with lidocaine ∗, Burdizzo castration with bupivacaine ■, rubber ring castration with bupivacaine ▲, surgical castration with bupivacaine ●.

7.4 Food intake

During the day of castration, lambs of the S groups were significantly ($p<0.01$) less frequently observed at eating or ruminating than lambs of all other treatment groups (Figure 8), and during the days 1 to 6 after castration there was still a trend ($p=0.069$) towards the same finding. The lambs of the B groups spent significantly ($p<0.01$) less time eating between 2.5 to 9 hours after castration than lambs of the RR and H groups.

Within the B groups, lambs with li anaesthesia spent significantly ($p=0.020$) less time eating than B/bu lambs during the first two hours after castration. But both groups didn't show any difference towards the H group. During the period 2.5 to 9 hours ($p=0.014$) after castration, within the RR groups, the li lambs spent significantly less time eating than bu lambs. Significant differences to the H group were not evident. Within the S groups, the time spent eating was significantly ($p=0.045$) less during the first two hours after castration when li as compared to bu was used.

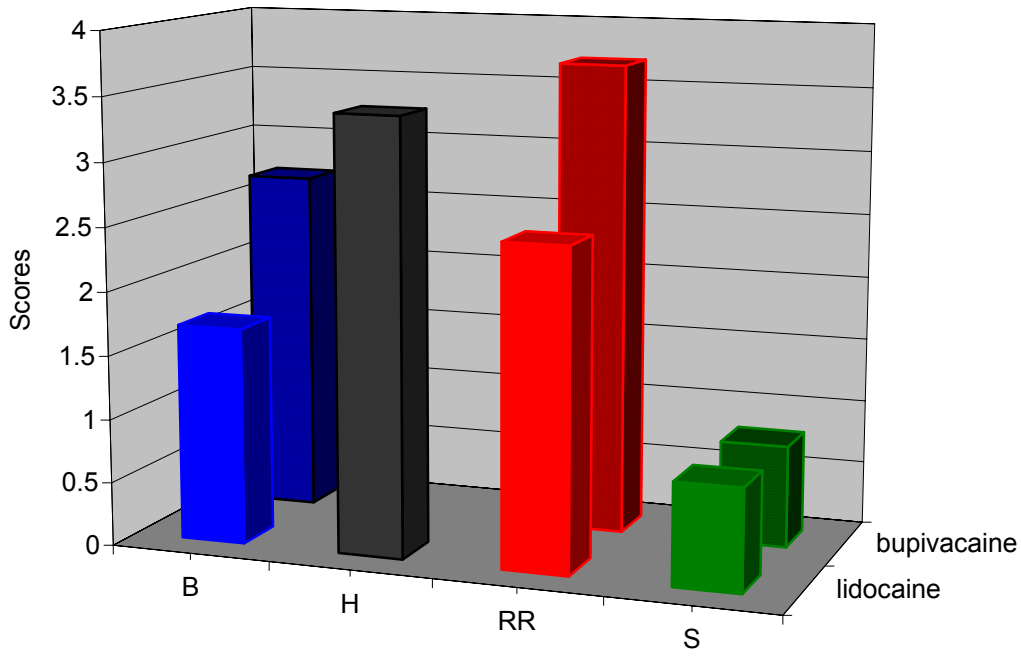

Figure 8: Average numbers of food intake recordings (scan samples) on the day of castration for lambs of the groups control (H), Burdizzo (B), rubber ring (RR) and surgery (S).

7.5 Proportions of abnormal postures

During the day of castration, abnormal postures were significantly ($p<0.01$) more often recorded in S lambs than in lambs of any other castration group (Table 3). Within the RR groups, the li lambs exhibited more often abnormal postures than the bu lambs: a trend ($p=0.079$) was found towards difference during the time period 20 minutes to 2 hours after castration, and significant differences ($p=0.028$) were found during the time period 2.5 to 9 hours after castration. Within the B and S groups, significant differences in abnormal postures between li and bu anaesthesia were not found. Examples of abnormal postures are presented in Figures 9, 10, 11 and 12.

Table 3: Group average of proportion of abnormal postures (in percent) during the respective observation periods after castration of lambs with the Burdizzo (B) method, the rubber ring (RR) method, the surgical (S) method, or after handling of a control group (H) anaesthetised with either bupivacaine (bu) or lidocaine (li).
min = minutes, h = hours, d= days

	Time after castration			
Castration method	**20 min - 2h**	**2.5 - 9 h**	**d 1 - 6**	**d 9 - 30**
B/bu	8.71	24.4	15.62	7.73
B/li	23.9	22.24	8.04	10.39
H/li	6.19	6.33	12.68	6.95
RR/bu	4.9	14.98	19.99	8.17
RR/li	20.21	31.47	29.72	12.35
S/bu	37.17	59.63	28.19	10.37
S/li	34.41	49.31	25.89	10.52

Figure 9: hind limbs apart and positioned further back than normal

Figure 10: arched back, tail stretched

Figure 11: hind legs extended

Figure 12: tail streched

7.6 Total behaviour

During the period 2.5 to 9 hours after castration, lambs of the B groups showed significantly ($p<0.01$) less active behaviour than lambs of the RR and H groups (Figure 13).

On the day of castration ($p=0.049$) and during the period day 1 to day 3 ($p=0.028$) after castration, within the B group, B/li lambs showed significantly

less active behaviour than B/bu lambs. Within the RR group, the RR/li lambs showed less active behaviour than the RR/bu lambs on the day of castration (p=0.034) and during the period day 1 to day 3 (p=0.041) after castration. But compared to the control handled lambs (H/li), the RR groups showed no significant difference. There was no significant difference between lidocaine and bupivacaine within the S groups.

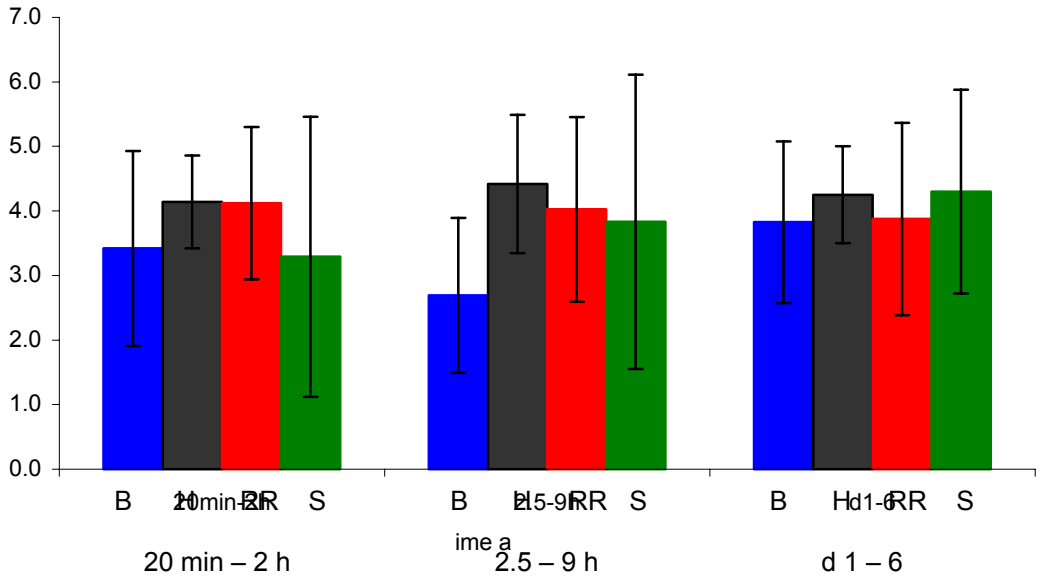

Figure 13:
Average scores of total active behaviour during the respective observation periods in relation to castration method.
B = Burdizzo, H = control handling, RR = rubber ring, S = surgical castration. min = minutes, h = hours, d = days

7.7 Scrotal condition and swelling

On the day of castration, the B groups showed a slight swelling of the scrotal neck and testes, which reached a maximum on day 1 (Figure 14). On day 7 after castration, the swelling was no longer obvious. During days 0 to 3, the scrotal circumference was significantly (p<0.01) increased in lambs of the B

groups as compared to the lambs of the H group. After castration, the lambs of B groups showed a small scab at the crushing area for 3 to 4 days.

After rubber ring castration, the testes dropped off after a mean (± SD) of 35 ± 6.9 days. Six out of 20 RR lambs showed purulent secretion at that time.

All S lambs developed moderate swelling of the scrotal neck. Seven out of 20 lambs showed severe swelling. On the day of castration, 17 out of 20 S lambs showed sanguineous or serous secretion. Four to 5 days after castration, 8 S animals showed even purulent secretion over a period of 4 to 6 days. One S lamb developed an afferent infection of the spermatic cord and 3 abscesses.

Complete wound healing took 8.4 (± 5.3) days for the B lambs. Wound healing took significantly longer for the RR lambs with 40.3 (± 5.4) days ($p<0.01$) and for the S lambs with 30.7 (± 4.8) days ($p<0.01$) as compared to the B lambs. Figures 15, 16, 17 and 18 represent different scrotal conditions.

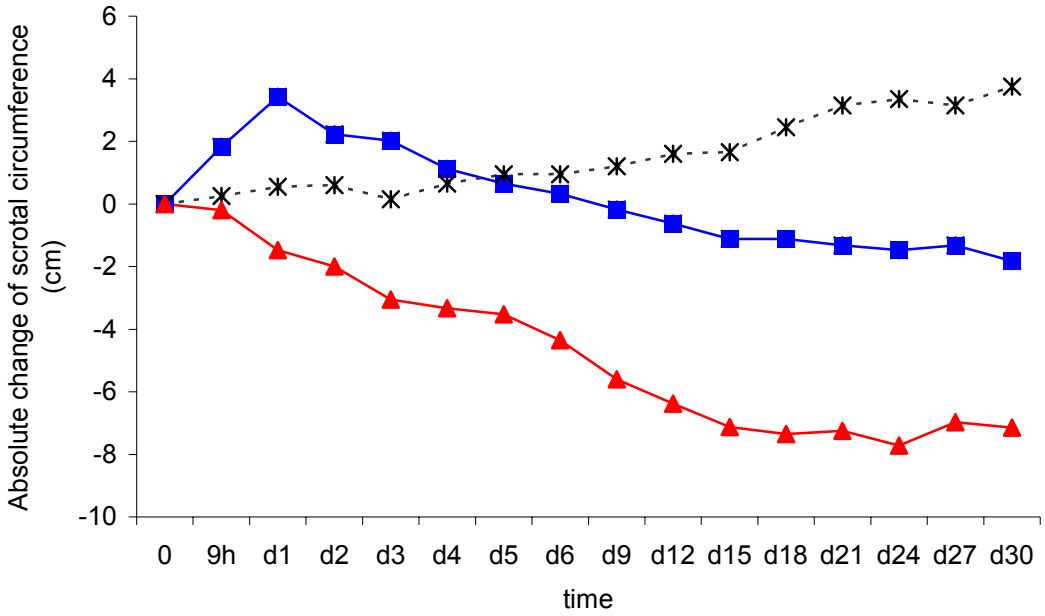

Figure 14:
Change of scrotal circumference in centimetres for the days 0 to 30 after treatment.
h = hours; d = days. Burdizzo castration ■, rubber ring castration ▲, control handling ∗.

Figure 15: day 44 after control handling

Figure 16: day 0, Burdizzo castration

Figure 17: day 21 after rubber ring castration

Figure 18: day 3 after surgical castration

7.8 Response to local palpation

During the first two hours after castration, significant differences among groups were not evident (Figure 19). During the time period 3 to 6 hours after castration, B lambs showed a significantly ($p<0.01$) stronger painful response to palpation than RR and H lambs, and lambs of the RR group showed a significantly ($p=0.019$) stronger response to local palpation than H lambs. At 9 hours after treatment, lambs of the S group showed a significantly ($p=0.028$) stronger response to local palpation than lambs of all other treatment groups. Between days 1 and 30 after castration, lambs of the RR group showed significantly ($p<0.01$) stronger responses to palpation than lambs of all other groups. Lambs of the S group showed during days 1 to 6 after castration

significantly ($p<0.01$) stronger painful responses to local palpation than lambs of the B and H groups.

Painful responses to local palpation were evident during a mean (± SD) of 1.3 ± 1.0 days in the B group, 12.3 ± 11.5 days in the RR group, and 4.9 ± 4.3 days in the S group.

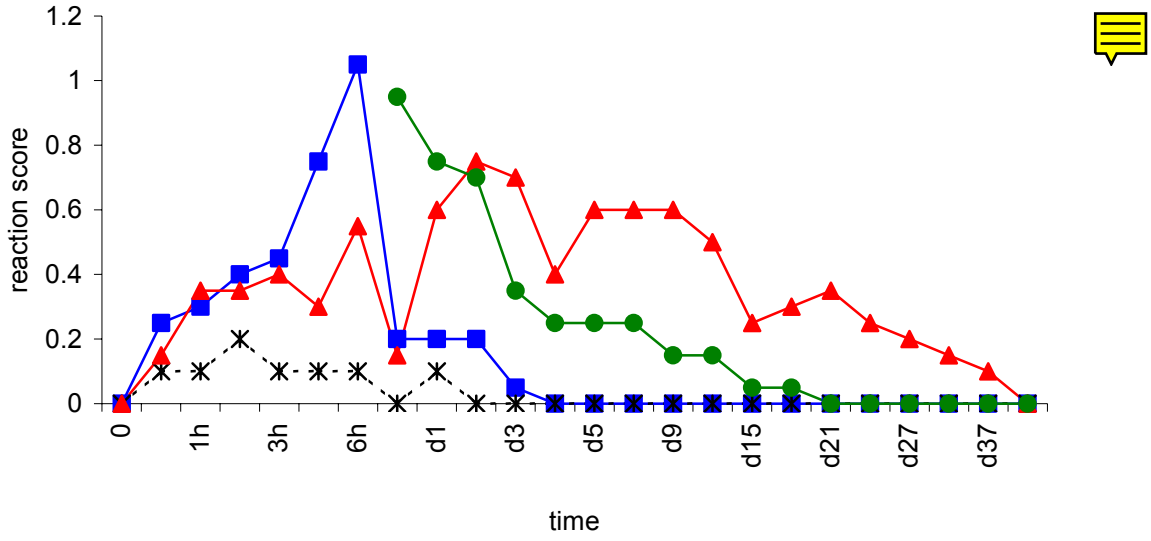

Figure 19:

Average pain scores to local palpation from 20 minutes before until 44 days after treatment. h = hours; d = days. Burdizzo castration ■, rubber ring castration ▲, surgical castration ●, control handling ✳.

7.9 Bodyweight

On the day before castration, there were no significant differences of bodyweight among the treatment groups (Figure 20). Over the first 3 weeks after treatment, the daily weight gain was significantly affected by the castration method. During the first 2 days after castration, S lambs lost a mean (± SD) of 2.5 ± 2.1 kg of bodyweight; whereas lambs of the other treatment groups kept their bodyweight or even gained some weight. Thus, the weight

gain was significantly smaller in lambs of the S group as compared to the lambs of all other treatment groups at day 2 ($p<0.01$) and to the lambs of H and B group at day 6 ($p=0.013$) and day 12 ($p=0.043$) after castration.

At day 21 and afterwards, the daily weight gain was no longer different among groups.

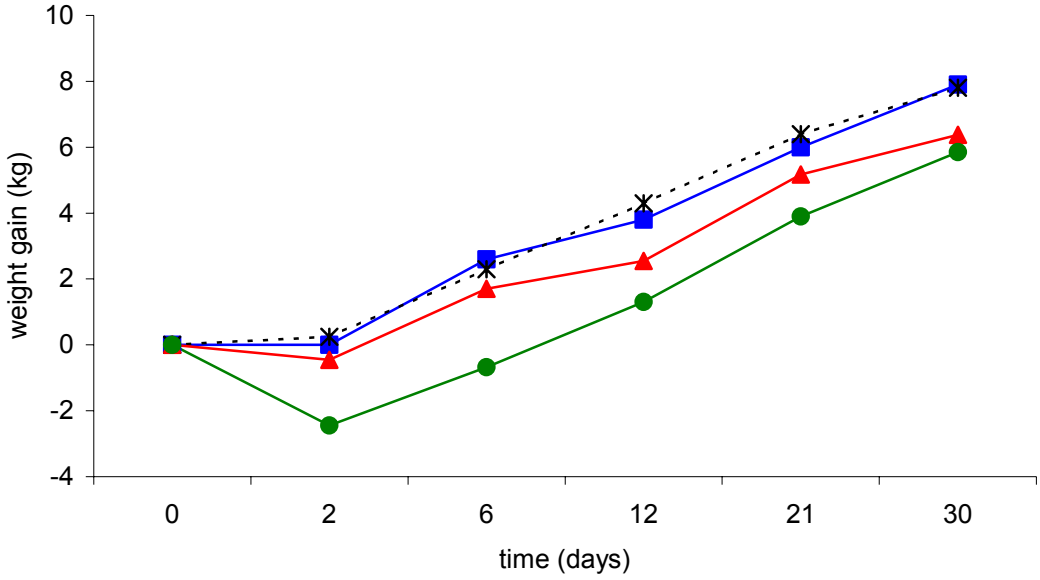

Figure 20:

Mean weight gain for the days 0 to 30 after treatment. Burdizzo castration ■, rubber ring castration ▲, surgical castration ●, control handling ∗.

7.10 Castration success

In all B lambs, the volume of scrotum and testes decreased compared to H lambs (Figure 21). After rubber ring castration, all lambs lost their scrotum and testes.

Figure 21: collected testes after culling. Left side: H lamb, right side: B lamb

7.11 Histology

The cells in the sex cords of B lambs were disintegrating, most cell walls existed no longer. No normal cell nuclei were apparent. In one B lamb, the spermatogenesis had begun before castration. On the slide of this lamb, few elongated spermatids were visible, but there were no free sperms and spermatogenesis had stopped. In none of the slides of the other B lambs, elongated spermatids or sperms could be found. The connective tissue between the sex cords was broadened by fibrocytes and macrophages, partly containing haemosiderin.

In 8 animals of the H treatment group, spermatogenesis and sperms were visible as an indicator of fertility. In the slides of the remaining 2 control lambs, large quantities of sex cords with supporting cells and spermatogonia were present.

8 Discussion

The results of this study may be summarized as follows: surgically castrated lambs showed significantly more often immediate behavioural pain response to castration than the lambs of all other treatment groups. The immediate pain response to Burdizzo castration was stronger than that to rubber ring castration. On the other hand, wound healing in lambs of the Burdizzo treatment group was faster and without complications as compared to lambs of the rubber ring treatment group. During the first 9 hours after castration, signs of pain and distress were lower when bupivacaine was used for local anaesthesia as compared to the use of lidocaine.

In the present study, the differences among the castration methods in behaviour, incidence of abnormal postures and cortisol levels were not as distinct as described in other studies, because lambs of all groups received local anaesthesia. That local anaesthesia is useful in either castration method is supported by the findings of Dinnis et al. (1997a), Kent et al. (1998), Mellor and Stafford (1999), Sutherland et al. (1999), Thornton and Waterman-Pearson (1999), and Mellema et al. (in revision). In our study, the anaesthetic effect was less pronounced in surgically castrated lambs as compared to the other treatment groups. This is in contrast to the study of Thornton and Waterman-Pearson (1999), who came to the conclusion, that after local anaesthesia none of the castration methods (rubber ring, combined ring and Burdizzo clamp, surgery) produced changes in cortisol concentrations that were significantly different from those seen in control lambs.

The cortisol concentrations after castration of lambs in the surgery treatment groups were significantly higher then those of lambs in the other treatment groups and did not return to pre-treatment levels until the last blood collection at 9 hours after castration. This is in accordance with the results of Lester et al. (1991), who, therefore, advised against the use of the surgical

castration. An other disadvantage of the surgical castration was reduced food intake resulting in weight loss during the first days after castration. Partly, this might be due to the sedation of the lambs in this treatment group. Although sedation was antagonised postoperatively, lambs were not completely awake for several hours and did, therefore, not eat as often as the lambs of the other treatment groups. Healing of the open wounds after surgical castration caused problems such as purulent secretion, prolonged pain, infection of the spermatic cord, and formation of abscesses. Furthermore, the surgical castration procedure used in this study was time-consuming. As a consequence of these results, the described surgical method can not be recommended for castration of lambs older than 10 weeks of age.

There are other surgical castration techniques described in the literature including drawing the testicles out with serrated tongs (Shutt et al., 1988), clamping the spermatic cord with artery forceps before being severed and cauterised with an electrically heated cautery distal to the forceps (Molony et al., 1993) or applying traction to the spermatic cord with manual rupture (Baird and Wolfe, 1998). These castration techniques are probably faster, but unlikely to evoke less pain.

The lambs castrated with the Burdizzo clamp showed an immediate behavioural response, indicating pain throughout the duration of the castration. However, during the first few hours after treatment, they showed less active behaviour than control- or rubber ring lambs. This is in agreement with the results of previous studies of Kent et al. (1995). Wound healing was completed within one week and complications did not occur. Burdizzo castration took a few minutes longer than the rubber ring method, and proper technique demands for an increased level of training as compared to application of a rubber ring. The difference of the size between Burdizzo castrated and intact testes was rather small. Therefore, success of castration

was difficult to be confirmed in the living animal. Histological examination, however, confirmed successful castration of all Burdizzo castrated lambs.

Rubber ring castration was the easiest and fastest of the compared methods. The response of the lambs to local palpation after castration was not as strong as the response of lambs in the surgery or Burdizzo treatment groups, but it persisted for a notedly longer period. Some of the lambs showed a painful response to palpation of the scrotal neck for more than 3 weeks. The difference in the response to palpation between lambs of the rubber ring and the Burdizzo treatment groups confirms previous findings of Molony (1995), reporting that long-term pain after rubber ring castration of calves can last for at least 42 days. In addition, the prolonged wound healing compared to Burdizzo castration and the occurrence of local infections do not favour the rubber ring method.

Both local anaesthetics, lidocaine and bupivacaine, did work well. Molony et al. (1997) applied bupivacaine for local anaesthesia because of the longer lasting effect, and he showed that bupivacaine is effective in reducing restlessness, time spent in abnormal postures and cortisol response in lambs after castration. These authors did, however, not perform a direct comparison between bupivacaine and lidocaine. In the present study, some differences were evident between the efficacy of lidocaine and bupivacaine on the day of castration in lambs of the Burdizzo and rubber ring treatment groups. On the first day after castration, the level of indicators of pain was significantly reduced when bupivacaine was used. This was to be expected due to the longer acting time of bupivacaine. For the surgical castration, it was not important which local anaesthetic was used. There were no differences concerning behaviour or postures between the two anaesthesia groups.

Independent on the method, castration always has to be performed with local anaesthesia. The actual law in Switzerland doesn't accept, that livestock owners castrate their animals themselves. But in January 2006, the new animal drug by-law comes into effect. This by-law allows, that after

sufficient training, livestock owners are allowed to perform local anaesthesia and castration of young ruminants themselves. The appeal of the animal protection law is currently in consultation in the parliament.

To simplify matters, livestock owner should castrate their lambs as early in life as possible, because of the better wound healing and the easier animal handling (Baird and Wolfe, 1998). If the ideal time in point was missed or a livestock owner prefers the castration of a group of lambs of different age categories at once by a veterinarian, we recommend the Burdizzo technique, because of the lower complication rate and the lack of long-term pain as compared to the rubber ring.

8.1 Conclusion

Lambs older than 10 weeks of age should not be castrated surgically. Rubber rings are fast and easy to apply, but this castration method may lead to local infection and long-term local pain. Acute behaviour pain response during Burdizzo castration was not completely abolished by either of the anaesthetics used in this study, but the raise in cortisol concentration after castration did not significantly differ from that observed in lambs of the mock-handled control treatment. Signs of local pain and wound healing of Burdizzo castrated lambs lasted for a few days only. Based on the results of our study, Burdizzo castration, combined with local anaesthesia using bupivacaine or lidocaine, can be recommended for the castration of lambs older than 10 weeks of age.

9 References

10 Acknowledgements

This study was granted by the Swiss Federal Veterinary Office (No. 2.03.01).

I would like to thank all people who helped me to bring this work to a good end. Special thank go to:

Adrian Steiner for being my supervisor and his fast and good corrections. Thank you for giving me the opportunity to work an such an interesting project.

Peppino Beffa and his family for technical support and the enjoyable accompaniment during the time I spent with them on the Bogigenhof.

Markus Doherr for his statistical support and corrections of the manuscript.

Beat Wechsler for his constructive inputs and very helpful comments on the manuscript.

Michael Stoffel for his assistance with the interpretation of the slides.

Mr. Winzap and his team for sterilising the surgical instruments and the cloths for free.

Sibylle Mellema-Aeschimann for all her help. Thank you very much for the interesting discussions, your good hints and your accompaniment through my whole work.