doi: http://dx.doi.org/10.5016/1806-8774.1999v1p87

ARBS

J P KASTELIC
Research Centre, Agriculture
and Agri-Food Canada,
Lethbridge, Alberta, CANADA

Scrotal/Testicular Thermoregulation in the Bull

Contents

Correspondence

Research Scientist (Reproductive Physiology)
Research Centre, Agriculture and Agri-Food Canada, Lethbridge, Alberta, Canada T1J 4B1
Telephone: 403-317-2236 - Fax: 403-382-3156
Email: kastelic@em.agr.ca

The author acknowledges the financial support of Agriculture and Agri-Food Canada, the Alberta Agricultural Research Institute and The Canada/Alberta Livestock Research Trust, Inc.

Abstract

Testicular temperature in bulls must be 2 to 6°C below body temperature to produce fertile sperm. Increasing testicular temperature results in defective spermatozoa, with recovery dependent upon the nature and duration of the thermal insult. The testicular vascular cone, consisting of the pampiniform plexus (a complex venous network) surrounding the highly coiled testicular artery, functions as a counter-current heat transfer system (heat is transferred from the artery to the vein). Heat loss from the scrotal surface (especially from the scrotal neck), scrotal sweating, relaxation of scrotal muscles, and complimentary temperature gradients (associated with blood vessels) in the scrotum and testes also reduce testicular temperature. Despite these features, the testes are very susceptible to temperature increases due to endogenous or exogenous factors (e.g. fever, high ambient temperature). The testes usually operate on the brink of hypoxia; increasing testicular temperature increases metabolism and therefore oxygen requirements, but blood flow changes little, and hence the testes become hypoxic. Future studies of scrotal/testicular thermoregulation in bulls should take into account heat loss from the scrotal surface, blood flow and oxygen saturation in testicular blood vessels and intratesticular tissues.

Key words: scrotum, testes, thermoregulation, semen, spermatozoa, fertility, bull.

Introduction

Bull fertility is of paramount importance in cattle production, as one bull may be responsible for perhaps 20 females under natural service conditions or hundreds of thousands via artificial insemination. Although few bulls are sterile (unable to reproduce), fertility varies considerably among bulls, especially from unselected populations. It is well known that testicular temperature in bulls must be 2 to 6°C below body temperature to produce fertile sperm and that elevated testicular temperatures reduce semen quality (Waites, 1970). Perhaps increased testicular temperature is the underlying cause of many cases of infertility in bulls. Although the counter-current exchange mechanism that cools arterial blood entering the testes is generally regarded as the principal means of reducing testicular temperature, there are many other features that reduce testicular temperature. The physiology of scrotal/testicular thermoregulation, the use of infrared thermography for assessment of scrotal surface temperature, and the effects of increased testicular temperature will be reviewed, with an emphasis on recent studies. In addition, suggestions will be made regarding future areas of investigation.

Anatomy and Physiology

Scrotum, Tunica Dartos and Cremaster

The scrotum has many features that help to regulate testicular temperature. Skin on the bovine scrotum is usually thin with little hair. Scrotal subcutaneous tissues have an extensive blood and lymphatic system, with blood vessels near the skin surface, facilitating radiation of heat (Dahl & Herrick, 1959). The warmest part of the scrotum (Coulter, 1988) is the neck (just below the attachment of the scrotum to the abdomen). A long, distinct scrotal neck provides considerable area for heat loss and moves the testes away from the abdomen. However, an extremely long scrotal neck may predispose the testes to trauma. Conversely, selection for large scrotal circumference (without regard to the length of the scrotal neck) may result in bulls with short, wide testes that are held closely to the body. Therefore, selection of breeding bulls should include assessment of scrotal and testicular shape and the proximity of the testes to the abdomen.

Two muscles are important in controlling testicular location relative to the abdomen. The tunica dartos, a thin sheet of smooth muscle underlying the scrotal skin, is controlled by sympathetic nerves and responds to changes in ambient temperature (Setchell, 1978). In warm environments, this muscle relaxes, allowing the testes to move away from the abdomen. In cold environments, contraction of this muscle (sustainable over prolonged periods) holds the testis close to the abdomen (Setchell, 1978). Although contraction of the cremaster muscle also draws the testis closer to the body, this is a striated muscle and therefore probably cannot sustain contraction for prolonged periods (Setchell, 1978).

Testicular Vascular Cone

The pampiniform plexus is a complex venous network surrounding the highly coiled testicular artery; the entire structure (venous network and artery) is properly called the 'testicular vascular cone' (Hees et al., 1984). The testicular vascular cone functions as a classical counter-current heat transfer system; heat is transferred from the warm blood in the testicular artery to the cooler blood in the testicular venous system. In bulls, the efficiency of heat transfer averaged 91% (Glad Sorensen et al., 1991). Furthermore, this is an important site of surface heat loss as the skin overlying the cone is usually the warmest area on the scrotum (Coulter, 1988).

Characteristics of the testicular vascular cone and scrotal surface temperature (SST) were recently studied in bulls that were 0.5, 1, 2, and 3 years of age (Cook et al., 1994). The testicular artery became profoundly larger between 6 months and 12 months of age (an interval during which most bulls reach puberty). Thickness of the wall of the testicular artery and the distance between the artery and the vein decreased with age; furthermore, both were less at the bottom of the cone

compared to the top. Thinner arterial walls and a shorter arterial-venous distance at the bottom of the cone may promote heat transfer. There was no significant effect of bull age on top, bottom, or average SST, or on top-to-bottom SST gradient. Therefore, despite profound changes in the size of the scrotum, testes, and testicular vascular cone with age, there were apparently compensatory mechanisms that maintained SST. Further study of the effects of puberty on scrotal/testicular thermoregulation in bulls is needed.

Local and Systemic Responses

Smooth muscle in cutaneous arterioles of the scrotum are innervated by postganglionic sympathetic neurons in the lumbar sympathetic chain (Langely & Anderson, 1895); stimulation of these neurons causes vasoconstriction (Langely & Anderson, 1895). An increase in scrotal temperature probably causes dilation of these arterioles by direct action of heat, reflex removal of sympathetic vasoconstrictor tone, and ultimately (if deep body temperature rises) complete removal of vasoconstrictor tone (Setchell, 1978).

Sweating is an important component of testicular cooling and has been well characterized in sheep. In Merino rams, scrotal sweat glands are larger and produce more sweat than those elsewhere on the body (Waites & Voglmayr, 1962). Apocrine sweat glands in the scrotum of rams discharge simultaneously; expulsion begins when scrotal surface temperature (SST) is about 35.5°C and occurs at a frequency of up to 10 discharges every hour (Waites & Voglmayr, 1963). In bulls, the volume of sweat glands per unit skin surface area of the scrotum was greater than that of other body regions (Blazquez et al., 1988). Sweating is a neural reflex, with afferent receptors in the scrotal skin and efferent sympathetic adrenergic nerves (Setchell, 1978).

There are also whole-body responses to increased temperatures. In fully fleeced rams, respiration rate increases when SST rises above 35 to 36°C (Setchell, 1978). Furthermore, when SST reaches 38 to 40°C, respiration rate increases to 200 breaths per minute and temperatures within the rectum and carotid artery can decline as much as 2°C in one hour (Waites, 1962). There is also peripheral vasodilation in the skin of the ears, abdomen and limbs but no change in the metabolic rate. It is noteworthy that warming flank skin (equal in area to the scrotum) does not cause these dramatic responses (Setchell, 1978). However, the responses are different in rams that have been shorn (wool removed). In these rams, skin temperature is decreased and the metabolic rate is increased (to keep them warm). Warming the scrotum in these rams results in a decrease in metabolic rate without an increase in respiration rate (Hales & Hutchinson, 1971). Similar studies have apparently not been conducted in bulls.

Surface and Internal Temperatures

Sixteen two-year-old beef bulls were used in a recent experiment to determine scrotal surface and internal scrotal/testicular temperatures in bulls (Kastelic et al., 1995). Infrared thermography was used to measure SST. Under caudal epidural

analgesia (xylazine HCl, 40 mg), scrotal subcutaneous temperature (SQT) and intratesticular temperature (ITT) were measured with needle thermistors at three locations in each testis: top, middle and bottom. Average temperatures (°C) at these locations were: 30.4, 29.8 and 28.8 (SST); 33.3, 33.0 and 32.9 (SQT); and 34.3, 34.3 and 34.5 (ITT). For SST, temperatures at all three locations were significantly different from one another. Top-to-bottom temperature gradients were 1.6 (SST), 0.4 (SQT), and -0.2°C (ITT). Therefore, the temperature gradient was most pronounced on the scrotal surface, small in the scrotal subcutaneous tissues, and was not present within the testicular parenchyma. Similar temperature gradients are present in rams (Kastelic et al., 1999). In a subsequent study (Kastelic et al., 1996[a]), it was shown that the scrotum and testes have opposing temperature gradients which apparently complement one another, resulting in a relatively uniform ITT. Furthermore, the scrotum substantially increased ITT, but SST was not significantly affected by the presence of a testis. These gradients may be due to vasculature. The scrotum is vascularized from top to bottom. However, the testicular artery (after exiting the ventral aspect of the testicular vascular cone) courses the length of the testis and then diverges into several smaller arteries that spread dorsally and laterally across the surface of the testis before entering the testicular parenchyma (Gunn & Gould, 1975). Therefore, the testis is vascularized from the bottom to the top. In a recent study (Kastelic et al., 1997) it was shown that blood within the testicular artery has a similar temperature at the top of the testis (just ventral to the testicular vascular cone) compared with the bottom of the testis, but was significantly cooler at the point of entry into the testicular parenchyma (intra-arterial temperatures 34.3, 33.4 and 31.7°C, respectively). Therefore, both the scrotum and testis are warmest at the origin of their vascular supply (top of scrotum, bottom of testis) with a reduction in temperature towards the opposite pole.

In the previous study in bulls (Kastelic et al., 1995), intraepididymal temperatures (IET) of the head, body and tail averaged 35.6 ± 0.2, 34.6 ± 0.1 and 33.1 ± 0.3°C. The difference between the IET of the head and the tail of the epididymis averaged 2.5°C. The IET of the head of the epididymis exceeded that of the top ITT, probably due to the proximity of the head of the epididymis to the testicular vascular cone, which is dorsal to the testis and is usually the area of highest SST. However, the tail of the epididymis, an important site for sperm storage and maturation, was slightly cooler than the testicular parenchyma.

Models of Testicular Thermoregulation

Although many components of the mechanisms controlling temperature regulation in the scrotum and testes have been studied, there have been few attempts to develop a comprehensive model. A computer model (Sealfon & Zorgniotti, 1991) based on analysis of an electronic circuit, has been developed to explain thermoregulation of human testes but has apparently received limited attention. This

model appears to predict human testis temperature data gathered in previous studies. The model accounts for countercurrent heat exchange in the testicular vascular cone and predicts, with an open loop analysis, that there is no feedback or regulation. In the absence of feedback, changes in ambient or body temperature affect testicular temperature. In contrast, most physiological systems have feedback mechanisms that attempt to maintain homeostasis. The model predicts that the countercurrent heat exchanger will become less effective as the temperature gradient across the exchanger becomes smaller and less heat can be transferred from the artery to the vein. The countercurrent heat exchanger cools arterial blood by transferring heat from the artery to the vein and the heat is subsequently dissipated from the scrotal surface. Therefore, reduced blood flow in the vein (e.g. varicocele of the testicular vein) or reduced heat loss from the scrotal surface (e.g. due to tight clothing) will result in higher testicular temperature, consistent with empirical observations. The model also correctly predicted that a fever will increase testicular temperature; the warmer arterial blood increases testicular temperature. In the human, it appears that any internal or external factor causing a temperature change will not activate a feedback mechanism to control the resulting change in testicular temperature.

In a recent study (Barros et al., 1999), testicular blood flow and oxygen uptake and the importance of blood flow versus metabolism as sources of testicular heat were determined in bulls. Eight post-pubertal Angus bulls (approximately 14 months of age and 500 kg), were used. The testicular artery and vein were exposed dorsal to one testis. Blood flow in the artery (measured with an electromagnetic flow monitor) was 12.4+1.1 ml/min (mean+SE). Blood in the artery was warmer (39.2+0.2 versus 36.9+0.4 ^{0}C, $P<0.001$) and had a higher percentage of hemoglobin saturated with oxygen than blood in the vein (95.3+0.7 versus 42.0+5.8%, $P<0.001$). Based on blood flow and hemoglobin saturation, oxygen used by one testis (1.2+0.2 ml of oxygen) would be expected to produce 5.8+0.8 calories of heat/minute, compared to 28.3+5.1 calories/minute attributed to the blood (approximately a 5-fold difference). Similarly, in the human model described above (Sealfon and Zorgniotti, 1991), the amount of heat carried into the testes by the blood was relatively much greater than the amount of heat produced by testicular metabolism. This study has important implications. The testis usually operates on the brink of hypoxia (Setchell, 1978). Increased temperature increases metabolism and therefore oxygen requirements also increase. However, studies in rams (Setchell, 1978) have shown that blood flow changes little in response to increases in testicular temperature and consequently the testes become hypoxic. Increasing blood oxygen saturation is not a practical option as the blood is nearly 100% saturated under normal conditions. Increasing blood flow would increase the delivery of oxygen, but would also bring considerable additional heat into the testes. Increasing heat loss from the scrotum is perhaps the best option. A comprehensive model of scrotal/testicular thermoregulation in the bull needs to include estimates of heat loss from the scrotal surface. Furthermore, measurement

of intratesticular tissue oxygen pressure would also be very useful; this has already been done in the testes of mice (Klotz et al., 1996).

Evaluation of Scrotal Surface Temperature

Infrared Thermography

Infrared thermography is a rapid, noninvasive method of assessing SST. The bull is restrained and the posterior surface of the scrotum is imaged by holding the camera approximately 1 meter behind the bull. A grey-scale or color image of the scrotum is produced. Immediate assessments can be made visually and the image saved for computerized image analysis. Recent developments in infrared systems have resulted in substantial reductions in the size (similar to a digital or video camera) and cost of infrared cameras, making this technology more convenient and practical for research and clinical evaluation of scrotal surface temperatures.

Scrotal thermograms of bulls with apparently normal scrotal thermoregulation had left-to-right symmetry and temperatures that were 4 to 6°C higher at the top of the scrotum than at the bottom (Purohit et al., 1985; Coulter, 1988). More random temperature patterns, often lacking left-to-right symmetry and having localized areas of increased temperature ("hot spots") were interpreted as abnormal thermoregulation of the testes or epididymides and the bulls usually had semen of poor quality (Purohit et al., 1985; Coulter, 1988). Conversely, not every bull with poor quality semen has an abnormal thermogram. In bulls with unilateral orchitis, the SST was greater over the affected testis compared to the other testis (Purohit et al., 1985).

In rams (Coulter et al., 1988) SST (measured by infrared thermography) was highly correlated with both scrotal subcutaneous temperature and with the temperature of a water-filled balloon (acting as a surrogate testis). However, subsequent investigations indicate that caution must be exercised when making inferences about intratesticular temperature based on measurement of SST (Kastelic et al., 1995).

Environmental Factors Affecting Infrared Thermography

Environmental factors affecting measurement of SST in bulls were recently studied (Kastelic et al., 1996[d]). Measurements can be performed at any time of the day, but should be performed at least 1 hour after rising and either prior to feeding or several hours thereafter. The scrotum should be dry. If the scrotum is wet or if must be moistened and cleaned, at least 30 minutes should be allowed for SST to return to normal. Although SST can be measured over a wide range of ambient temperatures, moderate to cool ambient temperatures (range approximately 5 to 15°C) are ideal. Ambient temperature has the greatest effect on bottom SST and the least effect on top SST. The SST gradient (difference between top and bottom SST) is inversely

related to ambient temperature. Abrupt changes in ambient temperature should be avoided as they may result in artifacts due to overcompensation. Ejaculation (either spontaneous or electroejaculation) results in an increase in SST in the area of the scrotum overlying the cauda epididymides (Kastelic et al., 1996[b]). Therefore, infrared thermography should be performed prior to semen collection.

Infrared Thermography for Breeding Soundness Evaluation

Thermography has been used as an adjunct to the standard breeding soundness examination. In one study (Lunstra & Coulter, 1997), 30 yearling bulls that were satisfactory on a standard breeding soundness examination, were each exposed to approximately 18 heifers for a 45-day breeding period. For bulls with a SST pattern that was classified as normal or questionable, pregnancy rates 80 days after the end of the breeding season were similar (83 ± 3%, n = 13 versus 85 ± 4%, n = 9), but were higher (P<.01) than pregnancy rates for bulls with an abnormal SST pattern (68 ± 4%, n = 8).

Effects of Increased Testicular Temperature

Increased Ambient Temperature

Bulls have been exposed to increased ambient temperature and the effect on semen quality has been determined in many studies. In one study (Casady et al., 1953), Guernsey bulls (n=2) were exposed to 37°C and 81.4% relative humidity for 12 hours per day for 17 consecutive days. The total number of spermatozoa, sperm concentration, and motility decreased profoundly but ejaculate volume and libido remained approximately constant. Approximately 30 to 40% of the spermatozoa were morphologically abnormal (mostly coiled tails and detached heads). Another two bulls were maintained continuously at 30°C and 73% relative humidity for 36 consecutive days, with a similar decrease in semen quality. In another study (Skinner & Louw, 1966), it was concluded that ambient temperatures of 40°C at a relative humidity of 35 to 45% for as little as 12 hours reduced semen quality.

Bos taurus bulls are more susceptible to high ambient temperatures than Bos indicus bulls (Skinner & Louw, 1966). In one study (Johnston et al., 1963), purebred Holstein, purebred Brown Swiss and crosses of these two breeds with Red Sindhi (a Bos indicus breed) were exposed to ambient temperatures of 40°C with 54% relative humidity for 8 hours per day for 7 consecutive days. The remainder of the 24-hour period the conditions were 28°C and 72% relative humidity. Purebred bulls had higher temperatures for the flank (39.5 versus 39.1°C), scrotal skin (38.2 versus 37.4°C) and rectum (40.6 versus 39.4°C) than did the crossbreds. In crossbred bulls, decreases in semen quality were less severe, occurred later and they recovered more rapidly. There were significant decreases in motility, sperm concentration and total

sperm number. There were marked declines in semen quality, especially in purebred bulls (approximately twice the increase in defective spermatozoa compared to crossbred bulls), 1 to 2 weeks after the onset of heat stress. The testes of 3 of the 4 purebred bulls became soft during heat stress. Sperm abnormalities included coiled and bent midpieces, and pyriform heads. There was improvement in semen quality 9 weeks after the heat stress (end of observation period); recovery was not complete at this time, but was more advanced in the crossbred bulls compared to the purebred bulls.

Scrotal Insulation

In a classical study (Lagerlof, 1938), insulation of the scrotum for 4 to 5 days resulted in many morphologically abnormal spermatozoa (particularly pyriform heads) and some decrease in motility. However, the number of spermatozoa was not affected and the bulls recovered in 4 to 6 weeks. Following a longer period of scrotal insulation (11 to 16 days), nearly all spermatozoa were morphologically abnormal and both motility and the number of spermatozoa were profoundly decreased. Sperm production was inhibited for approximately 4 months and an additional 3 months elapsed before sperm morphology returned to normal.

The scrotum of Hereford bulls (n = 12) was insulated for 0 (control), 24 or 72 hours (Austin et al., 1961). Sperm concentration decreased significantly 4 to 7 weeks after insulation in the group insulated for 72 hours, but decreased only slightly in the group insulated for 24 hours. Sperm motility was decreased 2 to 5 weeks after insulation (significant only at 3 weeks) and had returned to normal by 6 weeks. Both insulated groups had about 60% as many normal spermatozoa as non-insulated bulls 2 to 3 weeks after insulation, with no difference apparent by 6 weeks after insulation. The morphologic defects were mainly detached heads with a smaller increase in the incidence of coiled tails.

Short-term (10 or 20 hours) scrotal insulation resulted in spermatozoa with abnormal morphology from 3 to 9 weeks after insulation (Ross & Entwistle, 1979). Intermediate and B-type spermatogonia, spermatocytes through the long meiotic prophase and meiosis, and round and elongating spermatids (but not types A_0 and A_1 spermatogonia) were affected. Epididymal passage time (13.4 days) was similar in control and insulated bulls.

In one study (Wildeus & Entwistle, 1983) the scrotum of Bos indicus x Bos taurus bulls was insulated for 48 hours. Semen was collected every second or third day and all bulls were killed 23 days after insulation. There was no significant effect of insulation on sperm output. The nature and time (relative to the start of insulation) of morphologically abnormal spermatozoa were: decapitated, 6 to 14 d; abnormal acrosomes, 12 to 23 d; abnormal tails, 12 to 23 d; and protoplasmic droplets, 17 to 23 days. Therefore, scrotal heating affected spermatozoa in the caput epididymis as well as spermatids. Although daily sperm production was not affected, epididymal sperm reserves were reduced by nearly 50% (9.2 billion versus 17.4 billion), particularly

in the caput (3.8 vs 6.6 billion) and cauda (3.7 versus 9.5 billion). Perhaps selective resorption of abnormal spermatozoa in the rete testis and excurrent ducts may have reduced epididymal sperm reserves and the number of abnormal spermatozoa in the ejaculate. In a second study (Wildeus & Entwistle, 1986), similar bulls were insulated for 48 hours. The effects on semen quality were highly variable among bulls. The following defects were noted: decapitated sperm, 12 to 16 days; droplets, 16 to 20 days; and sperm tail abnormalities, 26 days. There was a marked reduction in the number of spermatozoa in the corpus and cauda 26 days after scrotal insulation, perhaps due to resorption of spermatozoa in the excurrent ducts.

In another laboratory, the scrotum of six Holstein bulls was insulated for 48 hours (Day 0 = initiation of insulation) and two ejaculates per day were collected with an artificial vagina every 3 days from Day -6 to Day 39 (Vogler et al., 1991, 1993). The number of spermatozoa collected were not significantly affected but the proportions of progressively motile spermatozoa decreased from 69% (prior to insulation) to 42% (on Day 15). The proportions of abnormal spermatozoa were not significantly different from Day -6 to Day 9 (19.6%), increased abruptly on Day 12 (47%) and peaked on Day 18 (86.3%). Although there was considerable variation among bulls in the type and proportion of abnormal spermatozoa that were present in the ejaculate, specific abnormalities appeared in a consistent chronological sequence as follows: tailless, Days 12 to 15; diadem, Day 18; pyriform and nuclear vacuoles, Day 21; knobbed acrosome, Day 27; and Dag defect, Day 30. When spermatozoa were examined immediately after collection, there were no differences between samples collected 3 to 9 days after insulation and those collected prior to insulation. However, following freezing, thawing and incubation at 37°C for 3 hours, there were significant differences in the proportion of progressively motile spermatozoa (30.8 versus 46.4%) and the proportion of spermatozoa with intact acrosomes (62.8 verus 73.0%) compared to semen collected prior to insulation (Vogler et al., 1991). Therefore, the stress of freezing plus post-thaw incubation enabled detection of changes that had occurred in spermatozoa that were in the epididymis at the time of scrotal insulation.

In a recent study (Fonseca & Chow, 1995), testicular degeneration was induced in 8 zebu bulls by insulating the scrotum for a period of 168 h. From 7 to 63 days after insulation, there was a decrease in sperm motility and concentration and an increase in the incidence of sperm abnormalities; severe oligoastenospermia was observed 7-21 days after treatment and oligonecrospermia from Day 21 to Day 63. Sperm regeneration began on Day 63 and was complete by Day 105. At the initial stages of the degeneration process, the most frequent abnormalities were proximal cytoplasmic droplets and sperm head and tail abnormalities; the knobbed sperm defect was observed most frequently in the middle of the period. During the degeneration process, the incidence of underdeveloped spermatozoa, proximal cytoplasmic droplets, sperm abnormalities and isolated sperm heads was 27, 19,15, and 12% respectively.

In a recent study (Barth & Bowman, 1994), scrotal insulation (4 days) and dexamethasone treatment (20 mg/day for 7 days) were used as models of testicular heating and stress, respectively. Semen was collected by electroejaculation two or three times weekly for 6 weeks after insulation or dexamethasone treatment. Some bulls seemed predisposed to produce spermatozoa with a particular abnormality. Pyriform heads, nuclear vacuoles, microcephalic sperm, and abnormal DNA condensation were more common in insulated than dexamethasone-treated bulls. Conversely, dexamethasone treatment resulted in an earlier and more severe effect on epididymal spermatozoa, an earlier and greater increase in distal midpiece reflexes, and and earlier increase in proximal and distal droplets. Notwithstanding, in general the types of defective spermatozoa and the time of their detection were similar for the two treatments.

Insulation of the Scrotal Neck

In a recent experiment (Kastelic et al., 1996[c]), the neck of the scrotum of 5 bulls was insulated for 7 days (Days 1 to 8) as a model of over-conditioned bulls that have considerable fat in the neck of the scrotum. Semen was collected by electroejaculation approximately every 3 days from Day -3 to Day 35. Spermatozoa within the epididymis or at the acrosome phase during insulation appeared to be most affected. Insulated bulls had twice as many spermatozoa with midpiece defects and four times as many with droplets on Day 5, fewer normal spermatozoa and three times as many with midpiece defects and droplets on Day 8, fewer normal spermatozoa on Days 15 and 18, and more spermatozoa with head defects on Days 18 and 21. Semen quality in insulated bulls had nearly returned to pre-insulation values by Day 35. In a second experiment, the scrotal neck was insulated for 48 hours. At 24 hours after insulation, the SST had decreased at the top of the testis but had increased at the bottom of the testis, resulting in a significant reduction in SST gradient. However, by 48 hours, SST was not significantly different from that prior to insulation. In contrast, SQT increased 2.0, 1.5 and 0.5°C at the top, middle and bottom of the testis, respectively, and ITT was increased 0.9°C at the corresponding three locations within the testis 48 hours after insulation (compared to pre-insulation). Therefore, compensatory mechanisms were apparently able to restore SST but not SQT or ITT.

Increased Epididymal Temperature

In the majority of animals, the cauda epididymis is somewhat or substantially cooler than the testes (Bedford, 1991). This lower temperature facilitates the function of the cauda as an important site of sperm storage. Experimentally moving the cauda into the abdomen and exposing it to deep body temperature rapidly disrupts the normal absorbtive and secretory functions of the cauda epithelium and changes the composition (ions and proteins) of the cauda fluid. Furthermore, sperm move much more rapidly through the cauda (transit time is decreased from

approximately 10 d to 3-4 d in rabbits) and therefore the number of spermatozoa stored in the cauda decreases dramatically (Bedford, 1991). Consequently, the number of sperm in the first ejaculate declines, with an even more dramatic decline in sperm number in successive ejaculates. In addition, the increased temperature seems to prematurely hasten sperm maturation as the time required for capacitation of hamster sperm is lower when they have been recovered from a cauda maintained at an elevated temperature (Bedford, 1991).

Summary of Increased Testicular Temperature

There are many potential causes of increased testicular temperature, including inflammation of the testes and/or scrotum, fever, or prolonged recumbency (e.g. due to lameness). When scrotal/testicular temperature is increased (e.g. under heat stress), semen morphology usually appears normal for a short period, corresponding to transit time through the epididymis, and then begins to decline (Barth & Oko, 1989) In some studies (Wildeus & Entwistle, 1983; Kastelic et al., 1996[c]), spermatozoa that would have been in the epididymis at the time of scrotal heating were morphologically abnormal when collected soon after heating. In another study (Vogler et al., 1991), changes in spermatozoa present in the epididymis at the time of scrotal insulation were manifest only after the spermatozoa were stressed. Sertoli and Leydig cell function seem adversely affected by heating, while germ cells are the most heat-sensitive cells in the testis (Waites & Setchell, 1990). All stages of spermatogenesis are susceptible to heating, with the extent of damage related to the extent and duration of the thermal insult (Waites &Setchell, 1990). Spermatocytes in meiotic prophase are killed by heat; sperm that are more mature have metabolic and structural abnormalities (Setchell et al., 1971). Heating the testis usually decreases the proportion of progressively motile and live spermatozoa, and increases the incidence of morphologically abnormal spermatozoa, especially head defects (Barth & Oko, 1989). Although there is considerable variation among bulls in the nature and proportion of defective spermatozoa, the order of appearance of specific defects is relatively consistent (Vogler et al., 1993; Barth & Bowman, 1994). Unless spermatogonia are affected, the interval from cessation of heating to restoration of normal spermatozoa in the ejaculate corresponds to the interval from the beginning of differentiation to ejaculation (Waites & Setchell, 1990). In most of the studies reviewed above, sperm morphology had returned to pre-treatment values within approximately 6 weeks of the thermal insult. Very prolonged or severe heating of the testes will prolong the interval for recovery. However, even when sperm cell morphology has returned to normal, their utilization may result in decreased fertilization rates and an increased incidence of embryonic death (Burfening & Ulberg, 1968). It appears that the decrease in semen quality associated with increased testicular temperature is ultimately related to the severity and duration of the increased temperature.

Concluding Remarks

Scrotal/testicular thermoregulation is clearly a complex phenomenon. Numerous local mechanisms, including countercurrent heat exchange, regulation of blood flow, position of the testes, and sweating, all have a role in keeping the testes cooler than body temperature. Despite these control mechanisms, the testes are very susceptible to temperature increases. Increasing testicular temperature results in defective spermatozoa, with recovery dependent upon the nature and duration of the insult. Further research is needed to improve our understanding of thermoregulatory mechanisms in the normal male and in those with infertility.

Acknowledgments

The author wishes to thank the many persons who have assisted him in this area of study.

References

Austin JW, Hupp EW, Murphee RL. Effect of scrotal insulation on semen of Hereford bulls. J Anim Sci 1961;20:307-10.

Barros CMQ, Brito LFC, Oba E, Cook RB, Coulter GH, Groves G, Olson M, Kastelic JP. Testicular blood flow and oxygen utilization in Angus bulls. Rev Brasil Repro Anim 1999;23:218-20.

Barth AD, Bowman PA. The sequential appearance of sperm abnormalities after scrotal insulation or dexamethasone treatment in bulls. Can Vet J 1994;35:93-102.

Barth AD, Oko RJ. Abnormal morphology of bovine spermatozoa. Ames (IA): Iowa State University Press, 1989:139.

Bedford JM. Effects of elevated temperature on the epididymis and testis: experimental studies. In: Zorgniotti AW, ed. Temperature and environmental effects on the testis. New York: Plenum Press, 1991:19-32.

Blazquez NB, Mallard GJ, Wedd SR. Sweat glands of the scrotum of the bull. J Reprod Fertil 1988;83:673-7.

Burfening PJ, Ulberg LC. Embryonic survival subsequent to culture of rabbit spermatozoa at 38º and 40º. J Reprod Fert 1968;15:87-92.

Casady RB, Myers RM, Legates JE. The effect of exposure to high ambient temperature on spermatogenesis in the dairy bull. J Dairy Sci 1953;36:14-23.

Cook RB, Coulter GH, Kastelic JP. The testicular vascular cone, scrotal thermoregulation, and their relationship to sperm production and seminal quality in beef bulls. Theriogenology 1994;41:653-71.

Coulter GH. Thermography of bull testes. In: Proceedings of the 12th Tech. Conf. Artific. Insem. Reprod. Milwaukee, WI; Columbia, MO: National Association of

Animal Breeders, 1988:58-63.

Coulter GH, Senger PL, Bailey DRC. Relationship of scrotal surface temperature measured by infrared thermography to subcutaneous and deep testicular temperature in the ram. J Reprod Fert 1988;84:417-23.

Dahl EV, Herrick. A vascular mechanism of maintaining testicular temperature by counter-current exchange. Surg Gynecol Obstet 1959;108:697-705.

Fonseca VO, Chow LA. Características seminais de touros zebus com degeneração testicular transitória. Arq Bras Med Vet Zootec 1995;47:707-16.

Glad Sorensen H, Lambrechsten J, Einer-Jensen N. Efficiency of the countercurrent transfer of heat and 133Xenon between the pampiniform plexus and testicular artery of the bull under in-vitro conditions. Int J Androl 1991;14:232-40.

Gunn SA, Gould TC. Vasculature of the testes and adnexa. In: RO Greep, EB Astwood, eds. Handbook of Physiology, Section 7, Endocrinology; Vol. 5, Male Reproductive System. Washington, DC: American Physiological Society, 1975:117-142.

Hales JRS, Hutchinson JCD. Metabolic, respiratory and vasomotor responses to heating the scrotum of the ram. J Physiol 1971;212:353-75.

Hees H, Leiser R, Kohler T, Wrobel KH. Vascular morphology of the bovine spermatic cord and testis. Light and scanning electron microscopic studies on the testicular artery and pampiniform plexus. Cell Tissue Res 1984;237:31-8.

Johnston JE, Naelapaa H, Frye JB. Physiological responses of Holstein, Brown Swiss and Red Sindhi crossbred bulls exposed to high temperatures and humidities. J Anim Sci 1963;22:432-6.

Kastelic JP, Coulter GH, Cook RB. Scrotal surface, subcutaneous, intratesticular, and intraepididymal temperatures in bulls. Theriogenology 1995;44:147-52.

Kastelic JP, Cook RB, Coulter GH. Contribution of the scrotum and testes to scrotal and testicular thermoregulation in bulls and rams. J Reprod Fertil 1996[a];108:81-5.

Kastelic JP, Cook RB, Coulter GH, Saacke RG. Ejaculation increases scrotal surface temperature in bulls with intact epididymides. Theriogenology 1996[b];46:889-92.

Kastelic JP, Cook RB, Coulter GH, Saacke, RG. Insulating the scrotal neck affects semen quality and scrotal/testicular temperatures in the bull. Theriogenology 1996[c];45:935-42.

Kastelic JP, Cook RB, Coulter GH, Wallins GL. Environmental factors affecting measurement of bovine scrotal surface temperature with infrared thermography. Anim Reprod Sci 1996[d];41:153-9.

Kastelic JP, Cook RB, Coulter GH. Contribution of the scrotum, testes, and testicular artery to scrotal/testicular thermoregulation in bulls at two ambient temperatures. Anim Reprod Sci 1997;45:255-61.

Kastelic JP, Cook, RB, Coulter, GH. Effects of ambient temperature and scrotal fleece cover on scrotal and testicular temperatures in rams. Can J Vet Res 1999;63:157-60.

Klotz T, Vorreuther R, Heidenreich A, Zumbe J, Engelmann U. Testicular tissue oxygen pressure. J Urol 1996;155:1488-91.

Lagerlof N. Infertility in male domestic animals. Vet Med 1938;33:550-61.

Langely JN, Anderson H. The innervation of the pelvic and adjoining viscera. Parts III and IV. J Physiol 1895;19:84 and 19:122.

Lunstra DD, Coulter, GH. Relationship between scrotal infrared temperature patterns and natural-mating fertility in beef bulls. J Anim Sci 1997;75:767-74.

Purohit RC, Hudson RS, Riddell MG, Carson RL, Wolfe DF, Walker DF. Thermography of the bovine scrotum. Am J Vet Res 1985;46:2388-92.

Ross AD, Entwistle KW. The effect of scrotal insulation on spermatozoal morphology and the rates of spermatogenesis and epididymal passage of spermatozoa in the bull. Theriogenology 1979;11:111-29.

Sealfon AI, Zorgniotti AW. A theoretical model for testis thermoregulation. In: Zorgniotti AW, editor. Temperature and environmental effects on the testis. New York: Plenum Press, 1991:123-35.

Setchell BP, Voglmayr JK, Hinks NT. The effect of local heating on the flow and composition of rete testis fluid in the conscious ram. J Reprod Fert 1971;24:81-9.

Setchell BP. The scrotum and thermoregulation. In: Setchell BP. The mammalian testis. Ithica NY: Cornell University Press, 1978:90-108.

Skinner JD, Louw GN. Heat stress and spermatogenesis in Bos indicus and Bos taurus cattle. J Applied Physiol 1966;21:1784-90.

Vogler CJ, Saacke RG, Bame JH, DeJarnette JM, McGilliard ML. Effects of scrotal insulation on viability characteristics of cryopreserved bovine semen. J Dairy Sci 1991;74:3827-35.

Vogler CJ, Bame JH, DeJarnette JM, McGilliard ML, Saacke RG. Effects of elevated testicular temperature on morphology characteristics of ejaculated spermatozoa in the bovine. Theriogenology 1993;40:1207-19.

Waites GMH. The effect of heating the scrotum of the ram on respiration and body temperature. Quart J Exptl Physiol 1962;47:314-23.

Waites GMH, Voglmayr JK. Apocrine sweat glands of the scrotum of the ram. Nature 1962;196:965-7.

Waites GMH, Voglmayr JK. The functional activity and control of the apocrine sweat glands of the scrotum of the ram. Aust J Agric Res 1963;14:839-51.

Waites GMH. Temperature regulation and the testis. In: The testis. Johnson AD, Gomes WR, Vandemark NL eds. Academic Press, NY, 1970;241-79.

Waites GMH, Setchell BP: Physiology of the Mammalian Testis. In: Lamming GE, ed. Marshall's Physiology of Reproduction, Fourth Edition, Volume 2, Reproduction in the Male. Edinburgh: Churchill Livingstone, 1990:1-105.

Wildeus S, Entwistle KW. Spermiogram and sperm reserves in hybrid Bos indicus x Bos taurus bulls after scrotal insulation. J Reprod Fert 1983;69:711-16.

Wildeus S, Entwistle KW. Effects of scrotal insulation and unilateral vasoligation on ejaculate characteristics and sperm reserves in the bull. Anim Reprod Sci 1986;10:11-21.