American Journal of Animal and Veterinary Sciences

Original Research Paper

Advanced Assessing Techniques of the Reproductive Performance of Bulls

Gulzat Turysbayeva, Isatay Jakupov, Aisulu Kuzerbayeva and Zhanargul Zharkimbayeva

Department of Veterinary Medicine, Faculty of Veterinary and Animal Husbandry Technology, S. Seifullin Kazakh Agro Technical University, Astana, Kazakhstan

Article history
Received: 22-08-2022
Revised: 26-12-2022
Accepted: 28-12-2022

Corresponding Author:
Isatay Jakupov
Department of Veterinary Medicine, Faculty of Veterinary and Animal Husbandry Technology, S. Seifullin Kazakh Agro Technical University, Astana, Kazakhstan
Email: ijakupov@bk.ru

Abstract: The sperm quality supplied by producers is an urgent problem in the field of livestock breeding. Therefore, monitoring the health of male producers and sperm quality is of great priority. The study aimed to assess the reproductive performance of bulls using laboratory, clinical, morphological, ultrasound and reflexological techniques. A total of 17 heads of Kazakh-white-headed, Holstein-Frieze and Simmental breeds were studied. The quality of sperm was evaluated by the laboratory, clinical, morphological, ultrasound and reflexological diagnostic methods were used to determine the physiological state of the genitals. The results revealed that 11 (65%) bulls were included with high and good reproductive bulls and 6 (35%) with reduced reproductive abilities. The reliability of the measuring method for the morphological development of the scrotum and ultrasound coincides with the data of sperm quality by 66.6% and studying sexual reflexes on a point scale by 50%. When assessing the male reproductive performance, measuring the morphological development of the testes, studying sexual reflexes on a point scale and ultrasound examination of the testes and their appendages are recommended.

Keywords: Bulls, Andrological Medical Examination, Clinical, Reflexological Examination, Evaluation of Sperm Quality

Introduction

One of the main tasks of animal husbandry is breeding animals with high reproductive capacity. Fertility is crucial for improving the reproductive performance of animals (Sabés-Alsina *et al*., 2019).

The best bull by origin, exterior and constitution is of breeding value only if it has sufficient sexual activity and can produce good quality seed. Therefore, the intensive use of breeding bulls under different circumstances is a quantitative and qualitative indicator of sperm quality and production (Abilov and Kolosova, 2007; Kapš *et al*., 2000; Butler *et al*., 2020).

The reproductive physiological functions of bulls are determined by clinical examination of the body systems and genitals, the severity of the manifestation of sexual reflexes and laboratory evaluation of sperm quality (Dahl *et al*., 2020; Barth, 2018; Johnston *et al*., 2013).

Clinical examination of the genitals in breeding bulls is carried out by examining the scrotum (symmetry, presence of scars, edema, rash and neoplasms), foreskin (configuration, condition of the hairline, presence of lesions and rash), palpation of the scrotum (temperature, thickness and mobility of layers), testes, their appendages and seminal tubules (position, shape, mobility, consistency, soreness and size), sigmoid bend and the terminal part of the penis (configuration, mobility and soreness) (Dahl *et al*., 2020; Brown and Vosloo, 2017; Kasimanickam *et al*., 2014).

Brazilian scientists (Carrer Filho *et al*., 2015; Lees *et al*., 2019) conducted a complete andrological examination (assessment of scrotum parameters and physico-morphological parameters of sperm) of 36 Bradford bulls. They divided the bulls according to andrological classification by Classification by Points (CAP) and a libido test in a pen lasting 15 min into four categories: Excellent from 9 to 10 points; very good from 7 to 8 points; good from 4 to 6 points; doubtful from 0 to 3 points.

Medvedev (2006) determined the reproductive ability of bulls using the circumference of the scrotum horizontally, the transverse circumference of the scrotum and the circumference along the sagittal line.

Ultrasound is recommended as an additional instrumental research method for evaluating the reproductive function of males (Jaskowski *et al*., 2004; Andrade *et al*., 2012; Mee *et al*., 2012) since it is non-

invasive and fast. Ultrasound can be used also to examine the accessory glands in their normal state in the pelvic canal (Tschoner *et al*., 2020; Bova *et al*., 2014; Michi *et al*., 2016).

The physiological state of reproduction depends on many biological and economic factors, one of which is the seed quality of the producers used. With artificial insemination, several thousand cows are inseminated and the use of an infertile or subfertile bull can cause huge damage to the industry. Therefore, the evaluation of the seed should be of a complex nature (Bagath *et al*., 2019). According to Tokysheva *et al*. (2023); Yu *et al*. (2020); Collantes-Fernández *et al*. (2019), 20% of bull producers are infertile and 40% have low fertilizing capacity.

There are numerous methods for assessing sperm quality. Diagnostic tests can reliably predict fertility manufacturers (Mazer *et al*., 2020; Orihuela and Galina, 2019; Lauder *et al*., 2020).

Currently, a reliable estimation of the reproductive qualities of males is increasingly using computer analyze the quality of the seed (SASA) and DNA technology (Al-Qarawi *et al*., 2002; Heimbürge *et al*., 2020; Chen *et al*., 2015; Fernandez-Novo *et al*., 2020).

The use of different methods of monitoring the health of bulls and determining their reproductive ability is relevant and useful for clinical practice.

In this regard, the study aimed to determine the reproductive ability of breeding bulls using laboratory, clinical, morphological, ultrasound and reflexological methods.

Materials and Methods

The research was conducted at the Department of Veterinary Medicine of the Kazakh Agrotechnical University named after Saken Seifullin, in JSC RCPJ "Asyl-Tulik", located in the Akmola region.

The work was carried out in 2018-2020 on 17 breeding bulls of different ages (2-9 years) meat (Kazakh white-headed), dairy (Holstein-Frisian) and combined (Simmental) breeds in the conditions of JSC "Asyl Tulik". The authors took into account the age of the bulls-producers.

To study the reproductive function of bulls, a laboratory assessment of the semen quality, a clinical study of the sexual apparatus and sexual reflexes were carried out.

The laboratory examinations of the semen included the smell, color, volume of ejaculate, concentration, density and mobility of sperm. the mobility was determined by visual microscopy methods and Computer Assisted Sperm Analysis (CASA) was also used. Computer technology (CASA) significantly increases the accuracy of the assessment and reduces time and improves the quality of control (Al-Qarawi *et al*., 2002; Heimbürge *et al*., 2020; Cooke *et al*., 2019).

An andrological medical examination was carried out by evaluating the macro and microscopic parameters of sperm, examining the state of the genitals and manifestations of sexual reflexes. The animals were divided according to the andrological classification (Jedraszczyk, 2003; Collier and Gebremedhin, 2015; Newcomer *et al*., 2017) into a group with high reproductive ability; good reproductive ability; reduced reproductive ability; and unable to reproduce.

The clinical examination of the genitals of bulls included inspection and palpation.

The morphological development of the seeds measurements included the circumference of the scrotum horizontally, the transverse circumference of the scrotum and the circumference of the scrotum along the sagittal line (Medvedev, 2006; Collier *et al*., 2017; Oguejiofor *et al*., 2019).

Ultrasound examination of the reproductive organs is a method that complements the clinical assessment and is of the greatest importance in cases of subclinical changes (Kolchina and Barashkin, 2011; Michael *et al*., 2019; Olson *et al*., 2019).

The echographic parameters of the genitals were carried out using an ultrasound scanner EMP Veterinary Ultrasound V9 (EMP Medical Headquarters, Shenzhen Emperor Electronic Technology Co., Ltd., China) with a convex sensor with a frequency of 2.5-3.5 MHz.

To study the severity of the manifestation of sexual reflexes in breeding bulls, they were evaluated by the method (Dahl *et al*., 2020; Hughes *et al*., 2014).

The content of the breeding bulls is stable with metal partitions and wooden floors. Forced exercise is provided once a day. The content of animals complies with veterinary and sanitary standards.

The air temperature in the premises for keeping bulls in winter is 10-12°C, in summer 20°C and in transition periods 12°C. Relative humidity 75%, light coefficient 1:10 lm.

Bulls are brushed and washed in the shower. For each manufacturer, there is a separate hair brush with an inscription of a nickname or number.

The feeding system is manual. Water supply from drinkers.

Manure removal is mechanized by the SMC 160 conveyor (scraper manure conveyor):

- Ventilation system and mechanism. Exhaust wall fans provide forced ventilation of the building: Air leaves the room through a fan, and fresh air enters through supply valves
- Fly protection measures, rodent protection measures:
- Preventive measures against insects (disinsection) and rodents (derivatization) are carried out according to plan and if necessary

All events involving animals were held following the "model law on responsible treatment of animals" of the Inter-Parliamentary assembly of the member states of the commonwealth of independent states (March 27, 2017 No. 46-15).

Fig. 1: Premises for keeping breeding bulls (general view)

Fig. 2: Bulls-producers, prepared to take the seed

The content of the breeding bulls is stable with metal partitions, wooden floors. Forced exercise is provided once a day. The content of animals complies with veterinary and sanitary standards in Fig.1 and 2.

The air temperature in the premises for keeping bulls in winter is 10-12°C, in summer 20°C, in transition periods 12°C. Relative humidity 75%, light coefficient- 1:10 lm.

Bulls are brushed, washed in the shower. For each manufacturer there is a separate hair brush with an inscription of a nickname or number.

The feeding system is manual. Water supply from drinkers.

Manure removal is mechanized by the TSN 160 conveyor (scraper manure conveyor):

- Ventilation system and mechanism. Exhaust wall fans provide forced ventilation of the building: Air leaves the room through a fan, and fresh air enters through supply valves
- Fly protection measures, rodent protection measures: Preventive measures against insects (disinsection) and against rodents (deratization) are carried out according to plan and if necessary

A typical room with producing bulls that meets sanitary and hygienic requirements and a favorable microclimate.

The statistical evaluation of the results was carried out by standard methods using statistical software Statgraphics Centurion XVII (StatPoint, USA) multifactor analysis of variance (MANOVA), LSD test. Statistical processing was performed in Microsoft Excel 2016 in combination with XLSTAT. Values were estimated using mean and standard deviations (Tokysheva *et al.*, 2022; Rioja-Lang *et al*., 2020).

Results and Discussion

The object of research was bulls-producers belonging to the Joint-Stock Company RCPZH "Asyl-Tulik", in the amount of 17 heads.

The freshly obtained sperm of healthy 17 bulls (100%) was characterized by a milky white color with a yellowish tinge, viscous creamy consistency, a specific smell and without foreign impurities.

The volume of ejaculate was 4.5 = 0.3 mL, 5.8 = 0.5 mL in Holstein Friesian and 5.3 = 0.6 in Simmental. The results are presented in Table 1.

The density and mobility of sperm from producers, the Kazakh white-headed (n = 7) and Holstein-Frisian breeds (n = 4) was an average and 8 points respectively. Five bulls-producers of Kazakh white-headed and one Simmental breed had below 8 points mobility.

The sperm concentration in the Kazakh white-headed breed (n = 12) was 1.1±0.06 billion/mL, 0.9±0.04 billion/mL in Holstein-Frisian and 0.9±0.07 billion/mL in the Simmental breed. Due to the decrease in sperm motility in 6 bulls (35.3%), an andrological medical examination of 17 breeding bulls was carried out.

To determine the quality of sperm, the state of the genitals and the manifestations of sexual reflexes, animals were divided into four groups: Group I with high reproductive ability; group II with good reproductive ability; group III with reduced reproductive ability; group IV unable to reproduce. The results are shown in Table 2.

Table 1: Indicators of sperm quality of breeding bulls for (14.03.19-06.08.19.)

	Breed	n	Number ejaculates	Ejaculate volume, mL	Sperm motility, score	Sperm concentration, billion/mL
1	Kazakh white-headed	7	30±0,03	4,5±0,3	8,1±0,1	1,1±0,06
2	Holstein	4	32±0,04	5,8±0,5	8±0,01	0,9±0,04
3	Simmental	1	30±0,03	5,3±0,6	7,2±0,01	0,9±0,07
4	Kazakh white-headed	5	30±0,03	5,06±1	6,2±0,7	1,1±0,09

Table 2: The result of the distribution of breeding bulls by groups based on the study of sperm

	Indicators			Breed		
Group	Ejaculate volume, mL	Sperm concentration, billion/mL	Sperm motility,	Kazakh white-headed score/*n* =12	Holstein /*n* = 4	Simmental /*n* = 1
I	At least 4-5	At least 0, 9	More than 8	1(8,3%)	-	-
II	At least 4 mL	More than 0,8	No more than8	6(50%)	4(100%)	-
III	Less than 3 mL	At least 0,8	At least7	2(16,7%)	-	(100%)
IV	Less than 2mL	Less than 0,8	Less than 7	3(25%)	-	-

Table 3: The results of the study of the morphological dimensions of the seeds of bulls-producers

Bulls-producers	Age	Horizontal circumference of the scrotum, mm	Transverse circumference of the scrotum, mm	Scrotum circumference along the sagittal line, mm
The testes are symmetrical	6,2±0,4	441±4,7	454±7,9	383±9,4
Testicles with asymmetry	6,3±1,6	390±13,5	430±13,5	360±13,5

Table 4: Results of the study of bulls by sexual reflexes

No	Group	n	Average, score
1	With a high and good reproductive capacity	10 Kazakh white-headed/4 Holstein	3,6±0,3
2	With a reduced reproductive capacity	2 Kazakh white-headed	2,6
3	Unable to reproduce	1 Simmental	2,2

Table 5: Reproductive capacity of breeding bulls by sperm quality, examination of the sexual apparatus, and sexual reflexes

			Group			
Indicators	Breed	n	I	II	III	IV
Sperm quality	Kazakh white-headed Holstein Simmental	12	1	5	3	3
The sexual apparatus	Kazakh white-headed Holstein Simmental	4		4		
		1				
The sexual reflex	Kazakh white-headed Holstein Simmental	12	9		3	
			1			
		4	4			
		1	1			
		12	4	6	2	
		4	4			
		1				1

Based on the laboratory results of semen quality, one animal (5.8%) of the Kazakh white-headed breed gave an ejaculate in a volume of at least 4-5 mL, a sperm concentration of at least 0.9 billion/mL and mobility of more than 8 points indicating high reproductive capacity.

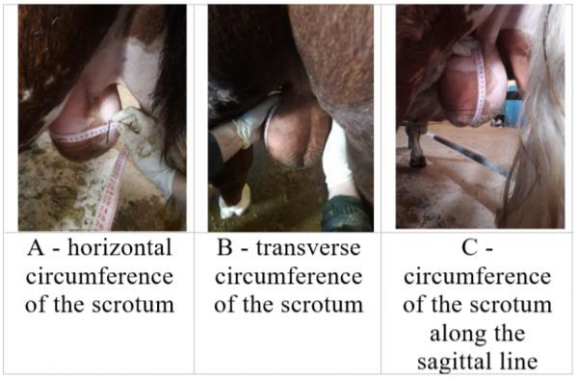
Four (23.5%) Holstein Friesian and six (35.3%) Kazakh white-headed breeds were assigned to the group with good reproductive ability. Bulls with an ejaculate volume of at least 4 mL, a sperm concentration of more than 0.8 billion /mL and mobility of no more than 8 points.

The group with reduced reproductive capacity included two (11.7%) Kazakh white-headed and one Simmental bull with an ejaculate volume < 3 mL, a sperm concentration of 0.8 billion/mL and sperm motility of 7 points. Bulls with reduced sperm motility were assigned to group 4 (unable to reproduce); 3 (17.6%) bulls of the Kazakh white-headed breed.

The results of clinical examination of the genitals have shown that 14 (82.3%) bull has a symmetrical scrotum, painless, without damage with well-defined mobility of layers; testes are symmetrical longitudinally oval with a well-defined outward bulge, elastic-elastic consistency, with a smooth surface and good mobility and painless. The penis is mobile, without mechanical damage or inflammation. Three (17.7%) Kazakh white-headed bulls have testicles with asymmetry of no more than 15-20% and they were classified as group III. At the same time, one bull in terms of sperm quality was assigned to group II and the other 2 bulls in terms of sperm quality to group IV.

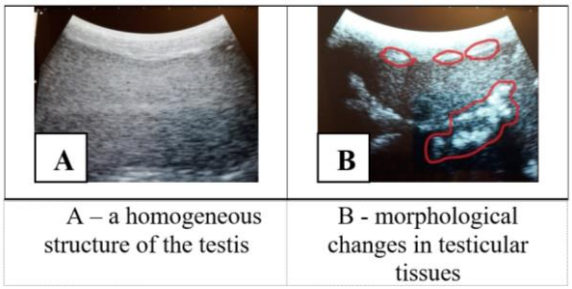
The morphological development of the testes was determined by three parameters (Medvedev, 2006): The circumference of the scrotum horizontally (A); the transverse circumference of the scrotum (B); the circumference of the scrotum along the sagittal line (C) (Fig. 3).

The results of the morphological dimensions of the testes of breeding bulls are shown in Table 3.



A - horizontal circumference of the scrotum | B - transverse circumference of the scrotum | C - circumference of the scrotum along the sagittal line

Fig. 3: Technique of measuring the scrotum and testicles in a bull



A – a homogeneous structure of the testis | B - morphological changes in testicular tissues

Fig. 4: Bull seeds during ultrasound examination

The horizontal circumference of the scrotum in 3 (17.7%) of the Kazakh white-headed breed (one bull from group II and 2 bulls from group IV) revealed an asymmetry of no more than 15-20% is 1.1 times smaller and the transverse circumference of the scrotum is 1.05 times, the circumference of the scrotum along the sagittal line is 1.06 times than in other 14 bulls (82.3%).

To determine the anatomical structure of the testes and assess the morphological state, an ultrasound examination was performed. When examining the testes, the sensor was applied to the caudal side of the testis. The ultrasound results are shown in Fig. 4.

About 11 bulls (65%) testes revealed smooth contours and an oval shape, the protein shell is viewed as a highly echogenic capsule and the mediastinum is in the form of a string of various diameters also of high echogenicity. Parenchyma is represented by a fine grained image with moderate echogenicity. The other six (35%) bulls revealed morphological changes with uneven contours expressed in the parenchyma in 4 bulls, while the quality of sperm (3 bulls of the Kazakh white-headed breed, 1 bull of the Simmental breed) revealed an abundance of hyperechogenic areas scattered in the parenchyma of the testicles, some of them showed acoustic dimming. The normal echogenicity of testes and appendages of testicles was determined in 2 bulls of the Kazakh white-headed breed, despite the poor quality of sperm.

The severity of sexual reflexes in breeding bulls was carried out on a 4-point scale based on the time, strength and nature of their manifestation. In bulls with good and high reproductive capacity, the activity of each sexual reflex was estimated at 3-4 points and the time of manifestation of all sexual reflexes should have been 1-2 min. The results of the study are shown in Table 4.

Ten bulls (83%) of the Kazakh white-headed and 4 (100%) of the Holstein-Frisian breed were assigned to the group with high and good reproductive ability on the scale of severity of sexual reflexes 3-4 points. Two (17%) bulls of the Kazakh white-headed breed were assigned to group III and 1 bull of the Simmental breed with inhibition of sexual reflexes was estimated at 2.2 points for group IV.

According to the laboratory evaluation of sperm quality, clinical examination of the reproductive apparatus by examination, palpation, morphological development of semen, ultrasound examination and study of sexual reflexes, animals were divided into 4 groups: With high reproductive capacity; with good reproductive capacity; with reduced reproductive capacity; and unable to reproduce (Table 5).

Out of the 17 bulls, high and good reproductive ability (I, II) on sperm quality in 11 bulls (65%), the study of the sexual apparatus in 14 bulls (82, 3%) and genital reflexes in 14 bulls (82.3%) were recorded.

Groups with low reproductive capacity and unable to reproduce (III, IV) were included in the semen quality of 6 bulls (35%) and the sexual apparatus and genital reflexes of 3 (17.7%) of the bulls.

The conducted studies allow us to assess the reproductive ability of breeding bulls and to carry out activities with bulls of 3 or 4 groups.

According to the results of laboratory studies, a decrease in sperm quality was noted in some bulls. andrological medical examination of animals by examining the genitals, sexual reflexes and sperm quality. Additionally, in the study of sexual reflexes on a point scale, measurements of morphological development and ultrasound of the testes were included.

Studies indicate a relationship between changes in the shape of the testes and laboratory assessment of sperm quality, in 2 bulls the sperm motility was 5±0.3 points and in 1 bull 8±0.01 points.

Our studies are consistent with the data of (Sammad *et al*., 2020) who determined the relationship between the frequency of changes in the shape of the testicles and andrological aspects in bulls of the Nellore breed. It was found that 99.61% of the animals had an oval testicle shape and were classified as healthy for breeding.

According to an ultrasound examination, 11 bulls with high and good reproductive capacity and a testis structure with moderate echogenicity were recorded. While in the other 6 bulls, with reduced reproductive capacity, 33.3% had

normal testicular echogenicity, despite the low quality of sperm recorded. The results are similar to the data of (Jaskowski *et al*., 2004; Ali *et al*., 2011) who determined that out of 10 bulls, 9 had poor sperm quality, despite this, with ultrasound examination, 4 of the echogenicity of the testes and appendages of the semen was normal. Ultrasound examination makes it possible to determine subclinical changes in the seeds and appendages, thereby increasing the effectiveness of andrological medical examination of bulls.

One of the methods for assessing the reproductive ability of bulls is the study of sexual reflexes by taking into account the time, nature and strength of their manifestation on a 4-point scale. The sexual reflexes of 17 bulls showed that 3 (17.7%) animals had a score below 3 points. These bulls, according to the quality of sperm, belong to group III and restless reproduction.

The results of the studies are consistent with the data of (Carrer Filho *et al*., 2015) who, in their studies, classified breeding bulls by Classification by Points (CAP) and by a 15 m libido test in a pen on a 0-10-point scale into excellent, very good, good and doubtful.

The sperm quality, 11 bulls (65%) are included in groups I and II and 6 bulls (35%) in groups III and IV.

Conclusion

The use of the sexual reflexes on a point scale, the morphological development of the semen and ultrasound examination of the semen and its appendages can improve the quality of evaluation of the reproductive ability of bulls.

Acknowledgment

Express our great gratitude to S. Seifullin Kazakh Agrotechnical University and the Chairman of the Board of JSC of the Republican Center of Breeding Breeding "ASYL TULIK" Seisenov B.S., as well as his team, for facilitating our research and support.

Funding Information

The authors have not received any financial support or funding to report.

Author's Contributions

Gulzat Turysbayeva: Conceived and planned the study. Did statistical analysis of data.

Isatay Jakupov and Aisulu Kuzerbayeva: Conducted lab work and drafted the manuscript.

Zhanargul Zharkimbayeva: Conceived and planned the study.

Ethics

The authors should address any ethical issues that may arise after the publication of this manuscript.

References

Abilov, A., Kolosova E., (2007). Dynamics of Ayrshire bull semen indicators. *Dairy and beef cattle breeding* 2: 23-27.

Ali, K. M., Ahmad, N., Akhtar, N., Ali, S., Ahmad, M., & Younis, M. (2011). Ultrasound Imaging of Testes and Epididymides of Normal and Infertile Breeding Bulls. *Pakistan Veterinary Journal*, *31*(4).

Al-Qarawi, A. A., Abdel-Rahman, H. A., El-Mougy, S. A., & El-Belely, M. S. (2002). Use of a new computerized system for evaluation of spermatozoal motility and velocity characteristics in relation to fertility levels in dromedary bulls. *Animal Reproduction Science*, *74*(1-2), 1-9. https://doi.org/10.1016/S0378-4320(02)00163-X

Andrade, A. K. G., Soares, A. T., Cartaxo, F. Q., Peña-Alfaro, C. E., & Guerra, M. M. P. (2012). Achados ultrassonográficos nos testículos e epidídimos de carneiros deslanados jovens e clinicamente sadios. *Arquivo Brasileiro de Medicina Veterinária e Zootecnia*, *64*, 371-379. https://doi.org/10.1590/S0102-09352012000200017

Bagath, M., Krishnan, G., Devaraj, C., Rashamol, V. P., Pragna, P., Lees, A. M., & Sejian, V. (2019). The impact of heat stress on the immune system in dairy cattle: A review. *Research in Veterinary Science*, *126*, 94-102. https://doi.org/10.1016/j.rvsc.2019.08.011

Barth, A. D. (2018). The use of bull breeding soundness evaluation to identify subfertile and infertile bulls. *Animal*, *12*(s1), s158-s164. https://doi.org/10.1017/S1751731118000538

Bova, T. L., Chiavaccini, L., Cline, G. F., Hart, C. G., Matheny, K., Muth, A. M., ... & Memili, E. (2014). Environmental stressors influencing hormones and systems physiology in cattle. *Reproductive Biology and Endocrinology*, *12*(1), 1-5. https://doi.org/10.1186/1477-7827-12-58

Brown, E. J., & Vosloo, A. (2017). The involvement of the hypothalamopituitary-adrenocortical axis in stress physiology and its significance in the assessment of animal welfare in cattle. *Onderstepoort Journal of Veterinary Research*, *84*(1), 1-9. https://doi.org/10.4102/ojvr.v84i1.1398

Butler, M. L., Bormann, J. M., Weaber, R. L., Grieger, D. M., & Rolf, M. M. (2020). Selection for bull fertility: A review. *Translational Animal Science*, *4*(1), 423-441. https://doi.org/10.1093/tas/txz174

Carrer Filho, L., Koetz, J., Barca, J., Menegassi, S. R. O., Pereira, G. R., dos Santos, M. D., ... & Lopes, F. G. (2015). Andrologic evaluation by points and libido test in corral of young bulls Braford. *Revista Brasileira de Higiene e Sanidade Animal*, *9*(2), 233-243. http://www.higieneanimal.ufc.br/.../867

Chen, Y., Arsenault, R., Napper, S., & Griebel, P. (2015). Models and methods to investigate acute stress responses in cattle. *Animals*, *5*(4), 1268-1295. https://doi.org/10.3390/ani5040411.

Collantes-Fernández, E., Moreno-Gonzalo, J., Sánchez-Sánchez, R., García-Bocanegra, I., Horcajo, P., & Ortega-Mora, L. M. (2019). Prevalence of bovine trichomonosis and associated risk factors in bulls from Spanish beef herds. *Theriogenology*, *128*, 116-121. https://doi.org/10.1016/j.theriogenology.2019.01.030

Collier, R. J., & Gebremedhin, K. G. (2015). Thermal biology of domestic animals. *Annu. Rev. Anim. Biosci*, *3*(1), 513-532. https://doi.org/10.1146/annurev-animal-022114-110659

Collier, R. J., Renquist, B. J., & Xiao, Y. (2017). A 100-Year Review: Stress physiology including heat stress. *Journal of Dairy Science*, *100*(12), 10367-10380. https://doi.org/10.3168/jds.2017-13676

Cooke, R. F., Moriel, P., Cappellozza, B. I., Miranda, V. F. B., Batista, L. F. D., Colombo, E. A., ... & Vasconcelos, J. L. M. (2019). Effects of temperament on growth, plasma cortisol concentrations and puberty attainment in Nelore beef heifers. *Animal*, *13*(6), 1208-1213 https://doi.org/10.1017/s1751731118002628

Dahl, G. E., Tao, S., & Laporta, J. (2020). Heat stress impacts immune status in cows across the life cycle. *Frontiers in Veterinary Science*, *7*, 116. https://doi.org/10.3389/fvets.2020.00116

Fernandez-Novo, A., Fargas, O., Loste, J. M., Sebastian, F., Perez-Villalobos, N., Pesantez-Pacheco, J. L., ... & Astiz, S. (2020). Pregnancy Loss (28-110 Days of Pregnancy) in Holstein Cows: A Retrospective Study. *Animals*, *10*(6), 925 https://doi.org/10.3390/ani10060925

Heimbürge, S., Kanitz, E., Tuchscherer, A., & Otten, W. (2020). Within a hair's breadth-Factors influencing hair cortisol levels in pigs and cattle. *General and Comparative Endocrinology*, *288*, 113359 https://doi.org/10.1016/j.ygcen.2019.113359

Hughes, H. D., Carroll, J. A., Sanchez, N. C. B., & Richeson, J. T. (2014). Natural variations in the stress and acute phase responses of cattle. *Innate Immunity*, *20*(8), 888-896. https://doi.org/10.1177/1753425913508993

Jaskowski, J. M., Urbaniak, K., Antosik, P., Galant, K., Zbylut, J., & Bukowska, D. (2004). Ultrasonographic evaluation of reproductive traits in young bulls. *Medycyna Weterynaryjna*, *60*(9), 994-997.

Jedraszczyk, J. (2003). Ultrasonography technique in bull reproductive organ examination. *Medycyna Weterynaryjna*, *59*(4), 311-314

Johnston, D. J., Barwick, S. A., Fordyce, G., Holroyd, R. G., Williams, P. J., Corbet, N. J., & Grant, T. (2013). Genetics of early and lifetime annual reproductive performance in cows of two tropical beef genotypes in northern Australia. *Animal Production Science*, *54*(1), 1-15. https://doi.org/ 10.1071/AN13043

Kasimanickam, R., Asay, M., Schroeder, S., Kasimanickam, V., Gay, J. M., Kastelic, J. P., ... & Whittier, W. D. (2014). Calm temperament improves reproductive performance of beef cows. *Reproduction in Domestic Animals*, *49*(6), 1063-1067. https://doi.org/10.1111/rda.12436

Kolchina, A. F., & Barashkin, M. I. (2011). Andrological medical examination of breeding bulls method instructions, Ural State Agricultural. *Academy*, p.24.

Kapš, M., Posavi, M., Stipić, N., & Mikulić, B. (2000). Genetic evaluation of semen and growth traits of young Simmental bulls in performance test. Agriculturae Conspectus Scientificus, 65(1), 15-20. https://hrcak.srce.hr/12507

Lauder, J. K., Marti, S., Schwartzkopf-Genswein, K. S., Jelinski, M. D., & Janzen, E. D. (2020). Measuring behavioral and physiological responses to pain mitigation for ovariectomy in Bos taurus yearling beef heifers. *Journal of Animal Science*, *98*(1), skz386. https://doi.org/10.1093/jas/skz386

Lees, A. M., Sejian, V., Wallage, A. L., Steel, C. C., Mader, T. L., Lees, J. C., & Gaughan, J. B. (2019). The impact of heat load on cattle. *Animals*, *9*(6), 322. https://doi.org/10.3390/ani9060322

Mazer, K. A., Knickerbocker, P. L., Kutina, K. L., & Huzzey, J. M. (2020). Changes in behavior and fecal cortisol metabolites when dairy cattle are regrouped in pairs versus individually after calving. *Journal of Dairy Science*, *103*(5), 4681-4690. https://doi.org/10.3168/jds.2019-17593

Medvedev, G. F. (2006). Physiology and pathology of the reproductive system of cattle: Monograph-Gorki: Belarusian State Agricultural Academy, p.216.

Mee, J. F., Geraghty, T., O'Neill, R., & More, S. J. (2012). Bioexclusion of diseases from dairy and beef farms: Risks of introducing infectious agents and risk reduction strategies. *The Veterinary Journal*, *194*(2), 143-150. https://doi.org/10.1016/j.tvjl.2012.07.001

Michael, J. D., Baruselli, P. S., & Campanile, G. (2019). Influence of nutrition, body condition, and metabolic status on reproduction in female beef cattle: A review. *Theriogenology*, *125*, 277-284. https://doi.org/10.1016/j.theriogenology.2018.11.010

Michi, A. N., Favetto, P. H., Kastelic, J., & Cobo, E. R. (2016). A review of sexually transmitted bovine trichomoniasis and campylobacteriosis affecting cattle reproductive health. *Theriogenology*, *85*(5), 781-791. https://doi.org/10.1016/j.theriogenology.2015.10.037

Newcomer, B. W., Cofield, L. G., Walz, P. H., & Givens, M. D. (2017). Prevention of abortion in cattle following vaccination against bovine herpesvirus 1: A meta-analysis. *Preventive Veterinary Medicine*, *138*, 1-8. https://doi.org/10.1017/S1466252319000057

Olson, C. A., Carstens, G. E., Herring, A. D., Hale, D. S., Kayser, W. C., & Miller, R. K. (2019). Effects of temperament at feedlot arrival and breed type on growth efficiency, feeding behavior, and carcass value in finishing heifers. *Journal of Animal Science*, *97*(4), 1828-1839. https://doi.org/10.1093/jas/skz029

Orihuela, A., & Galina, C. S. (2019). Effects of separation of cows and calves on reproductive performance and animal welfare in tropical beef cattle. *Animals*, *9*(5), 223. https://doi.org/10.3390/ani9050223

Oguejiofor, C. F., Thomas, C., Cheng, Z., & Wathes, D. C. (2019). Mechanisms linking Bovine Viral Diarrhea Virus (BVDV) infection with infertility in cattle. *Animal Health Research Reviews*, *20*(1), 72-85. https://doi.org/10.1017/S1466252319000057

Rioja-Lang, F. C., Connor, M., Bacon, H. J., Lawrence, A. B., & Dwyer, C. M. (2020). Prioritization of farm animal welfare issues using expert consensus. *Frontiers in Veterinary Science*, *6*, 495. https://doi.org/10.3389/fvets.2019.00495

Sabés-Alsina, M., Lundeheim, N., Johannisson, A., López-Béjar, M., & Morrell, J. M. (2019). Relationships between climate and sperm quality in dairy bull semen: A retrospective analysis. *Journal of Dairy Science*, *102*(6), 5623-5633. https://doi.org/10.3168/jds.2018-15837

Sammad, A., Wang, Y. J., Umer, S., Lirong, H., Khan, I., Khan, A.,... & Wang, Y. (2020). Nutritional physiology and biochemistry of dairy cattle under the influence of heat stress: Consequences and opportunities. *Animals*, *10*(5), 793. https://doi.org/10.3390/ani10050793

Tokysheva, G., Makangali, K., Uzakov, Y., Kakimov, M., Vostrikova, N., Baiysbayeva, M., & Mashanova, N. (2022). The potential of goat meat as a nutrition source for schoolchildren. *In Potravinarstvo Slovak Journal of Food Sciences* 16, pp. 398-410. https://doi.org/10.5219/1763

Tokysheva, G., Tultabayeva, T., Mukhtarkhanova, R., Zhakupova, G., Gorbulya, V., Kakimov, M., & Makangali, K. (2023). The study of physicochemical and technological properties of boiled sausage recommended for the older adults. *Potravinarstvo Slovak Journal of Food Sciences*, 17, 16-29. https://doi.org/10.5219/1806

Tschoner, T. S., Zablotski, Y., Knubben-Schweizer, G., & Feist, M. (2020). Effect of xylazine administration before laparoscopic abomasopexy to correct left displaced abomasum on markers of stress in dairy cows. *Journal of Dairy Science*, *103*(10), 9318-9331. https://doi.org/10.3168/jds.2020-18523

Yu, H., Morota, G., Celestino Jr, E. F., Dahlen, C. R., Wagner, S. A., Riley, D. G., & Hulsman Hanna, L. L. (2020). Deciphering cattle temperament measures derived from a four-platform standing scale using genetic factor analytic modeling. *Frontiers in Genetics*, *11*, 599. https://doi.org/10.3389/fgene.2020.00599