International Journal of Veterinary Sciences and Animal Husbandry

ISSN: 2456-2912
VET 2024; 9(1): 1166-1168
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www.veterinarypaper.com
Received: 25-11-2023
Accepted: 29-12-2023

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Gross anatomical and biometrical study of cervix during different phases of estrus cycle in buffalo (*Bubalusbubalis*)

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Abstract
An anatomical and biometrical study was conducted on the cervix of 50 buffaloes across different stages of the estrus cycle. As the animals approached estrus, there was a noticeable increase in uterine tone, reaching its peak during estrus. Following estrus, there was a gradual reduction in uterine tone. The average length of the cervix during proestrus, estrus, metestrus, diestrus, and anestrus was recorded as 6.72±0.26, 6.89±0.17, 6.65±0.35, 7.04±0.28, and 7.00±0.19 cm, respectively. Concurrently, the average circumference of the cervix during these phases was measured as 16.45±0.25, 17.50±0.27, 17.24±0.24, 17.13±0.37, and 16.05±0.41 cm, respectively. The average number of cervical rings during proestrus, estrus, metestrus, diestrus, and anestrus was documented as 3.60±0.22, 3.90±0.23, 3.20±0.24, 3.40±0.30, and 3.00±0.25, respectively. These findings provide valuable insights into the variations in cervix dimensions and characteristics throughout the different phases of the estrus cycle in buffaloes.

Keywords: Cervix, biometry, proestrus, estrus, metestrus, diestrus and anestrus

Introduction
The uterus holds a pivotal role in the reproductive tract, undergoing discernible transformations throughout the four phases of the estrus cycle: proestrus, estrus, metestrus, and diestrus. These cyclic alterations are orchestrated by the impact of pituitary hormones on the ovary. The configuration and functions of the uterus exhibit variations corresponding to reproductive status, as highlighted by Deshpande (1994) [4], depicting the dynamic interplay of pituitary-ovarian activity. A comprehensive understanding of the uterus's structure and function during different stages of the estrus cycle is imperative (Shukla *et al*., 1973) [21]. However, in the context of buffaloes, there is a notable scarcity of comprehensive information pertaining to the biometrical changes occurring in sexual organs (Khan, 1989) [15]. Although reports on biometrical studies of female genitalia in Murrah, Jaffri, and Nili Ravi buffaloes are available, limited information is accessible concerning non-descript buffaloes (Napolean and Quayam, 1996) [18].

Material and Methods
The research involved 50 buffaloes, categorized into five groups corresponding to different phases of the estrus cycle, namely, proestrus, estrus, metestrus, diestrus, and anestrus, each group comprising ten samples. To prepare the samples, they were meticulously cleaned under running tap water to eliminate tissue debris, fascia, and blood clots. Various gross anatomical observations and biometric parameters, including uterus weight (in grams), length, maximum circumference, and maximum diameter of the cervix, were measured in centimeters. This was done using a weighing balance, non-stretchable thread, scale, and vernier caliper. The number of cervical rings was also recorded. The collected data underwent statistical analysis following the methodology outlined by Snedecor and Cochran (1967), and the ensuing results are discussed below.

Result and Discussion

The cervix, recognized as the posterior constricted segment of the uterus, establishes anterior communication with the uterus body and posterior opening into the vagina. These observations correspond with the descriptions outlined by Hafez (1980) [13], Frandson *et al*. (1994) [10], and Dyce *et al*. (2002) [7] in cows. Moreover, the current study reaffirms the identification of the cervix as a sphincter-like structure, a characterization previously documented by Hafez (1980) [13] and Dyce *et al*. (2002) [7] in cows.

The lumen of the cervix exhibited a constricted appearance, filled with mucosal folds, and the cervix's wall was recognized as the thickest part of the uterus. Comparable findings were documented by Raghavan (1964) [19], Hafez (1980) [13], Frandson *et al*. (1994) [10], Dyce *et al*. (2002) [7], and Ghosh (2006) [12] in cows. It is noteworthy that Hafez (1980) [13] and Frandson *et al*. (1994) [10] specifically described the mucosal folds as annular rings arranged in spirally transverse interlocking ridges in cows.

The cervix concluded with the os uteri, a diminutive opening situated on a rounded prominence at the anterior extremity of the vagina. This observation aligns with the findings of Asdell (2002) [1] and Ghosh (2006) [12] in cows, although Ghosh (2006) [12] specifically referred to the depression on the projected part of the cervix into the vagina as the external os. Notably, Deshpande and Velhankar (1994) [4] previously highlighted that the buffalo's uterus is relatively well-defined and large, possessing a smaller body and a narrower, shorter cervix compared to cows. They also reported a bluish-grey color in the buffalo's uterus. In contrast, the current study observed a pale to pink color in the uterus, suggesting potential variations attributable to breed differences.

The uterus underwent discernible changes across the estrus cycle. Several days preceding estrus, there was a noticeable rise in uterine tone, intensifying at the onset of estrus. This phase was marked by turgidity, erectness, and coiling upon palpation. Subsequent to estrus, the uterus reverted to a soft and flaccid state during the metestrus and diestrus phases. These findings align with observations made by Laing *et al*. (1988) [17] in cows and Franz and Wright (2001) [9] in domestic animals.

Length of the cervix (cm)

The length of the cervix during different phases of the estrus cycle exhibited a range of values: 5.80 to 8.00 cm in proestrus, 6.12 to 8.00 cm in estrus, 5.50 to 8.50 cm in metestrus, 5.30 to 8.00 cm in diestrus, and 6.00 to 8.00 cm in anestrus. The corresponding average lengths were 6.72±0.26 cm, 6.89±0.17 cm, 6.65±0.35 cm, 7.04±0.28 cm, and 7.00±0.19 cm, respectively. Notably, these differences in average values were statistically non-significant. Comparable findings were reported by Kaikini (1974) [14] in buffaloes, Laing *et al*. (1988) [17] in cows, Khan (1989) [15] in pregnant buffaloes, Napolean and Quayam (1996) [18] in non-descript buffaloes, and Ali *et al*. (2006) [2] in cows.

However, variations in cervix lengths were noted in studies conducted by Drennan and Macpherson (1966) [6] in heifers, El-Sheikh and Abdelhadi (1970) [8] in Egyptian buffaloes, Kodagali *et al*. (1971) [16] in Surti buffaloes, Getty (1975) [11] in cattle, Hafez (1980) [13] in cows, Khan (1989) [15] in Nili Ravi buffaloes, Chauhan and Adamu (1990) [3] in African Zebu cattle, Franz and Wright (2001) [9] in cows, Asdell (2002) [1] in cows, and Ghosh (2006) [12] in cattle. The reported cervix lengths in these studies ranged from 4.77 to 10 cm. Such variations may be attributed to differences in species, size, breed, age, and reproductive status. Sane *et al*. (1964) and (1965) recorded cervix lengths of 7.69 and 8.09±0.13 cm in Murrah and Jaffrabadi buffaloes, respectively, indicating potential breed-related differences.

Maximum circumference of the cervix (cm)

The maximum circumference of the cervix exhibited variability during different phases of the estrus cycle: ranging from 15.16 to 18.30 cm in proestrus, 16.00 to 19.00 cm in estrus, 16.00 to 18.33 cm in metestrus, 15.30 to 19.00 cm in diestrus, and 14.50 to 19.00 cm in anestrus. The corresponding average circumferences were 16.45±0.25 cm, 17.50±0.27 cm, 13.61±0.65 cm, 17.93±0.37 cm, and 16.05±0.41 cm, respectively. The differences between the average values were deemed statistically significant at a 5% level. Moreover, the means with at least one common superscript did not differ significantly, while the maximum circumference of the cervix with no common superscript exhibited significant differences. In contrast, Ali *et al*. (2006) [2] reported an average cervix circumference of 8.40±0.21 cm in non-descript cows, suggesting potential variations related to the species.

Maximum diameter of the cervix (cm)

The maximum diameter of the cervix displayed variation across different phases of the estrus cycle: ranging from 3.71 to 5.12 cm in proestrus, 4.11 to 5.10 cm in estrus, 3.70 to 6.00 cm in metestrus, 4.00 to 5.20 cm in diestrus, and 3.50 to 6.00 cm in anestrus. The respective average diameters were 4.47±0.14 cm, 4.42±0.09 cm, 4.61±0.23 cm, 4.77±0.13 cm, and 4.71±0.23 cm. Importantly, the differences between the average values were statistically non-significant. These findings align with observations made by Chauhan and Adamu (1990) [3] in African Zebu cattle and Asdell (2002) [1] in cows. However, notable differences were noted in studies by Hafez (1980) [13] and Franz and Wright (2001) [9], where cervix diameters in cows ranged from 3 to 4 cm and 5 cm, respectively, indicating potential species-related variations.

Number of cervical rings

The count of cervical rings during proestrus, estrus, metestrus, diestrus, and anestrus displayed variability, ranging from 3 to 5, 3 to 5, 2 to 4, 2 to 5, and 2 to 4, respectively. The corresponding averages were 3.60±0.22, 3.90±0.23, 3.20±0.24, 3.40±0.30, and 3.00±0.25, and importantly, the differences between these average values were statistically non-significant. These findings align with similar observations reported by Kodagali *et al*. (1971) [16] and Kaikini (1974) [14] in Surti and Nagpuri buffalo, Hafez (1980) [13], and Napolean and Quayam (1996) [18] in buffalo, as well as Asdell (2002) [1] in cows. However, contrasting results were noted in studies conducted by Sane *et al*. (1964) [20] in Murrah buffalo, Drennan and Macpherson (1966) [6] in cows, Khan (1989) [15] in Nili Ravi buffalo, and Ali *et al*. (2006) [2] in cows, where the reported number of cervical rings was 3, 4.03, 4.46±0.03, and 6, respectively. Importantly, the number of cervical rings was not found to be influenced by pregnancy.

Table 1: Range of the biometrical observations of various parameters of the cervix of buffalo (*Bubalusbubalis*) in different phases of estrus cycle

Sr. No.	Parameters	Proestrus	Estrus	Metestrus	Diestrus	Anestrus
1.	Weight of uterus (gm)	500.00 - 750.00	550.00 - 800.00	500.00 - 750.00	500.00 - 850.00	500.00 - 800.00
2.	Number of cervical rings	3.00 - 5.00	3.00 - 5.00	2.00 - 4.00	2.00 - 5.00	2.00 - 4.00
3.	Length of the cervix of uterus (cm)	5.80 - 8.00	6.12 - 8.00	5.50 - 8.50	5.30 - 8.00	6.00 - 8.00
4.	Circumference of the cervix of uterus (cm)	15.16 - 18.30	16.00 - 19.00	16.00 - 18.33	15.30 - 19.00	14.50 - 19.00
5.	Diameter of the cervix of uterus (cm)	3.71 - 5.12	4.11 - 5.10	3.70 - 6.00	4.00 - 5.20	3.50 - 6.00

Table 2: Statistical analysis of the biometrical observations of various parameters of the cervix of buffalo (*Bubalusbubalis)* in different phases of estrus cycle

Sr. No.	Parameter	Mean±SE					Coefficient of Variation	F value
		Proestrus	Estrus	Metestrus	Diestrus	Anestrus		
1.	Weight of uterus (gm)	635.00^{b}± 25.87	650.00^{b}±25.82	625.00^{b}±26.08	655.00^{b}±39.75	645.00^{b}±30.23	14.00	2.839*
2.	Number of cervical rings	3.60±0.22	3.90±0.23	3.20±0.24	3.40±0.30	3.00±0.25	21.62	1.843^{NS}
3.	Length of the cervix of uterus (cm)	6.72±0.26	6.89±0.17	6.65±0.35	7.04±0.28	7.00±0.19	12.08	1.614^{NS}
4.	Circumference of the cervix of uterus (cm)	16.45^{abc}±0.25	17.50^{a}±0.27	17.24^{a}±0.24	17.13^{ab}±0.37	16.05^{bc}±0.41	7.48	2.837*
5.	Diameter of the cervix of uterus (cm)	4.47±0.14	4.42±0.09	4.61±0.23	4.77±0.13	4.71±0.23	13.64	2.31^{NS}

:** ($p \leq 0.05$), *:** ($p \leq 0.01$) and NS**:** Non-significant.

Note: Mean with at least one common superscript do not differ significantly.

Summary and Conclusion

The cervix, functioning as the posterior sphincter-like segment of the uterus, displayed a constricted lumen featuring annular rings arranged in spirally transverse interlocking ridges. Its culmination was marked by the os uteri, a small opening on a rounded prominence at the anterior extremity of the vagina. Throughout proestrus and estrus, there was an observable increase in uterine tone, characterized by a turgid, erect, and coiled state. In metestrus, diestrus, and anestrus, the uterus reverted to a soft and flaccid condition. All recorded average values for the biometrical observations in the study were found to be statistically non-significant, except for the weight of the uterus and the maximum circumference of the cervix, which exhibited statistically significant differences at a 5% level of significance.

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