The prenatal development in swamp buffalo (*Bubalus carabanensis*)

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Summary

There are morphological and reproductive physiological differences between swamp buffalo (*Bubalus carabanensis*) and river buffalo (*Bubalus bubalis*). The development of fetus weight and fetus biometry was reported in river buffalo and other animals but not in swamp buffalo. The aim of this study was to describe the inherent variability in fetus related measurements during swamp buffalo pregnancy. The data is based on measurements of 267 fetuses and 5 new born claves from swamp buffalo. The results show that a significant linear correlation exists between estimated age of fetuses and parameters of fetus sizes. There were correlations between crown-rump length (CRL) and other fetal parameters, as well as between fetus weight and its parameters. In conclusion, our data indicated that the feasibility and value of fetal measures in swamp buffaloes being used for the evaluation of fetal development.

Key words: Swamp buffalo, Prenatal development, Fetus, Pregnancy

Introduction

Buffaloes are an important domestic animal in Asia. According to the recent FAO statistics, the buffalo (*Bubalus bubalis*) population in the world is presently about 168.7 million, among them 161.9 million are located in Asia (96.0%) (Cruz, 2010). The domestic water buffaloes (*Bubalus bubalis*) in Asia include the river and swamp types. They differ in morphology, genetics and behavior. The name swamp buffalo is commonly used to refer to all the native water buffaloes in southeast Asia and some in the south of China (Borghese and Mazzi, 2005). The term river buffalo refers to all types of buffaloes distributed across India and other countries (Borghese and Mazzi, 2005). Morphologically, swamp buffaloes are much closer to the wild type than river buffaloes and are mainly used as draft animals and for meat production. Genetically, the river buffalo has 50 chromosomes while the swamp buffalo has 48 (Borghese and Mazzi, 2005). Swamp buffalo play an important role in total agriculture production and economic income of small farm holders.

Slaughterhouse studies of river buffalo fetuses from Egypt and India indicated that all body parameters increase steadily in size during gestation. These studies concluded that the CRL is the most satisfactory criterion for estimating the age of the river buffalo fetuses (Singh *et al*., 1963; Abdel-Raouf and El-Naggar, 1968; Abdel-Raouf and El-Naggar, 1970). The prenatal development of river buffalo and the correlation between weight and age of fetus and other parameters have been reported (Singh *et al*., 1963). There are also differences in reproductive physiology between swamp and river buffalo, most obviously reflected in mean gestation lengths: 330 days in swamp buffalo and 310 days in river buffalo. Furthermore, the calf birth weight of swamp buffalo is less than that in river buffalo (29 and 35 kg,

respectively) (Barile, 2005).

Prenatal development of laboratory, domestic and many wild mammals has been reported, however, to our knowledge, there have been no reports concerning prenatal development in swamp buffalo. In the present study, the correlation between CRL and weight with different fetal parameters, such as: nose-rump length, head length, body cross length in swamp buffalo during gestation is reported.

Materials and Methods

Fetus of pregnant swamp buffalo (n=267 thereof 148 males and 119 females) were collected from slaughterhouses in the suburb of Hanoi, Vietnam. The new born calves (n=5: 2 males and 3 females) were collected from a farm in Thainguyen province, Vietnam.

Measurement of concepts and new born calve parameters

The parameters were measured following Joubert (1956) (Fig. 1): 1) CVRL was taken with flexible steel tape across the dorsum from the fore head (midway between the eyes) (A) to the tail head (B); 2) Nose-rump length was taken with flexible steel tape across the dorsum from the nose (C) to the tail head (B); 3) Eye-rump length straight was measured from the forehead at point (A) to the tail head at point (B); 4) Ear-rump length was taken from the top head (midway between the ear) (D) to the tail head (B); 5) Head length was measured from top of the head (D) to the nose (C) (midway between the ears); 6) Head width was the horizontal distance between the points just in front of the ears; 7) Face width was measured by caliper-square between two eyes; 8) Head circumference was measured at the biggest place of the head (at the height of the ears); 9) Body cross length was measured from the tuber spine scapulae (E) to the head tail (B); 10) Chest depth was taken vertical to the line of the 6th rib (immediately behind the thoracic limbs) from dorsal surface of the back (G) to the ventral border of the sternum (H); 11) Chest circumference was measured with a measuring tape parallel to the 6th rib, point (I) to (J); 12) Radius-Ulna was taken from F to F1; 13) Metacarpal was taken from F1 to F2; 14) Tibia was taken from H to H1; 15) Tarsal-Metatarsal was taken from H1 to H2; 16) Umbilical cord circumference was measured at the biggest place by measuring tape.

The weight of fetuses was also recorded by scale with accuracy of 1 g in early fetus.
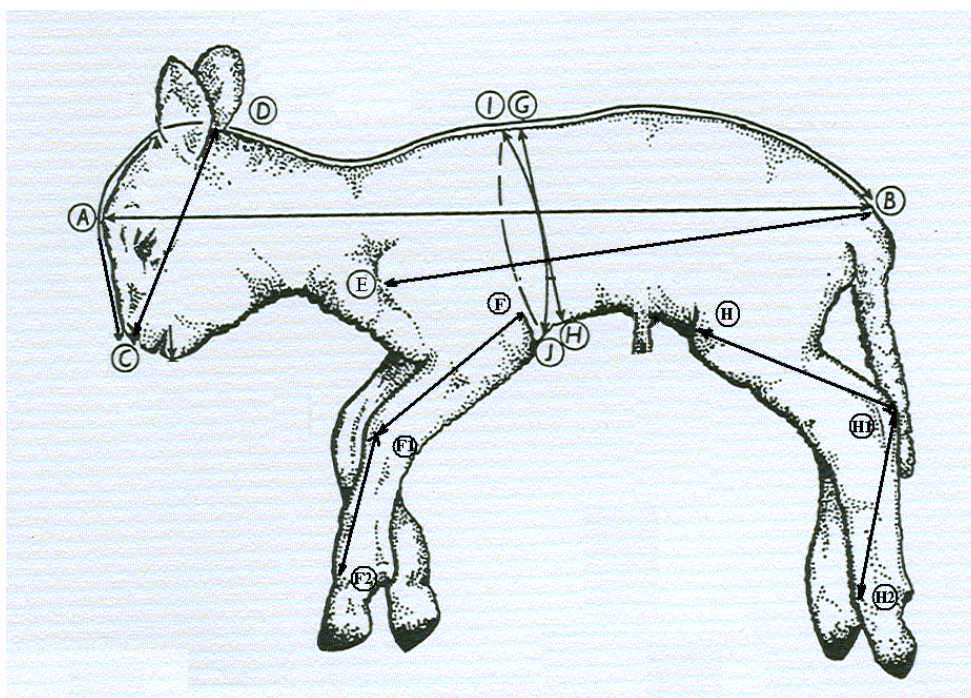

Fig. 1: Positions of fetal measurements (Joubert, 1956)

Determination of fetal age and sex

During gestation, the approximate fetal age was estimated using the CRL as previously described by Singh *et al*. (1963). The fetal age estimation tools were determined as overall polynomial regression function:

$y = 0.0011x^3 - 0.1411x^2 + 8.5715x + 17.9$ (r = 0.999)

where,
y: CRL (cm)
x: The estimated fetal age (days) (Ali and Fahmy, 2008)

Depending on the stage of fetal development, the sex of the fetus was determined by examination of the external genitalia. The area from the umbilical cord to the tail was scrutinized to identify the apparent genital tubercle, while the area between the two hind limbs was examined for the appearance of the scrotum or the udder. The fetus was recorded as male when the genital tubercle was located immediately caudal to the abdominal attachment of the umbilical cord or by presence of scrotum between the two hind limbs, while the fetus was recorded as female when the genital tubercle was located toward the base of the tail or by presence of the udder between the two thighs (Ali and Fahmy, 2008).

Statistical analysis

The data was performed using Microsoft Office Excel. The correlation models were fitted to evaluate the relationship between gestational age and each of the studied parameters. The line of best fit using the most appropriate regression function is presented.

Results

The measurements of 17 parameters of swamp buffalo fetus were shown in Table 1. The average values for different fetal measurements were shown at monthly intervals. The fetuses were distributed mainly from days 91 to 240 (125 fetuses, corresponding to 46.8% of total).

Correlation between CRL and measurement of other parameters

The correlation between CRL and measurements of different parts of the fetal body is shown in Table 2. During gestation, the relations are best described by the 1st order function, also shown in Table 2. The correlation of coefficients significance was very high and no differences were demonstrated between male and female fetuses (P>0.05). The lines of best fitting through the combined data points estimated from the fetal measurements during gestation are expressed as a 1st regression function as follows:

$y = ax + b$

where,
x: CRL (cm)
y: The value of body fetus measurements (cm)

The results of correlation between CRL and four distinct fetal head measurements are presented in Table 3. In general, the correlations were best fitted with 2nd level function. Highest correlations (male, female and total) were observed regarding CRL and head circumference, while the lowest correlation was observed between CRL and male face width.

The relative contributions of CRL towards the total measurement of fetal leg measurements during gestation are shown in Table 4. The data presentation indicates that the measurement of fetal leg and CRL have relative best fit by using a power function.

The results obtained after calculating the correlation between CRL and umbilical cord circumferences are shown in Table 5. In early pregnancy period the regression between CRL and umbilical cord circumference of male and female was considered, whereas a clear difference appeared in late pregnancy.

Correlation between fetal weight and measurement of other parameters

The correlations between fetal weight and fetal length are shown in Table 6. A power correlation exists between fetal length and weight of fetus as follows:

$y = ax^b$

in which,
y: Fetal length
x: Fetal weight

Based on 267 observations, the correlation between CRL and weights was

Table 1: Average (Mean±SD) of fetal measurements of the developing swamp buffalo fetuses during the successive 30 day periods of gestation

No.	Parameters	N.	Period of pregnancy (days)										
			30-60	61-90	91-120	121-150	151-180	181-210	211-240	241-270	271-300	301-330	New born
			7	23	34	28	38	47	40	26	14	10	5
1	CRL (cm)		5.37 ±2.90	10.25 ±1.46	14.93 ±2.65	22.75 ±2.08	30.81 ±3.67	43.14 ±4.48	54.50 ±4.73	65.72 ±4.35	74.79 ±5.45	85.5 ±5.97	86.8 ±5.67
2	Noise-rump length (cm)		6.04 ±3.04	11.20 ±1.59	17.10 ±2.26	26.10 ±2.76	35.53 ±3.94	49.30 ±4.43	61.40 ±4.99	74.27 ±5.40	83.21 ±5.68	95.90 ±6.03	98.2 ±5.07
3	Eye-rump length straight (cm)		3.77 ±1.46	8.03 ±1.23	12.81 ±1.45	19.61 ±1.57	26.67 ±2.71	37.55 ±3.21	47.69 ±3.40	57.54 ±2.31	65.64 ±2.02	75.20 ±4.96	79.0 ±3.65
4	Ear rump length (cm)		3.3 ±1.67	6.6 ±0.93	10.34 ±1.46	15.99 ±1.62	22.74 ±2.71	32.72 ±3.96	41.18 ±4.12	52.35 ±4.91	58.71 ±4.58	68.4 ±5.85	69.8 ±5.97
5	Body cross length (cm)		2.73 ±0.38	4.57 ±0.88	7.78 ±1.20	12.5 ±1.15	17.32 ±1.85	25.00 ±3.96	31.70 ±3.17	39.69 ±4.35	44.14 ±3.16	52.80 ±3.77	55.4 ±4.01
6	Chest circumference (cm)		4.15 ±0.58	6.01 ±1.02	10.07 ±1.89	15.01 ±1.53	20.67 ±2.08	28.80 ±2.87	36.85 ±3.49	43.00 ±8.57	48.57 ±4.03	58.00 ±3.71	71.4 ±3.87
7	Chest depth (cm)		1.78 ±0.38	2.33 ±0.40	3.99 ±0.67	6.35 ±0.74	8.70 ±1.29	12.51 ±1.48	16.15 ±1.76	19.42 ±1.17	21.79 ±1.42	24.85 ±1.45	31.0 ±1.64
8	Head length (cm)		1.95 ±0.17	2.94 ±0.58	4.93 ±0.91	7.83 ±0.79	10.48 ±1.41	14.10 ±1.07	17.40 ±2.03	20.23 ±1.44	22.36 ±2.90	23.80 ±1.11	25.6 ±1.43
9	Head width (cm)		1.03 ±0.13	1.68 ±0.31	2.58 ±0.49	4.16 ±0.48	5.05 ±0.84	6.83 ±0.72	8.37 ±1.18	9.69 ±1.33	10.50 ±1.16	11.86 ±1.04	12.3 ±1.25
10	Head circumference (cm)		4.35 ±0.66	6.60 ±0.91	10.07 ±1.90	14.54 ±1.06	19.02 ±1.78	24.63 ±2.55	30.63 ±2.26	36.06 ±1.80	39.46 ±2.89	43.25 ±2.46	47.4 ±2.19
11	Face width (cm)		0.55 ±0.06	0.89 ±0.26	1.48 ±0.47	2.59 ±0.55	3.50 ±0.56	4.48 ±0.59	5.40 ±0.77	6.40 ±0.91	6.89 ±0.71	7.75 ±0.63	8.0 ±0.81
12	Radius-Ulna (cm)		0.40 ±0.10	0.79 ±0.26	1.41 ±0.26	2.36 ±0.37	3.27 ±0.61	5.04 ±0.87	7.05 ±1.34	10.37 ±2.27	11.07 ±1.33	13.70 ±1.84	14.0 ±1.96
13	Metacarpal (cm)		0.77 ±0.12	1.29 ±0.34	2.78 ±0.41	3.72 ±0.35	5.46 ±0.67	8.19 ±1.04	11.04 ±1.46	14.46 ±1.47	17.00 ±1.57	20.35 ±1.42	21.6 ±1.86
14	Tibia (cm)		0.57 ±0.15	1.10 ±0.31	2.15 ±0.45	3.70 ±0.51	5.27 ±0.86	8.22 ±1.13	11.56 ±1.67	15.82 ±1.74	18.86 ±2.01	22.60 ±1.63	25.8 ±1.78
15	Tarsal-Metatarsal (cm)		1 ±0.10	1.57 ±0.39	2.79 ±0.54	4.88 ±0.55	6.84 ±0.90	10.80 ±1.37	14.32 ±1.92	19.35 ±1.78	22.64 ±2.44	26.90 ±2.81	28.8 ±2.63
16	Umbilical cord circumference (cm)		1 ±0.18	1.24 ±0.39	1.99 ±0.46	3.01 ±0.47	3.96 ±0.58	5.18 ±0.69	6.22 ±0.86	6.70 ±0.95	7.00 ±0.68	7.35 ±1.23	
17	Weight (kg)		0.007 ±0.01	0.037 ±0.05	0.097 ±0.06	0.326 ±0.11	0.893 ±0.36	2.479 ±0.73	4.95 ±1.16	9.037 ±1.79	12.27 ±2.80	18.87 ±3.54	25.8 ±4.06

Table 2: Correlation between CRL (y) and body measurements (x)

Parameter	Sex	Regression	R^2
Noise rump length	Total	y = 1.2679x + 1.3352	0.988
	Males	y = 1.2702x + 1.3533	0.989
	Females	y = 1.2642x + 1.3321	0.987
Eye rump length straight	Total	y = 1.1265x + 0.8702	0.985
	Males	y = 1.1378x + 0.6674	0.985
	Females	y = 1.1116x + 1.1497	0.984
Ear rump length	Total	y = 0.9123x - 1.4152	0.983
	Males	y = 0.9223x - 1.6617	0.983
	Females	y = 0.899x - 1.0892	0.983
Body cross length	Total	y = 0.698x - 1.2174	0.991
	Males	y = 0.7092x - 1.4788	0.992
	Females	y = 0.6834x - 0.8783	0.991
Chest circumference	Total	y = 0.7612x + 0.2591	0.982
	Males	y = 0.7743x - 0.0533	0.986
	Females	y = 0.7442x + 0.658	0.978
Chest depth	Total	y = 0.3399x - 0.2845	0.977
	Males	y = 0.3452x - 0.371	0.979
	Females	y = 0.3328x - 0.1665	0.976

Table 3: Correlation between four fetal head measures (x) and CRL (y) and weight (y)

Parameter	Sex	CRL		Weight	
		Regression	R^2	Regression	R^2
Head length	Total	$y = -0.0019x^2 + 0.465x - 0.603$	0.973	$y = 19.599x^{0.2748}$	0.963
	Males	$y = -0.0018x^2 + 0.4611x - 0.4643$	0.973	$y = 10.622x^{0.3077}$	0.969
	Females	$y = -0.0019x^2 + 0.4704x - 0.781$	0.973	$y = 10.473x^{0.3064}$	0.932
Head circumference	Total	$y = -0.0021x^2 + 0.7173x + 1.1838$	0.982	$y = 10.533x^{0.3084}$	0.953
	Males	$y = -0.002x^2 + 0.7166x + 1.1162$	0.985	$y = 19.678x^{0.2755}$	0.982
	Females	$y = -0.0021x^2 + 0.7146x + 1.3103$	0.977	$y = 19.606x^{0.2707}$	0.944
Head width	Total	$y = -0.0007x^2 + 0.2031x + 0.1566$	0.941	$y = 5.2472x^{0.2859}$	0.942
	Males	$y = -0.0005x^2 + 0.1972x + 0.2546$	0.944	$y = 5.3192x^{0.2838}$	0.958
	Females	$y = -0.0008x^2 + 0.208x + 0.0633$	0.939	$y = 5.1807x^{0.2864}$	0.924
Face width	Total	$y = -0.0006x^2 + 0.1463x - 0.1676$	0.925	$y = 3.3009x^{0.3204}$	0.930
	Males	$y = -0.0006x^2 + 0.1466x - 0.1539$	0.917	$y = 3.3273x^{0.3202}$	0.938
	Females	$y = -0.0006x^2 + 0.145x - 0.1714$	0.936	$y = 3.271x^{0.3204}$	0.916

Table 4: Correlation between fetal leg measures (x) and CRL (y) and weight (y)

Parameter	Sex	CRL		Weight	
		Regression	R^2	Regression	R^2
Radius-Ulna length	Total	$y = 0.0518x^{1.2756}$	0.970	$y = 3.6809x^{0.4151}$	0.956
	Males	$y = 0.0482x^{1.2928}$	0.976	$y = 3.6393x^{0.4142}$	0.962
	Females	$y = 0.0567x^{1.2545}$	0.963	$y = 3.7178x^{0.4145}$	0.948
Metacarpal length	Total	$y = 0.099x^{1.2222}$	0.987	$y = 5.8902x^{0.3982}$	0.970
	Males	$y = 0.0983x^{1.2241}$	0.990	$y = 5.9082x^{0.395}$	0.978
	Females	$y = 0.1x^{1.2198}$	0.982	$y = 5.8637x^{0.4031}$	0.957
Tibia length	Total	$y = 0.0685x^{1.3305}$	0.984	$y = 5.8487x^{0.4335}$	0.964
	Males	$y = 0.0664x^{1.3388}$	0.988	$y = 5.8537x^{0.4317}$	0.974
	Females	$y = 0.0714x^{1.3197}$	0.979	$y = 5.8171x^{0.4382}$	0.953
Tarsal-Metatarsal length	Total	$y = 0.1138x^{1.2561}$	0.983	$y = 7.576x^{0.409}$	0.963
	Males	$y = 0.1161x^{1.2531}$	0.989	$y = 7.6696x^{0.4064}$	0.979
	Females	$y = 0.1113x^{1.2593}$	0.975	$y = 7.5143x^{0.4102}$	0.940

Table 5: Correlation between umbilical cord circumference (x) and CRL (y) and weight (y)

Sex	CRL		Weight	
	Regression	R^2	Regression	R^2
Total	$y = -0.0012x^2 + 0.1873x - 0.184$	0.897	$y = 3.8531x^{0.2706}$	0.889
Males	$y = -0.001x^2 + 0.1786x - 0.0418$	0.908	$y = 3.9025x^{0.2705}$	0.924
Females	$y = -0.0013x^2 + 0.1964x - 0.3449$	0.885	$y = 3.7892x^{0.2769}$	0.847

best described by the use of the power order function (Table 6). The correlation coefficients had no significant difference (P>0.05) between male and female. The correlations between fetal weight and fetal body measures are strongly expressed as:

$$y = ax^b$$

in which,
y: Fetal body measure
x: Fetal weight

The correlation between fetal weight and fetal head measurements is shown in Table 3. A correlation existed between measurements of fetal head and weight of fetus and can be described as:

$$y = ax^b$$

in which,
y: The measurements of fetal head
x: Fetal weight

The correlation between fetal weight and measurements of fetal leg are shown in Table 4. A correlation existed between length of fetal leg and weight of fetus being best fitted by the power function as:

$$y = ax^b$$

in which,
y: Measurements of fetal leg
x: Fetal weight

Table 6: Correlation between fetal weight (y) and body measurements (x)

Parameter	Sex	Regression	R^2
CRL	Total	$y = 28.047x^{0.3328}$	0.967
	Males	$y = 28.021x^{0.3362}$	0.974
	Females	$y = 28.082x^{0.327}$	0.955
Noise rump length	Total	$y = 37.215x^{0.3208}$	0.961
	Males	$y = 37.25x^{0.3245}$	0.964
	Females	$y = 37.238x^{0.3131}$	0.954
Eye rump length straight	Total	$y = 32.63x^{0.3203}$	0.953
	Males	$y = 32.716x^{0.3237}$	0.958
	Females	$y = 32.517x^{0.3149}$	0.942
Ear rump length	Total	$y = 28.321x^{0.3253}$	0.974
	Males	$y = 23.989x^{0.3514}$	0.968
	Females	$y = 23.832x^{0.3443}$	0.952
Body cross length	Total	$y = 18.253x^{0.3494}$	0.973
	Males	$y = 18.324x^{0.3496}$	0.981
	Females	$y = 18.191x^{0.3503}$	0.960
Chest circumference	Total	$y = 21.785x^{0.3224}$	0.971
	Males	$y = 21.865x^{0.3242}$	0.984
	Females	$y = 21.664x^{0.3197}$	0.947
Chest depth	Total	$y = 9.2176x^{0.3394}$	0.962
	Males	$y = 9.3144x^{0.3365}$	0.974
	Females	$y = 9.0782x^{0.3449}$	0.944

In all parameters the correlations of coefficients were very high ($R^2 \geq 0.94$).

The correlation between fetal weight and umbilical cord circumference is shown in Table 5. A function relation existed between fetal weight and umbilical cord circumference of fetus by:

$$y = ax^b$$

in which,
y: Umbilical cord circumference
x: Fetal weight

The correlation of coefficient had no significant difference between male and female.

Discussion

In bovine, Thomsen (1975) reported the ability prediction of gestational age in bovine fetus based on body length, head circumference and weight. This research concludes the correlation of the above parameters is valid for time interval corresponding to gestational age of approximately 9.9 to 34.8 weeks. Since 1991 there have been reports concerning the prenatal assessment of weight and dimension of the camel conceptus (Hussein et al., 1991). The report indicates that the coefficients of correlation between the CRL and other parameters (total conceptus length, weight hump circumference, chest circumference and the length of the radius and tibia) were highly significant in camel (Hussein et al., 1991).

This study uses large samples to describe the growth of buffalo fetuses during the pregnancy. It focuses on measuring a number of specific body parts and on determining the correlation between each of them. Currently, the precision of the measurements was judged by correlation coefficients between growth of fetal parts and organs, and gestational age, whereas the highest correlation was found with CRL and weight. In buffalo, the correlations between CRL and age of fetuses were used for 1st function during gestation (Schmidt et al., 1964; Abdel-Raouf and El-Naggar, 1968; Hussein et al., 1991) or 2nd function in first trimester (Ali and Fahmy, 2008). However, the information in growth rate approaching term have been recognized previously in

bovine (Eley *et al.*, 1978), and evidence for a decline of growth rate have been presented for sheep (Rattray *et al.*, 1974); and Human (McKeown *et al.*, 1976; Johnsen *et al.*, 2006). So that, the correlation was described by the 3rd function (Singh *et al.*, 1963; Rattray *et al.*, 1974; Eley *et al.*, 1978) or exponential function (Joubert, 1956).

The present study was a description of the correlation between age and weight of fetus and many parameter measurements during the pregnancy development of the swamp buffalo. Though descriptions of the correlations during gestational stages in the river buffalo have been reported (Singh *et al.*, 1963), it appears that the development in the buffalo species, river buffalo and swamp buffalo, is closely related. In all correlations, although there are some differencies of correlation coefficient between male and female but there are no significant differences between them. Eley *et al.* (1978) have shown that the growth rate of male is significantly higher than female in period less than 100 days in bovine.

In conclusion, these results indicate that the correlation coefficients between the estimated age of fetus and weight with other measurement parameters (total fetus length, body fetus measure, and the leg measure) were highly significant in swamp buffalo. The knowledge gained from this study forms a basis for further research in prenatal development in the swamp buffalo.

Acknowledgements

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