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Original Research

Transabdominal Laparoscopy and its Diagnostic Potential for the Evaluation of Abnormalities of Genitalia in Bovines

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Abstract

The present study comprised evaluation of transabdominal laparoscopy (TAL) as a reproductive diagnostic tool for abnormalities of genitalia in cows (n=28). Feed, but not water was withheld for a minimum period of 24 h and sedation with xylazine and local infiltration of portal sites was done. Blood and serum samples (n=8) were collected at pre (1 h) and post (1 and 24 h) laparoscopy for CBC, hormone and biochemical analysis. In the present study, both flank approaches proved feasible for evaluation of genitalia with direct trocar as better alternative for creating pneumoperitoneum compared to Veress needle method. Optimum site of laparoscopic port and instrument port was determined. TAL revealed ovarian abnormalities in 18 (85.7%), oviduct and adnexal pathologies in 17 (80.9%) and uterine abnormalities in 9 (42.8%) cows. Significant difference of cortisol at post 1 h and AST at 24 h post laparoscopy was observed only with respect to blood and serum analysis. In conclusion, TAL can be successfully done through flank approach for evaluation of genitalia in bovines and is very useful for diagnosing genital abnormalities especially oviduct and adnexa.

Keywords: Abnormalities, Bovines, Genitalia, Trans-abdominal Laparoscopy

Introduction

Laparoscopy is a procedure in which laparoscope is used to look at the organs and tissues through a small incision in the abdomen. Presently, it is used commonly as a diagnostic and intervention tool for various gynaecological procedures in humans. In veterinary practice, laparoscopy played an important role to enhance diagnosis and management of fertility (Sofi *et al*., 2018; Delaune *et al*., 2020). Laparoscopy has been used for diagnosis of gross reproductive abnormalities and for surgical intervention like paraovarian cyst removal in female animals (Sofi and Singh, 2018). Laparoscopy is now considered a standard procedure in the investigation and evaluation of infertile human females (Parveen and Khanam, 2010) and it allows direct visualization to reveal different pathologies. Although use of laparoscopy as a diagnostic tool for infertility evaluation is presently frequent in equines (Arnold and Love, 2013) but its use in bovines for infertility assessment is very meagre. So, the present study was conducted to standardize and assess the diagnostic potential of transabdominal laparoscopy for evaluation of abnormalities of genitalia in bovines.

Materials and Methods

Stray cattle (n=28) housed in university dairy farm of Himachal Pradesh Agricultural University, Palampur-HP (Temperate, 32.12°N, 76.53°E, 1219 m meters above sea level) constituted the study group. Cows were brought to the Gynaecology section of TVCC for examination of the genitalia through transabdominal laparoscopy (TAL). Feed, but not water was withheld for a minimum period of 24 h. The animal was restrained in standing crush designed for laparoscopy and sedated with xylazine (Xylaxin; Indian Immunological Ltd) @ 0.03 mg/kg BW (IM). Both left and right flank approach in standing position was applied for transabdominal laparoscopy. Portable laparoscope was used and all the laparoscopic instruments used were of KARL STORZ (Gmbh and Co. KG, Tuttlingen/Germany).

Laparoscopic Procedure

Paralumbar fossa was aseptically prepared for TAL and the procedure was done under local anesthesia using 2% lignocaine. The ports for laparoscope and instruments were made at different sites on both left and right flank to evaluate the optimum site for examination of genitalia. The measurements of the sites were taken with scale and noted. Two techniques were used for creating pneumoperitoneum: 1) Veress needle technique and 2) Direct entry method using a cannula (6 mm diameter). After optimum pneumoperitoneum was achieved, a 0° viewing angle rigid laparoscope (10 mm, 57 cm) was inserted through the cannula and the tract was searched and visualized on the monitor. Palpation probe and claw forceps inserted through the instrument ports were used to manipulate the genital tract for complete examination. The ovary, oviduct and uterus including cervix was examined and all significant observations were noted and recorded including video recording for further analysis. After complete examination, laparoscope and instruments were removed and CO_2 was allowed to escape through the portals. The incision sites were sutured with one or two interrupted sutures with standard post-operative care. The sutures were removed after 10-12 days of laparoscopic procedure.

Hematology and Serum Analysis

Blood samples (n=8) were collected from animals pre (1 h) and post (1 and 24 h) laparoscopy for hematology and serum analysis. Approximately 7 ml blood was collected in EDTA vials for complete blood count and equal volume was collected for separation of serum. Complete blood count was done immediately in automatic hematology analyzer (Mindray, BC-2800Vet). Serum analysis involved assay of hormones viz; cortisol, T3 and T4 and enzymes *viz.* ALT, AST, AP, GGT and total protein (TP). Cortisol was assayed by enzyme immunoassay kit (DRG Instruments, GmbH, Germany) and T3 and T4 by using commercially available ELISA kit (Calibiotech Inc., Austin). Similarly, ALT, AST, AP, GGT and TP were measured in serum by colorimetric method using semi-automatic biochemistry analyzer (Photometer 5010, ROBERT RIELE GmbH & Co, KG, GERMANY) with the commercially available kits (AGGAPPE DIAGNOSTICS LTD, Kerala, India).

Statistical Analysis

The data was analyzed by using statistical software SPSS version-16.

Results and Discussion

In the present study, laparoscopy was conducted in 28 animals and out of which genitalia of 21 animals were successfully examined (Table 1).

Table 1: Overview of different approaches, entry methods and complications of TAL

Parameter	Total	Left Flank	Right Flank	Veress needle method	Direct entry method
Total laparoscopy done	28	15	13	16	12
Successful laparoscopy	21	11	10	11	10
Failed during standardization	4	3	1	4	-
Failed after standardization	3	1	2	2	1
Rumen puncture	5	5	-	5	-
CO_2 effusion	3	0	3	2	1

Laparoscopic examination failed initially in 4 animals during standardization and in 3 animals examination was aborted due to either extra peritoneal CO_2 effusion, presence of intraabdominal adhesion or obstruction by omento-serosal layer. Although, the present study showed that laparoscopic examination of genitalia can be done at 24 h of fasting time but it has been observed that increase in fasting time reduced chances of rumen puncture on left flank approach and enabled movement of laparoscope and instrument during examination. This is in agreement with Patel *et al*. (2014). In the present study, both left and right flank approach was feasible for evaluation of genital tract with minor limitations which is in agreement with the findings reported earlier by different workers (Bleul *et al*., 2005). However, in the right side, hindrance to the movement of laparoscope and instruments due to rumen especially when fasting time is shorter was absent. Further, there was no chance of rumen puncture on the right side but laparoscopic examination was hampered in few cases due to omento-serosal layer and mesentery on right side which has not been observed on left flank approach which is also in accordance with the observations found by Bleul *et al.* (2005). Direct trocar technique using 6 mm trocar cannula unit proved better alternative for creating pneumoperitoneum than the Veress needle method as reported earlier by Vilos *et al*. (2013). Intraabdominal insufflation pressure maintained during examination was 6-10 mmHg with an average of 8 mmHg. At this insufflation pressure, examination was done comfortably as higher pneumoperitoneum lead to straining and physical discomfort by the animal during examination. These observations are in agreement with earlier reports (Maiti *et al*., 2013). Although, there are reports of laparoscopy at higher intraabdominal pressure (12 mmHg; Pizzi *et al*., 2010) but in the present study, it has been observed that the animal became restless and had signs of colic when intraabdominal pressure was increased to 10 mmHg or above.

In the present study, optimum site of laparoscopic port in both flank approaches was 8-10 cm cranial to the tip of tuber coxae and 6-8 cm ventral to the transverse processes of lumbar vertebrae at the junction of middle and caudal third flank. Similarly, optimum site of instrument port both in right and left flank approach was 18-20 cm ventral to the tip of tuber coxae and 2-3 cm cranial to that point. However, in few laparoscopic examinations second instrument port in both flank approaches was made at 10-12 cm ventral to the tip of tuber coxae and 4-6 cm cranial to that point. The optimal portal sites were determined by testing various locations and those described above were optimum for examination of genitalia in cattle. The CO_2 consumption for each laparoscopic examination was 45-80 litres (average: 60 litres) and the amount varied with abdominal size and duration of examination. The average time for complete laparoscopic examination was 60-90 minutes. However, the time required varied with the entry technique, size of abdomen and cooperation by the animal. All the animals recovered uneventfully without any complications.

Among 21 laparoscopic examinations, ovarian abnormalities were diagnosed in 18 (85.8%), oviduct and adnexal pathologies in 17 (80.9%) and uterine abnormalities in 9 (42.8%) cows (Table 2, Figure 1-14). In one animal, perioophoritis (Figure 5) was present involving right ovary in which ovarian surface was shaggy and fully encapsulated without any functional structures, as also reported earlier by Purohit (2014). Hydrosalpinx (Figure 6) was seen as distended tubes containing fluid with tubes twisted, closely apposed and was frequently associated with peri-tubular adhesion. Parovarian cyst was diagnosed by presence of transparent glistening fluid filled structure attached to but separate from the ovary (Figure 8) or in the adnexa (Figure 9).

Table 2: Different abnormalities of genitalia diagnosed by TAL (n=21)

Genital part	Abnormality		No		%age
Ovary	True anestrus		2		9.5
	Ovarobursal adhesion	Right	8	13	61.9
		Left	4		
		Both	1		
	Cystic ovaries	Right	1	1	4.8
	Ovarian abscess			1	4.8
	Perioophoritis			1	4.8
	Total			**18**	**85.7**
Oviduct and Adnexa	Hydrosalpinx	Right	2	4	19
		Left	2		
	Oviductal adhesion	Right	4	9	42.8
		Left	4		
		Both	1		
	Parovarian cyst	Right	2	3	14.3
		Left	1		
	Tubo-ovarian abscess	Right	1	1	4.8
	Total			**17**	**80.9**
Uterus	Uterine adhesion			4	19
	Abnormal color with nodules			4	19
	Uterine cyst			1	4.8
	Total			**9**	**42.8**
Total Abnormalities detected: **44**					

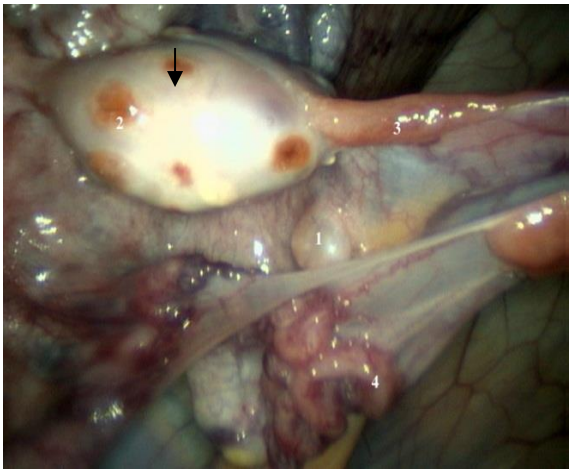

Figure 1: Laparoscopic image of inactive ovary (arrow); absence of follicles and CL.

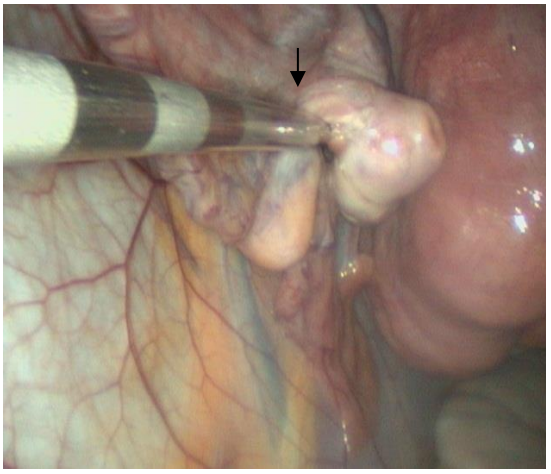

Figure 2: Laparoscopic image of ovarobursal adhesion (arrow); presence of fibrous tissue attached to ovary.

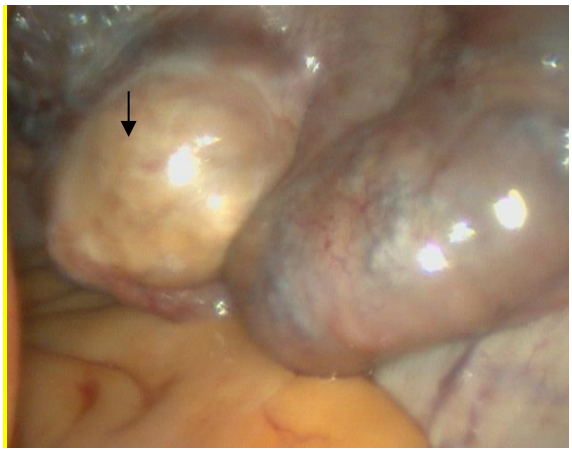

Figure 3: Laparoscopic image of luteal cyst (arrow).

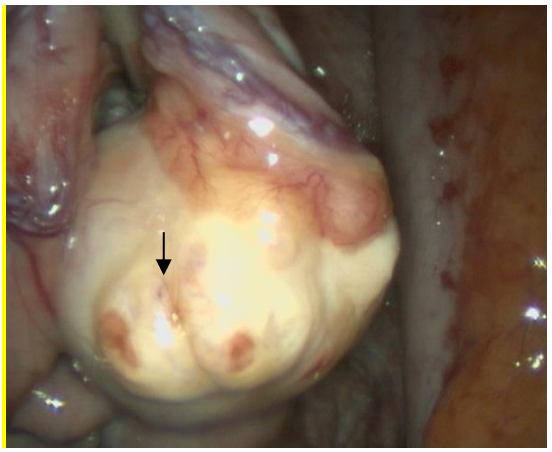

Figure 4: Laparoscopic image of ovarian abscess (arrow).

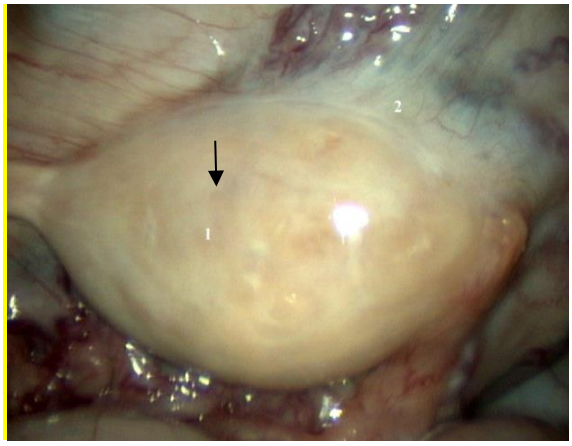

Figure 5: Laparoscopic image of perioophoritis (arrow); whole ovary is encapsulated.

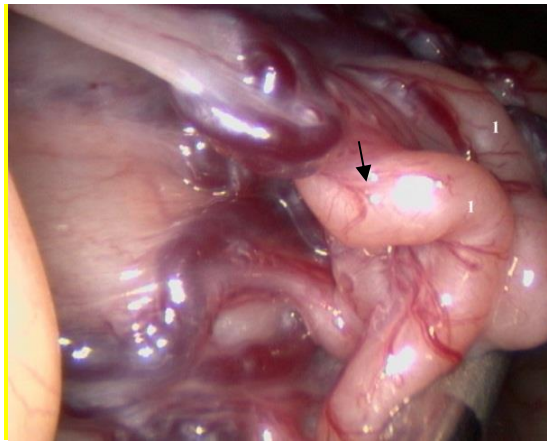

Figure 6: Laparoscopic image of hydrosalpinx (arrow); dilated oviduct with fluid.

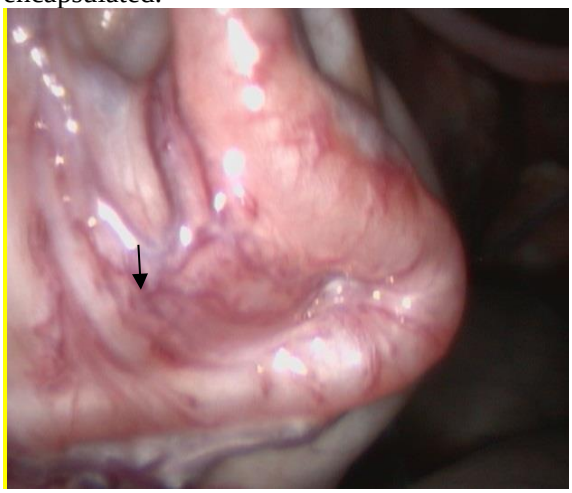

Figure 7: Laparoscopic image of oviductal adhesion (arrow).

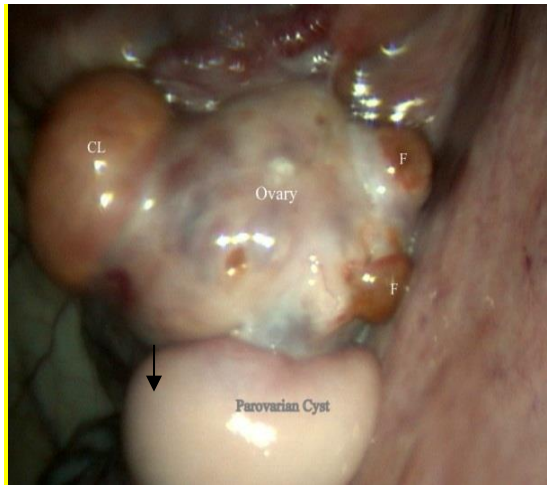

Figure 8: Laparoscopic image of parovarian cyst besides ovary (arrow).

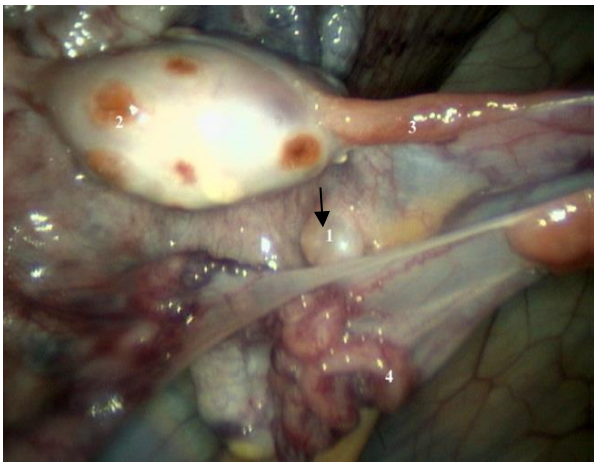

Figure 9: Laparoscopic image of parovarian cyst in adnexa (arrow); presence of small glistening cyst.

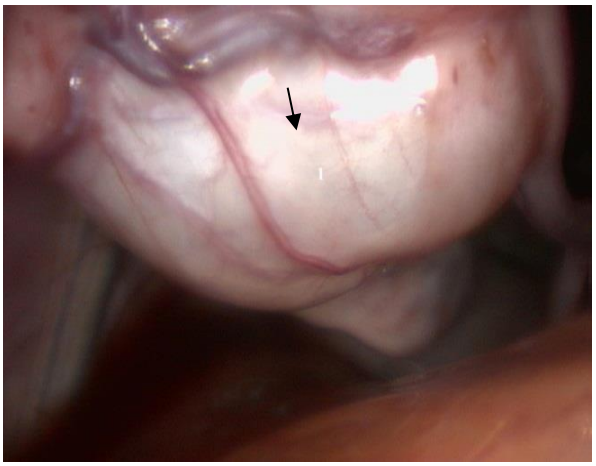

Figure 10: Laparoscopic image of tubo-ovarian abscess (arrow).

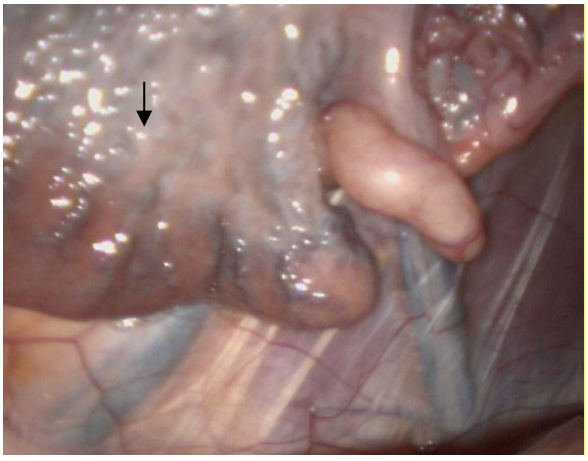

Figure 11: Laparoscopic image of uterine adhesion (arrow).

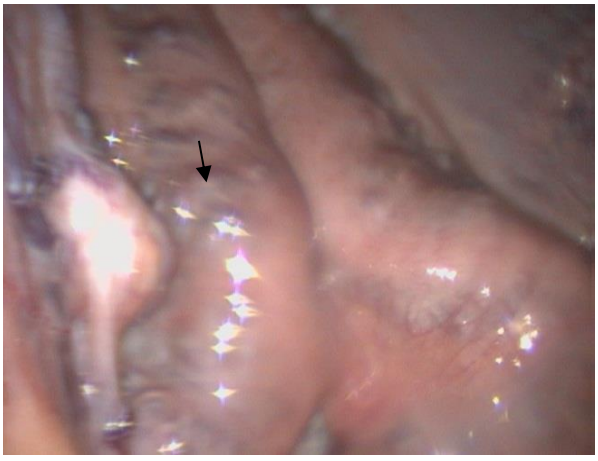

Figure 12: Laparoscopic image of abnormal color of uterus with nodules (arrow).

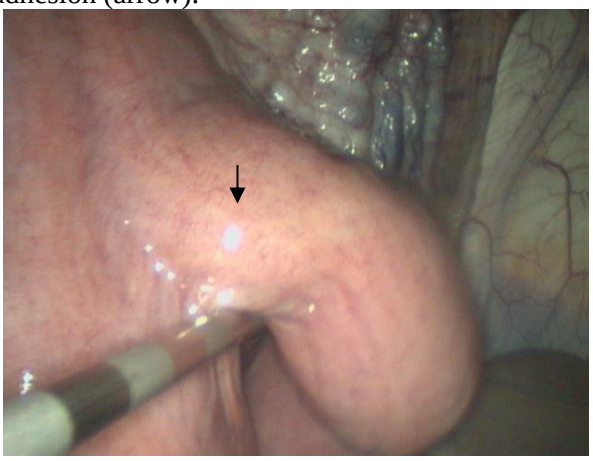

Figure 13: Laparoscopic image of normal uterus (arrow).

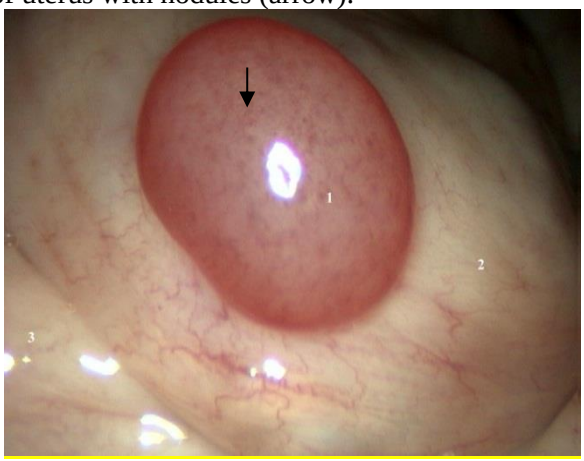

Figure 14: Laparoscopic image of uterine cyst (arrow).

Split sign as criteria for diagnosing parovarian cyst attached to ovary was also done with the help of palpation probe as it was confused with the true ovarian cyst. On laparoscopy, there were also seen single or multiple fleshy looking, red with blue hue nodules on the surface of the uterus with bluish discoloration (Figure 12) in contrast to normal uterine horns (Figure 13) seen as reddish colored tapering tubes clearly distinct from other organs. However, exact diagnosis was not made as further diagnostic procedures like biopsy or histology was not done. In one animal, uterine cyst (Figure 14) was observed as small pinkish fluid filled structure on the dorsal surface of uterus.

Complete blood count of cows at pre- and post-laparoscopy revealed no significant difference between haematological parameters. These findings are in agreement with that of Maiti *et al.* (2013). Heparinized blood was collected before and after anaesthesia, 30, 60 and 120 minutes after establishment of capnoperitoneum and 24 h after laparoscopy and no significant changes were observed in PCV, Hb and TLC when comparisons were made between the groups at different time intervals (Maiti *et al.*, 2013). Results of serum analysis (Table 3) revealed significant difference of cortisol between pre and post 1 h but the level reached non-significant value at post 24h laparoscopic examination.

Table 3: Hormone and enzyme analysis (n=8) at pre (1 h) and post (1 and 24 h) laparoscopy (Mean ± S.E)

Parameter	Pre 1 h	Post 1 h	Post 24 h
Cortisol (ng/ml)	0.93^{ac}± 0.08	1.21^{bc}± 0.07	1.12^{cab}± 0.05
Triiodothyronine (T3) (ng/ml)	0.79± 0.07	0.82±0.13	1.07±0.14
Thyroxine (T4) (µg/dl)	0.298±0.03	0.306±0.02	0.380±0.04
Alanine transaminase (ALT)(U/L)	30.9±3.92	32.6±4.20	39.7±4.72
Aspartate transaminase (AST)(U/L)	79.8^{ab}±10.7	88.4^{ba}±9.6	134.7^{c}±10.8
Alkaline phosphatase (AKP) (U/L)	70.0±8.4	71.1±11.1	79.5±4.8
Gamma-glutamyltranspeptidase (GGT)(U/L)	10.8±1.01	11.6±1.21	11.7±1.4
Total Protein (g/dl)	6.99±0.29	6.83±0.29	6.78±0.11

Mean values within the same row with different superscripts differ significantly ($p \leq 0.05$)

In accordance with the present findings, there was transient rise in plasma cortisol level in ewes undergoing laparoscopic examination (Martin *et al*., 1981) and cortisol levels started to increase at 30 minutes after capnoperitoneum and reached to peak at 120 minutes after capnoperitoneum, which were significantly higher than the base value at before and 24h laparoscopy procedure (Maiti *et al*., 2013). However, no significant difference was found in the serum level of T3 and T4 before and after laparoscopy as reported earlier (Dutta *et al*., 2010). There was a consistent but non-significant increase in ALT, GGT and alkaline phosphatase at post 1 and 24 h compared to 1 h before laparoscopy but serum level of AST reached significantly higher level at 24 h post laparoscopy as reported by Latimer *et al*. (2010) Similarly, ALT and AST were within the normal range in all the animals before and after laparoscopy as reported earlier (Maiti *et al*., 2013). With respect to total protein, no significant difference was observed at pre- and post-laparoscopy. Thus, the study revealed that laparoscopy examination of genital tract in bovines did not adversely affect the haematology and plasma chemistry and can be concluded as safe procedure in bovines.

Conclusion

Transabdominal laparoscopic for evaluation of genitalia can be successfully done through flank approach under local anaesthesia at an intraabdominal pressure of 7-8 mmHg using CO_2 for insufflation in bovines with direct entry technique as better alternative technique for creating pneumoperitoneum. TAL is a safe procedure and enables to diagnose many conditions especially oviductal and adnexal abnormalities which are difficult to be accurately delineated either manually or by ultrasound examination.

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Conflict of Interests

There is no conflict of interest.

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