Risk factors associated with surgical site infections in cattle undergoing enterectomies

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SUMMARY

Post-operative surgical site infections (SSIs) have a detrimental impact on patient mortality, morbidity, and medical expenses. There has not been any research done on SSIs in cattle following enterectomies. Few studies have documented surgical site infection rates in cattle after caesarean section and right paramedian abomasopexy, respectively. Therefore, the purpose of the current study was to determine various risk factors responsible for surgical site infections (SSIs) in cattle having enterectomies. The present study was conducted in cattle (n=30) suffering from intussusceptions and underwent laparo-enterectomies to evaluate rate of SSIs and the risk variables associated with it. Pre-operative, intra-operative, and post-operative parameters were noted, and each case was monitored for 35 days following discharge on alternate day. These parameters were then compared between cattle that developed SSIs and the cattle that did not develop SSIs. The overall rate of SSIs in laparo-enterectomies was 40% (12/30). The pre-surgical risk factors; Increased days of illness (6.42 ± 0.48 days), increased age (3.13 ± 0.29 years), hypoalbuminemia (2.53 ± 0.08 g/dl), ineffective scrubbing (n=3), increased TLC level (24236 ± 2206.7/mm^3), and increased PCV (41.64 ± 1.38%) were found to be associated with a higher risk for SSIs in cattle. Intra-operative risk factors included; contamination (n=1), increased mean duration of procedure (117.33 ± 2.53 minutes), increased blood loss (540.83 ± 56.10 ml), use of subcutaneous sutures and operation by surgeons with less level of experience. Post-operative risk factors included; more than 2 days hospital stay (n=6), dehiscence of subcutaneous suture material (n=9), severe hypoalbuminemia (<2.5 g/dl, n=9) and anemia (<8 g/dl, n=7) in post-operative days. In conclusion, cattle with intussusception require emergency laparo-enterectomy, which is a clean contaminated surgery. These surgeries possess a high risk of post-operative infections. The poor prognostic factors for SSIs in cattle include; hypoalbuminemia, anaemia, increased TLC and PCV, intra operative blood loss, increased duration of surgery and use of subcutaneous sutures.

KEY WORDS

Bovines; laparo-enterectomies; nosocomial infections; predictors of surgical site infections; risk factors.

INTRODUCTION

Despite improvements in surgical techniques, the accessibility of antibiotics, and aseptic procedures in the modern era, surgical site infections (SSIs) continue to pose a serious threat to patients undergoing invasive surgeries[1]. According to the centers for disease control and prevention (CDC), a surgical site infection is defined as an infection that appears in the vicinity of the surgical site within 30 days following the surgery or up to 90 days following the procedure if an implant is used[2]. SSIs fall into one of three categories: superficial if limited to skin and subcutaneous tissue, deep when they impact muscle and fascia, or organ space when they affect a body cavity.
SSI rates have previously been reported to vary from 1.3-25.9% in cows after c-section[3], 10.6-58.3% in cows after right paramedian abomasopexy[4] and 10-37% in horses after laparotomy[5]. According to Weese (2008), post-operative surgical site infections (SSIs) have serious consequences that include patient morbidity, patient death, higher treatment costs, longer hospital stays, client and veterinarian dissatisfaction, potential liability, and unfavourable public perceptions[6].
As per the authors knowledge, studies on the evaluation of SSIs in large ruminants undergoing enterectomy is not reported. Therefore, the present work was planned to initiate a systematic clinical study on the assessment of pre-operative, intra-operative and post-operative risk factors responsible for SSIs in cattle undergoing laparo-enterectomies.

MATERIALS AND METHODS

The present study was conducted on cattle (n=30) suffering

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from intussusceptions and underwent laparo-enterectomies. All the surgeries were performed on the standing cow through the right paralumbar fossa under local anesthesia using a line block. Pre-operative, intra-operative and post-operative parameters were recorded for each case. Further, the study was divided into cattle that developed SSIs (n=12) and cattle (n=18) that did not develop SSIs. Pre-operative parameters included patient parameters (Age of cattle, duration of illness, haemato-biochemical parameters like estimation of haemoglobin (Hb), packed cell volume (PCV), total leukocyte count (TLC) and albumin levels, swabbing of surgical site for microbiological examination after the surgical site scrub (Figure 1) (scrubbing was done using antiseptic solution containing chlorhexidine gluconate- 7.5% v/v, cetrimide- 15% w/v and isopropyl alcohol- 7% v/v at recommended dilution for 5 minutes), swabbing of scrubbed hands for microbiological examination, checking of optimum temperature requirement of sterilization using chemical indicators (Sterility indicator strips were placed in each surgical pack (before placing the pack in the autoclave)) and Swabbing of the surgical pack (After opening the sterile surgical pack, sterile swabs were used for taking samples from random instruments and sent for culture test. Lack of visible colonies on the culture indicates the pack to be completely sterile). In all the cases, a combination of enrofloxacin @ 5 mg/kg body weight and gentamicin @ 4 mg/kg body weight were used half hour before making skin incision. Intra-operative parameters included level of experience (faculty member/ postgraduate/ undergraduate student surgeon) and classification of surgical wound as clean contaminated or contaminated based on the degree of intra-operative contamination. Other intra-operative parameters included duration of procedure (the time from skin incision to that of skin closure), intra-operative blood loss assessed by swab weighing method and volumetric method, surgical technique, suturing technique and suture materials used and number of people (students, owner(s), attendants and others) present in the operation theatre at the time of surgery.

Each surgical team consists of 3 members (1 faculty member and 2 postgraduate students/ 3 postgraduate students/ 2 post-graduate student and 1 under-graduate student). Faculty member included were having more than 15 years of experience in bovine practice. Post-graduate student surgeons included were having more than 1 year of experience in bovine practice. The role of under-graduate students was limited to scrubbing of the patient and passing of the surgical instruments. Under-graduate students were not associated with handling of tissues or the surgical procedure. All the surgeries were performed under the supervision of the senior surgeons.

Post-operative parameters included incision care by sterile dressing for 2 days post-operatively, antimicrobials administration (enrofloxacin @ 5 mg/kg body weight and gentamicin @ 4

Figure 1 - Photograph showing Swab taking from the surgical site.

Figure 2 - Incision care by sterile gauze (a) and additional clean cloth (b).

mg/kg body weight) for the next 4 days, direct observation of the surgical site for 2 days and estimation of haemoglobin and albumin immediately after the surgery and on day 1 and day 2 post-operatively. The surgical wound was covered with sterile dressing (Figure 2a) and the cattle were kept in a clean place. The wound was further covered by another clean cloth (Figure 2b). This layer of clean cloth was changed every day or sooner if became wet, as it might become source of contamination especially when wet and used for prolonged duration. Swabs of the surgical site were taken at the immediate post-operative period, on the first and second post-operative day and processed for presence of infection.

Each case was discharged after 2 days and then was followed for SSI's up to 35 days post-operatively. Patients were assessed for systemic (fever) and local (pain, swelling, purulent drainage) signs of infections. Wound dehiscence or suture dehiscence if any, was recorded. Time of suture removal was also recorded. All the data was compared using student t-test between cattle with surgical site infections and without surgical site infections to find out the predictors of surgical site infections.

RESULTS AND DISCUSSION

The total rate of SSIs following laparo-enterectomies in cattle was 40% (12/30), whereas 60% (18/30) of cases were free of surgical site infections. Using the CDC classification, 83.33% (10/12) of the cattle had superficial SSIs and 16.67% (2/12) had deep incisional SSIs. Risk factors associated with SSIs are classified as pre-operative, Intra-operative and post-operative risk factors.

Preoperative Risk Factors

The risk of surgical site infections in cattle that underwent laparo-enterectomies increased with increasing mean duration of illness, increasing age, hypo-albuminemia, ineffective scrubbing, increased TLC level and increased PCV. Pre-operative risk factors for surgical site infections are summarized in **Table 1**. Duration of illness in the present study was correlated with the duration of anorexia and reduced water intake. Prolonged anorexia was observed in the pre-operative period in cattle with SSIs, which might have decreased hepatic albumin secretion leading to hypoalbuminemia[7]. In addition to this, capillary leakage of albumin into the interstitial space and increased catabolism of proteins in intestinal diseases might have further decreased the albumin levels[8]. A lower pre-operative serum albumin level (2.53 ± 0.08g/dl) may be the cause of the higher incidence of SSIs seen in our study. Hypoalbuminemia has been reported earlier as a strong risk factor for development of SSIs[9]. SSIs due to increased age of cattle in the present study might be due to increased likelihood of comorbidities, malnutrition (as most of the cattle (n=7) that developed SSIs were associated with hypoalbuminemia) and decrease in body immunological efficiency, causing more extensive SSIs. Wilson et al. (1995) reported that the risk of incisional infection in horses (n=274) undergone celiotomies seems to be greatest at age older than 1 year[10]. Increased PCV and TLC level were observed in cattle that developed SSIs. Increased TLC values might indicate intense systemic inflammation or remote infection, which further possess a great risk for SSIs[11].

Other pre-operative factors associated with SSIs were ineffective scrubbing of surgical site (n=1) and surgeon hands (n=2). *Staphylococcus aureus* was the organism isolated in all the three cases in the pre-operative culture as well as from the SSI samples. Culture results of the swabs taken after break in asepsis in the pre-operative, intra-operative and post-operative period are shown in **Table 2.** Inadequate pre-operative preparations might have been responsible for the SSIs development in these cases. Transmission of microbial pathogens by the hands of health care workers during patient care plays a crucial role in development of SSIs, particularly superficial SSIs[12]. Culture results from the instrument swabs and change of colouration of sterility strips showed the packs to be completely sterilized.

Intraoperative Risk Factors

Intra-operative risk factors associated with SSIs in the present study were intra-operative contamination (n=1), increased mean duration of procedure (134 minutes), increased blood loss (540.83 ± 56.10 ml), use of subcutaneous sutures and operation by students with less level of experience**.** Intra-operative risk factors for surgical site infections are summarized in **Table 3.**

A total of 29 procedures were categorized as Clean-contaminated and 1 as contaminated based on break in asepsis and spilling of GIT contents. Out of the clean-contaminated procedures, there were eleven SSIs (ten superficial and one deep incisional SSI). The contaminated procedure led to the deep incisional SSI. Swab was taken immediately from the contaminated site and the cultural result showed the presence of *E.coli* as shown in **Table 2.** Complexities of the surgeries associated with the wound classifications can be another reason for increased rate of SSI in the present case as surgeries involving GI tract carry an inherent risk of bacteria laden contents spilling into wounds causing severe contamination and often sepsis and death[13].

Table 1 - Stratification of pre-operative risk factors responsible for surgical site infection.

Pre-operative parameters	Cattle with SSIs (n=12)	Cattle without SSIs (n=18)
Duration of illness (days)	6.42 ± 0.48*	4.89 ± 0.40
Age (years)	3.13 ± 0.29**	1.97 ± 0.19
Albumin level (g/dl)	2.53 ± 0.08*	2.79 ± 0.06
TLC (/mm^3)	24236 ± 2206.7**	10461 ± 1019.2
PCV (%)	41.64 ± 1.38*	37.6 ± 0.61
Haemoglobin (g/dl)	9.88 ± 0.36	10.28 ± 0.20

The superscript ((**) and (*) show significant difference between the same variables among cattle with SSIs and without SSIs at 1% and 5% level of significance.

Table 2 - Microbiological assessment of swabs taken after break in asepsis or sterility in the pre-operative, intra-operative and post-operative period.

	Break in asepsis		
	Ineffective scrubbing of surgical site (n=1)	Ineffective scrubbing of hands (n=2)	Intra-operative contamination (n=1)
	Culture results of the swabs taken		
Pre-operative period	*Staphylococcus aureus*	*Staphylococcus aureus*	No colonies were observed
Intra-operative period	Not taken	Not taken	*E. coli*
Immediate post-operative period	*Staphylococcus aureus*	*Staphylococcus aureus*	*E. coli*
Post-operative period (Day 1)	*Staphylococcus aureus*	*Staphylococcus aureus*	*Staphylococcus aureus* and *E. coli*
Post-operative period (Day 2)	*Staphylococcus aureus*	*Staphylococcus aureus*	*Staphylococcus aureus* and *E. coli*
SSI swab	*Staphylococcus aureus*	*Staphylococcus aureus*	*Staphylococcus aureus* and *E. coli*

In the present study, cattle with SSIs (n=12) had a mean surgical duration of 117.33 ± 2.53 minutes while cattle without SSIs had duration of 93.00 ± 2.63 minutes. Increased duration of surgery might have increased the risk of SSIs development due to prolonged exposure of wound to environment leading to more chances of inoculation of micro-organisms, prolong trauma and stress associated with increased duration of procedure[14]. Additionally, a prolonged operative time is often related to increased blood loss which contributes to tissue hypoxia[9].

Intra operative blood loss was assessed by swab weighing method and volumetric method (Figure 3). The Mean ± S.E of blood loss in cattle without SSIs (n=18) was 322.50 ± 21.04 ml while the blood loss

In cattle with SSIs (n=12) was 540.83 ± 56.10 ml. All the cattle (n=8) having increased intra operative blood loss (>500 ml) were found to have SSIs. Ejaz et al. (2017) also reported increased blood loss (>600ml) as a risk factor for causing SSIs in patients following major abdominal surgeries[15].

The increased blood loss in this study may have led to a drop in post-operative haemoglobin levels, which in turn may have lowered immunological function, produced tissue hypoxia, and slowed the healing of wounds. Hypoxia reduces oxygen and tissue perfusion, which in turn reduces tissue regeneration and promotes bacterial growth[9].

The present study showed reduced percentage of SSIs with double layered abdominal muscle closure as compared to double layered abdominal muscle closure along with subcutaneous layer and single layered abdominal closure either alone or with subcutaneous layer. In the present study, it was observed that in maximum number of cases (n=9) the subcutaneous sutures were the source of infection. Subcutaneous suture material in the dehiscence subcutaneous layer might act as foreign material and a source of nutrition for the bacteria and thus increases the risk of SSIs. Coomer et al. (2007) also reported two-layer abdominal closure (without subcutaneous layer) as a safe alternative means of achieving ventral midline abdominal closure in horses as compared to three layers (2 layered abdominal closure and the subcutaneous layer)[16]. The Present study showed the increased risk

Table 3 - Stratification of intra-operative risk factors responsible for surgical site infection.

Intra operative parameters		Cattle with SSIs	Cattle without SSIs
Type of surgery	Clean-contaminated	11/12	18/18
	Contaminated	1/12	0
Duration of surgery (Minutes)		117.33 ± 2.53*	93 ± 2.63
Amount of blood loss (ml)		540.83 ± 56.10*	322.50 ± 21.04
	250 ml	0	7
	250-500 ml	4	11
	> 500 ml	8	0
Abdominal closure layers	Double layer abdominal muscle closure (n=8)	1/8 (12.5%)	7/8
	Double layer abdominal muscle closure + subcutaneous suture (n=9)	4/9 (44.44%)	5/9
	Single layer abdominal muscle closure (n=7)	4/7 (57.14%)	3/7
	Single layer abdominal muscle closure + subcutaneous layer (n=6)	3/6 (50%)	3/6 (50%)
Level of experience	Post-graduate students only (Surgeries= 12)	4/12 (33.33%)	8/12 (66.67%)
	Post-graduate and undergraduate students (Surgeries= 8)	5/8 (62.5%)	3/8 (37.5%)
	Post-graduate students and faculty members (Surgeries= 10)	3/10 (30%)	7/10 (70%)
Number of people at the time of surgery		6.67 ± 0.40	5.5 ± 0.22

Values having different superscript (*) in a column differ significantly from each other ($p<0.05$) at 5% level of significance.

Figure 3 - Photographs showing intra-operative blood loss estimation by swab weighing and volumetric method.

of SSIs with subcutaneous layer which could be due to dehiscence of subcutaneous layer in the post operative period. The main reasons for dehiscence might include loss of strength in absorbable sutures, breakage of sutures, knot failure and tissue failure. Excessive strain on the suture line might probably be the greatest cause of wound dehiscence and the strain could occur during recovery or during the postoperative period[17].

The average mean of persons in the present study that were present at the time of surgery in cattle that developed postoperative SSIs (n=12) were higher (6.67 ± 0.40) than the average number of persons that were present at the time of surgery (5.5 ± 0.22) in cattle that didn't developed SSIs (n=18). The difference was not statistically significant. Eugster et al. (2004) also reported increased incidence of SSIs to be associated with increased number of persons at the time of surgery[18]. It might be possible that the number of persons in the operating room could be an indicator of another risk factor. Therefore, a limitation on personnel in the operating room is still a valid recommendation for a reduction of SSI frequency.

There was no variation in number of members of surgical team. Each team consists of 3 surgeons. The study showed that faculty and post graduate (P.G) students got minimum number of SSIs (30%) followed by post graduate (P.G) (33.33%) and finally by post graduate (P.G) and undergraduate students (U.G) (62.5%). The decreased percent of SSIs by faculty and P.G students could be due to increased level of experience of the faculty members as they have done a greater number of laparo-enterectomies as compared to post graduate and undergraduate students[19]. The higher percentage of SSIs by post graduate (P.G) and undergraduate students (U.G) might be due to prolonged operations due to technical delays related to level of competence. This prolongation in operation time exposes the tissues to atmosphere causing desiccation and contamination which makes the wound susceptible to developing post-operative infection.

Postoperative Risk Factors

The abdominal incision was covered with a sterile gauze for a period of 2 days due to the fact that the continuity of skin gets restored within 48 hours (physical closure of wound). The dressing might act as a physical barrier to protect the wound until the continuity of the skin in restored (within about 48 hours)[20].

Suture or wound dehiscence was not evident in any case in the post-operative day 1 and 2. Each case was then followed up to 35 days on every alternative day through home visits and through telephone. The mean duration of diagnosis of SSI was 8.08 ± 0.93 days. The area of infection showed signs of warmth, swelling and drainage. Grönlund and Bergström (2013) also reported that median duration of 7 days for SSIs diagnosis in horses[12]. The mean time of skin suture removal was 12.42 ± 0.40 days. Post operative risk factors for surgical site infections are summarized in **Table 4.**

Out of 12 cases that developed SSIs, 3 cattle were positive for *staphylococcus aureus* in the immediate post-operative period which might be due to ineffective scrubbing of hands, surgical site and environment contamination while 1 was positive for both *staph*ylococcus aureus and *E.coli* (Contaminated surgery). During surgery, microorganisms tend to repopulate an area following decontamination and this occurs at a much quicker rate than the original colonization took place. Gram positive staphylococcal bacteria might also be able to contaminate the site during surgery even with aseptic procedures and leads to SSIs[21].

Haemato-biochemical parameters like Hb and albumin were also estimated at immediate post-operative period, at 24 hours and at 48 hours. Severe hypoalbuminemia (2.28 ± 0.11 g/dl) and lower Hb values (8.13 ± 0.33 g/dl) were observed at the post operative day 2 in cattle with SSIs. The decreased albumin level in the immediate post-operative period would be due to leakage of albumin at damaged site, decreased production of albumin by liver, blood loss and surgical induced inflammation (systemic and tissue) which further redistributes the fluid to interstitial space from plasma thus further decreasing albumin level[22]. The stress of surgery might deplete protein reserves and lead to multiple organ dysfunctions and greater susceptibility to nosocomial infections[23]. Patients with hypoalbuminemia are at risk of impaired systemic and intestinal immune function, as well as decreased digestive and absorptive capacity due to the altered architec-

Table 4 - Stratification of post-operative risk factors responsible for surgical site infection.

Post operative parameters		Cattle with SSIs	Without SSIs
Post-operative contamination of surgical site (n=3)		3/3	0
Duration of hospital stay (Days)	Average	2.5 ± 0.31	2.33 ± 0.26
	2 days (n=18)	6/18 (33.33%)	12/18 (66.67%)
	More than 2 days (n=12)	6/12 (50%)	6/12 (50%)
Post-operative Hb level	Immediate	8.68 ± 0.40	9.36 ± 0.20
	At 24 hours	8.4 ± 0.32*	9.14 ± 0.16
	At 48 hours	8.13 ± 0.33*	8.94 ± 0.15
Post-operative albumen level	Immediate	2.00 ± 0.09**	2.34 ± 0.07
	At 24 hours	2.22 ± 0.10*	2.54 ± 0.07
	At 48 hours	2.28 ± 0.11*	2.57 ± 0.06
Subcutaneous suture Dehiscence (n=9)		9	0

Values having different superscript (**) in a column differ significantly from each other ($p<0.01$) at 1% level of significance. Values having different superscript (*) in a column differ significantly from each other ($p<0.05$) at 5% level of significance.

ture of the gut barrier. A deficiency of protein can impair wound healing (capillary formation, fibroblast proliferation, proteoglycan synthesis, collagen synthesis, and wound remodeling) and affects immune system, with resultant decreased leukocyte phagocytosis and increased susceptibility to infection[9].

The higher incidence of SSIs in our study might be due to the lower pre and post operative (2.53 ± 0.08g/dl and 2.28 ± 0.11g/dl) serum albumin level. Nine cattle that developed SSIs were found to have severe hypoalbuminemia (<2.5 g/dl) at the postoperative day 2 which might be due to the surgical stress or increased blood loss in the intra-operative period[23]. Six of these cattle were having extremely low albumin levels (<2 g/dl). The cattle without SSIs were found to have only mild hypoalbuminemia at post operative day 2. Thus, presence of severe hypoalbuminemia at post operative day 2 (<2.5 g/dl) has been found to be positively associated with development of SSIs. Post operative hypoalbuminemia has been reported earlier as a risk factor for the development of SSI after abdominal surgeries[23]. According to Labgaa et al. (2017), early postoperative decreases in serum albumin on the first postoperative day are linked to worse outcomes and infections[24].

A decreased Hb level (<8 g/dl) was found at the post operative day 2 in majority of the cattle (n=7) that developed SSIs. The mean Hb level in cattle that developed SSIs at post operative day 2 was 8.13 ± 0.32 g/dl whereas the mean Hb level in cattle that did not develop SSIs at the post operative day 2 was higher (8.94 ± 0.15 g/dl). This could be due to increased amount of blood loss in cattle that developed SSIs**.** Due to post operative anaemia, there might be tissue hypoxia, nutrient deplete wound microenvironment, impaired wound healing and decreased immunity which was favorable for bacterial multiplication[9]. Anaemia has been reported earlier to be linked with increased incidence of SSIs following abdominal surgeries[11].

Majority of the cattle (n=18) were discharged after 2 days. Out of these cattle, 6 got SSIs with a percentage of 33.33% (6/18) and for the cattle (n=12) that stayed for more than 2 days, 50% (6/12) showed SSIs. As previously mentioned by Roberts (2013), it has been demonstrated that a longer hospital stay is associated with a greater incidence of SSIs[25]. Longer hospital stay (more than 2 days) might be due to the post-operative complications like hypoalbuminemia. Isgren et al. (2017) also reported increased duration of hospital stay as a risk factor for SSIs development in horses[26].

CONCLUSION

Clean contaminated surgeries possess a high risk of post-operative infections. The poor prognostic factors for SSIs in cattle include; hypoalbuminemia, anaemia, increased TLC and PCV, intra operative blood loss, increased duration of surgery and use of subcutaneous sutures.

Conflicts of interest

The authors have no conflicts of interest with anyone.

Authors contribution

Author 1: Study design, acquisition, analysis, and interpretation of data, drafting and revising manuscript, approved final article; Author 2: Acquisition, analysis, and interpretation of data, drafting and revising manuscript, approved final article; Author 3: Study design, drafting and revising manuscript, approved final article.

Acknowledgements

The authors are grateful to the Sher-e-Kashmir University of Agricultural Sciences and Technology of Jammu for proving the necessary facilities for conducting this study.

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