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Investigations on pathomorphological alterations in ruminants affected with gastrointestinal tract disorders

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Abstract

Gastrointestinal disorders are one of the most important factors that affect the productive and reproductive performance of the animals. Comprehensive studies on such disorders is lacking therefore the present work was planned to aim at patho-biological investigation of gastrointestinal tract disorders affecting large and small ruminants. The current investigation was conducted on 65 cases (bovine, ovine and caprine) brought to the Department of Veterinary Pathology, LUVAS, Hisar from August, 2019 to January, 2020. The investigation included the detailed post-mortem examination of gastrointestinal tract and associated organs. mortality pattern study with relation to digestive disorders, gross pathology and histopathology. Bacteriological and parasitic studies were also carried out to investigate the etiology of gastrointestinal tract disorders. Generalized clinical signs such as weakness, anorexia, diarrhoea, dehydration, tympany and abdominal pain and distention were observed in affected animals. Maximum mortality was observed in more than two-year age group (47.7%) and in calves of upto 6 months age group (38.5%). Main gastrointestinal tract disorders affecting ruminants were acute necro-hemorrhagic type or chronic/granulomatous in nature involved with multiple etiological factors as bacterial infections (Escherichia coli and other opportunistic pathogens), paratuberculosis (Johne's disease), coccidiosis and mixed parasitic infections of Strongyle spp., Strongyloides spp. and Buxtonella sulcata complicating the pathology of such disorders.

Keywords: Gastrointestinal tract, pathology, enteritis, hepatitis, lymphadenitis, ruminant

Introduction

Gastrointestinal tract disorders in ruminants are one of the major concerns to veterinarians as well as livestock owners and are usually reported throughout the year that led to high mortality and morbidity among ruminants. The system includes the digestive tract and its associated glands with main function as digestion, absorption and assimilation of nutrients. It is one of the largest and heaviest organ systems of ruminants which normally contain a mixture of commensal and potentially pathogenic organisms. During healthy condition there is continuous interaction between the host and microorganisms resulting in a continuous steady state. The common conditions affecting the gastrointestinal tract are traumatic reticuloperitonitis, diaphragmatic hernia, abscesses, adhesions, impactions, dilatations and motility disturbances which are discernible by clinical signs like anorexia, dullness, recurrent or persistent tympany and abdominal distension (Chanie and Tesfaye, 2012) ^[4].

Most of these conditions are very intricate to distinguish from each other. According to Sharma *et al.* (2015) ^[23], more than 15 percent of all the natural deaths in dairy and beef animals are due to abomasal diseases and resulting peritonitis which are the part of forestomach disorders complex. The gastrointestinal disorders may also occur due to multiple infectious or non-infectious etiological factors including parasites, bacteria, viruses, fungi, foreign bodies (nails, wires, polythene bags etc) and neoplastic conditions (Zachary *et al.*, 2012) ^[30]. There is very limited literature available on comprehensive studies on gastrointestinal tract disorders affecting ruminants. Therefore, the present work was planned to aim at etio-pathomorphological investigation of gastrointestinal tract disorders affecting large and small ruminants.

Materials and Methods

The current investigation was carried out on 65 cases (bovine, ovine and caprine) brought to the Department of Veterinary Pathology, LUVAS, Hisar for post-mortem examination for six months duration i.e. from August, 2019 to January, 2020.The complete history of the cases was recorded including the species, clinical signs and symptoms and treatment if given to the clinically affected animals.

Pathological studies included detailed gross and histopathological examination of gastrointestinal tract organs at the time of post-mortem examination. The ruminant carcasses were divided into different age groups (I-V) to study the age wise mortality pattern. During post-mortem examination forestomach, abomasum, intestine, mesenteric lymph nodes and liver were thoroughly examined and pathological changes were classified according to the lesions observed. For histopathological examination, representative tissue pieces showing lesions were collected in 10% neutral buffered formalin and processed by standard paraffin embedding technique and stained with routine haematoxylin and eosin stain (Luna, 1968) ^[8]. Bacteriological and parasitic studies were carried out to investigate the etiology of gastrointestinal tract disorders. Bacteriological isolations were attempted from the heart blood, liver and mesenteric lymph nodes of all the animals. Identification and biochemical characterization of each bacterial isolate was done by using Vitek-2 system (Bauer et al., 1966)^[1]. Parasitological examination was carried out by standard floatation and sedimentation techniques (Soulsby, 1982)^[26].

Results and Discussion

Generalized clinical signs such as weakness, anorexia,

diarrhoea, dehydration, tympany, abdominal pain and distention were observed in most of the carcasses. Age, species and sex wise distribution of mortality in ruminants is presented in table 1. Age-wise distribution of mortality revealed maximum mortality in more than 2 years age group (47.7%) and young ones of up to 6 months age group (38.5%). Sex-wise mortality data revealed that mortality in females (buffalo, cattle and sheep) was higher than their male counterparts. Month-wise mortality distribution revealed maximum in the month of September (29.2%) followed by December, October, November, January and August. McConnel *et al.* (2010) ^[10] and Londhe (2010) ^[7] also reported higher mortality in animals above one year of age in bovine.

They suggested that higher mortality in adult animals might be attributed to the stage of their high production and high risk of suffering from traumatic and infectious diseases. A relatively high mortality rate in younger calves might be due to inadequate management or due to improper feeding regime after withdrawal of milk. Higher mortality in female is due to more reports of death as compared to male because farmers are more interested in keeping females (Sushma et al., 2016) [27]. The month wise mortality is presented in table 2 and which revealed that maximum mortality was in the months of September (29.2%) followed by December (23.1%), October (21.5%), November (16.9%), January (6.2%) and August (3.1%). More or less similar results have been reported by Lehreena (2008) [6] and Londhe (2010)^[7]. High mortality in above said month might be due to seasonal variations which makes animals particularly the voung calves susceptible to many infectious diseases and pathological conditions due to seasonal stress.

Table 1. Percent ac	e-wise	species and	sex-wise mortali	ity in	ruminants affected	with	gastrointestinal	tract disorders
Table 1. I ciccin ag	30-wise,	species and	sex-wise mortan	ity m	runnants arrected	with	gasuomusunai	tract disorders

(n=65)															
	A = 2	Animals													
Groups	Age	Buffalo		Cattle		Tatal Sheep		T . 4 . 1	Goat		T. 4 - 1	Grand total with			
	(mus)	Μ	F	Total	Μ	F	Total	Μ	F	Total	Μ	F	Total	percentage	
Ι	0-6	11	7	18	2	3	5	0	0	0	2	0	2	25 (38.5%)	
II	6-12	0	1	1	0	0	0	1	1	2	0	0	0	3 (4.6%)	
III	12-18	0	2	2	1	0	1	0	2	2	0	0	0	5 (7.7%)	
IV	18-24	0	0	0	0	0	0	1	0	1	0	0	0	1 (1.5%)	
V	≥24	1	16	17	0	4	4	0	7	7	1	2	3	31 (47.7%)	
Т	otal	12	26	38	3	7	10	2	10	12	3	2	5	65 (100%)	
%	age	18.5	40	58.5	4.6	10.8	15.4	3.1	15.4	18.5	4.6	3.1	7.7	100	

 Table 2: Percent month wise mortality in ruminants affected with gastrointestinal tract disorders

(n=65)

Times a suited		Mortal	D emonstrate $(0/)$					
Time period	Buffalo Cattle Sheep Goat		Goat	Total	Tercentage (70)			
August, 2019	1	1	0	0	2	3.1		
September, 2019	13	2	3	1	19	29.2		
October, 2019	10	2	1	1	14	21.5		
November, 2019	8	2	1	0	11	16.9		
December, 2019	6	1	6	2	15	23.1		
January, 2020	0	2	1	1	4	6.2		
Total cases	38	10	12	5	65	100		

The main gross and histopathological lesions observed in gastrointestinal tract organs are presented in Table 3 and 4, respectively. Most prominent gross lesion affecting forestomach was circulatory disturbances (18.5%) followed by perforated reticulum associated with foreign bodies (7.5%; Fig 1), impaction and tympanic rumen (6.2%). Morwal *et al.* (2019)^[11] also reported similar conditions in buffaloes at 6-9 year age (most productive age) and concluded that this might be related to the production and sudden dietary changes. Histopathological

examination revealed acute reticulitis (Fig 2) in 20% cases followed by acute omasitis (12.3%; Fig 3), acute rumenitis (7.7%; Fig 4), generalized congestion and hemorrhages (6.2%). Similar to our findings Kumar (2018)^[5] also reported rumenitis, reticulitis and omasitis in ruminants. In support to our findings, acute omasitis has also been reported by Puspha (2013)^[16] and Sharma (2013)^[22]. According to earlier workers rumenitis has been found to be associated with ruminal acidosis (Pacheco and Cruz, 2015; Uzal *et al.*, 2015)^[13, 29] which is most often caused

by sudden changes in diet, the consumption of concentrate which are rich in fermentable carbohydrates (Brust et al., 2015) ^[3]. Present study reported the presence of plastic bags intermixed with feed contents in rumen as major cause of ruminal impaction in bovine carcasses. Berrie et al. (2015) [2] suggested fodder scarcity, environmental contamination and poor standards of animal rearing as major factors that cause animals to ingest and accumulate plastic foreign bodies in their rumen. Problem is more often encountered in bovines as they graze close to the ground making them more vulnerable to ingestion of plastic foreign bodies. Caprine are less susceptible to the ingestion of foreign bodies because of their well-developed sensitive prehensile organs and selective feeding behaviour. The penetrating sharp foreign bodies that can accumulate in the reticulum cause focal abscessation, peritonitis, diaphragmatic and pericardial perforation which is called as traumatic reticulopericarditis. Similar to our findings, Ramprabhu et al. (2003) ^[18] also reported incidence of 23.38% in buffalo suffering from traumatic reticuloperitonitis. Similar to our observation Radostits et al. (2007)^[17] stated that omasum is involved in many gastrointestinal tract disorders but clinical signs and symptoms due to omasal dysfunction are seldom observed and primary diseases of the omasum, such as omasal impaction and omasitis are rare. Major lesions observed in abomasum were vascular changes (congestion, hemorrhages and edematous mucosal folds) in 23 (35.4%) cases followed by mucosal ulcers in 7 (10.8%) cases. Histopathological examination revealed abomasitis in 29 (44.6%) cases followed by vascular changes in 12 (18.5%) cases and ulcerative lesion in 2 (3.1%) cases. Kumar (2018)^[5] and Tiwari et al. (2018)^[28] also reported the similar pathological findings in abomasum. Abomasal ulcers in ruminants have also been elucidated to be associated with stress, concentrated rations and concurrent diseases (Rebhun, 1995)^[20]. Gross lesions observed in intestine were vascular changes (46.2%) followed by transverse folds/corrugations (13.8%), catarrhal exudate in lumen (9.2%) and reddish necrotic mucosa (9.2%; Fig.5). Seema et al. (2004)^[21] also reported the vascular changes in bovine carcasses. Similar corrugations and

thickening of the intestinal mucosa have been also reported by Singh *et al.* (2017)^[24] in goats with subclinical paratuberculosis. Histopathological examination of intestine revealed necrohemorrhagic enteritis (Figs. 6) in 43 (67.7%) cases followed by chronic granulomatous enteritis (Fig. 7) in 9 (13.8%), catarrhal enteritis (Fig. 8) in 7 (10.8%), fibrino-necrotic enteritis in 3 (4.6%) and suppurative enteritis in 2 (3.1%) cases. In corroboration to our findings, Marodia (2017)^[9] and Poonia (2018)^[15] also reported similar changes in intestines. Similar results regarding granulomatous enteritis were reported by Perez *et al.* (2005)^[14] and Siyaseelan *et al.* (2011)^[25].

The mesenteric lymph nodes in the present study showed enlargement due to vascular changes (congestion and hemorrhages), necrotic foci and induration with caseation in 18 (27.7%), 13 (20%) and 2 (3.1%) cases, respectively. More or less similar results were also reported by Rano *et al.* (2012) ^[19] and Tiwari *et al.* (2018) ^[28]. Histopathological findings revealed acute lymphadenitis (Figs. 9) in 15 (23.1%) cases, chronic lymphadenitis in 14 (21.5%) cases, necrotic lymphadenitis in 6 (9.2%) cases, circulatory disturbances as well as parasitic lymphadenitis in one (1.5%) case. More or less similar findings in mesenteric lymph nodes were also reported by Sushma *et al.* (2016) ^[27] and Tiwari *et al.* (2018) ^[28].

Gross changes observed in liver were pale/white necrotic areas (Fig. 10) in 17 (26.2%) cases followed by hepatomegaly with congestion and mottling in 16 (24.6%) cases, induration in 14 (21.5%) cases and hydatid cysts in 4 (6.2%) cases. More or less similar findings were also reported in bovine by Tiwari *et al.* (2018) ^[28] and in ovine by Mundotiya (2017) ^[12]. Sushma *et al.* (2016) ^[27] also reported induration and hepatomegaly in case of bovine, ovine and caprine. Histopathology of liver revealed degenerative changes in hepatocytes in 25 (38.5%) cases followed by acute focal hepatitis (Fig. 11) in 16 (24.6%) cases, necrotic hepatitis in 14 (21.5%), circulatory disturbances in 6 (9.2%) and cirrhosis (Fig. 12) in 2 (3.1%) cases.

	Gross lesions	Buffalo	Cattle	Sheep	Goat	Total	Percentage
	Vascular changes (Congestion & Hemorrhages)	6	3	2	1	12	18.5
	Ruminal Impaction	3	0	1	0	4	6.2
Fore-stomach	Perforated reticulum (Foreign body syndrome/Traumatic reticuloperitonitis)	5	0	0	0	5	7.7
	Tympanic rumen	2	0	0	0	2	3.1
	Total	16	3	3	1	23	
	Percentage (%)	42.1	30	25	20	35.4	
	Vascular changes (Congestion & Hemorrhages with edematous mucosal folds)	10	3	8	2	23	35.4
Abomasum	Abomasal ulcer	6	0	1	0	7	10.8
Aboinasum	Total	16	3	9	2	30	
	Percentage (%)	42.1	30	75	40	46.2	
	Vascular changes (congestion and hemorrhages)	20	2	7	1	30	46.2
	Catarrhal exudate in lumen	3	2	0	1	6	9.2
Tutostino	Necrotic mucosa	1	2	1	2	6	9.2
Intestine	Transverse folds/corrugations	8	1	0	0	9	13.8
	Total	32	7	8	4	51	
Abomasum Intestine Mesenteric Lymph nodes	Percentage (%)	84.2	70	66.7	80	78.5	
	Enlargement with vascular changes (congestion and hemorrhages)	13	3	1	1	18	27.7
	Enlarged and necrotic foci	7	2	2	2	13	20.0
Mesenteric Lymph	Induration and caseation	0	0	2	0	2	3.1
nodes	Total	20	5	5	3	33	
	Percentage (%)	52.6	50	41.7	60	50.8	
	Hepatomegaly associated with congestion and mottling	10	3	2	1	16	24.6
	Pale/white necrotic areas	9	2	4	2	17	26.2
T in an	Induration with distended gall bladder	11	1	2	0	14	21.5
Liver	Hydatid cysts	3	0	1	0	4	6.2
	Total	33	6	9	3	51	
	Percentage (%)	86.8	60.0	75.0	60	78.5	

Table 3: Gross pathological lesions observed in gastrointestinal tract of ruminants

0			Anim	T (1	D		
Organs	Parameters	Buffalo	Cattle	Sheep	Goat	Total	rercentage
	Vascular changes (Congestion & Hemorrhages)	2	1	1	0	4	6.2
	Rumenitis	2	2	1	0	5	7.7
Γ	Reticulitis	9	1	2	1	13	20.0
Equatomoch	Haemorrhagic reticulitis with hyperkeratosis	1	0	0	0	1	1.5
Forestomach	Omasitis	4	1	2	1	8	12.3
Γ	Haemorrhagic omasitis with hyperkeratosis	0	1	1	0	2	3.1
Γ	Total	18	6	7	2	33	
Γ	%age	47.4	60	58.3	40	50.8	
	Vascular changes (Congestion & Hemorrhages)	10	2	0	0	12	18.5
Ē	Abomasitis	14	1	11	3	29	44.6
Abomasum	Necrotic abomasitis	1	1	0	0	Total Percentage 4 6.2 5 7.7 13 20.0 1 1.5 8 12.3 2 3.1 33	
	Total	25	4	11	3	43	
F	%age	65.8	40	91.7	60	66.2	I Percentage 6.2 7.7 20.0 1.5 12.3 3.1 3 3.1 3 3.1 3 18.5 44.6 3.1 2 10.8 67.7 4.6 3.1 13.8 0 3.1 23.1 21.5 9.2 3.1 18.5 1.5 9.2 3.1 18.5 1.5 9.2 3.1 18.5 1.5 9.2 38.5 24.6 21.5 3.1 3.1
	Catarrhal enteritis	4	1	2	0	7	10.8
Intestine	Necro-haemorrhagic enteritis	28	6	7	3	44	67.7
	Fibrino-necrotic enteritis	2	1	0	0	3	4.6
	Suppurative enteritis	0	0	2	0	2	3.1
	Granulomatous enteritis	4	2	1	2	9	13.8
	Total	38	10	12	5	65	
	% age	100	100	100	100	100	
	Circulatory disturbances (Congestion & Hemorrhages)	2	0	0	0	2	3.1
Forestomach Abomasum Intestine Mesenteric lymph nodes Liver	Acute lymphadenitis	7	3	3	2	15	23.1
	Chronic lymphadenitis	8	2	3	1	14	21.5
F	Necrotic lymphadenitis	3	0	2	1	Total Percentage 4 6.2 5 7.7 13 20.0 1 1.5 8 12.3 2 3.1 33 50.8 12 18.5 29 44.6 2 3.1 43 66.2 7 10.8 44 67.7 3 4.6 2 3.1 9 13.8 65 100 2 3.1 15 23.1 15 23.1 15 23.1 14 21.5 6 9.2 2 3.1 12 18.5 1 1.5 52 80 6 9.2 25 38.5 16 24.6 14 21.5 2 3.1	
Mesenteric lymph nodes	Parasitic lymphadenitis	0	1	1	0	2	otal Percentage 4 6.2 5 7.7 13 20.0 1 1.5 8 12.3 2 3.1 33 60.8 12 18.5 29 44.6 2 3.1 43 6.2 7 10.8 44 67.7 3 4.6 2 3.1 9 13.8 65 100 2 3.1 15 23.1 14 21.5 6 9.2 2 3.1 12 18.5 1 1.5 52 80 6 9.2 25 38.5 16 24.6 14 21.5 2 3.1 63 97
	Chronic Granulomatous lymphadenitis	8	2	1	1	12	18.5
Ē	Acute Suppurative lymphadenitis	0	0	1	0	1	1.5
Ē	Total	28	8	11	5	52	
Γ	% age	73.7	80	91.7	100	80	
	Circulatory disturbances (Congestion & Hemorrhages)	3	2	1	0	6	9.2
Ē	Degenerative changes	17	2	4	2	25	38.5
Ē	Focal acute hepatitis	7	4	2	3	16	24.6
Liver	Necrotic hepatitis	10	1	3	0	14	21.5
	Cirrhosis	0	1	1	0	2	3.1
	Total	37	10	11	5	63	
F	%age	97.4	100	91.7	100	97	

Table 4: Histopathological lesions observed in gastrointestinal tract of ruminants



Fig 1: Perforated reticulum with necrosed tract (arrow) due to penetration of pointed objects through reticulum (Adult buffalo)



Fig 2: Acute hemorrhagic reticulitis characterized by extensive hemorrhages (arrow) in mucosa (Adult buffalo, *Micrococcus luteus*) H&E 400X



Fig 3: Acute hemorrhagic omasitis characterized by necrosis of mucosal epithelium (arrow), hemorrhages and infiltration of leucocytes (Adult sheep, *Streptococcus spp.*) H&E 200X



Fig 4: Rumenitis characterized by necrosis of mucosal epithelium (arrow) and infiltration of leucocytes in epithelial mucosa (Cattle-calf) H&E 100X



Fig 5: Hemorrhagic intestine with frank blood in the intestinal lumen and congested mucosa (Adult buffalo)



Fig 6: Necrohemorrhagic enteritis with severe hemorrhages in mucosa along with infiltration of leucocytes (Adult buffalo, *Micrococcus luteus* infection) H&E 200X



Fig 7: Granulomatous enteritis characterized by thickened mucosa with infiltration of epitheloid cells, macrophages, lymphocytes in sub mucosa (Cattle calf, MAP positive) H&E 200X



Fig. 8: Catarrhal enteritis characterized by hyperplasia of goblet glands and infiltration of leucocytes in mucosa (Adult sheep, *Strongyle spp.* infection) H&E 200X

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Fig. 9: Acute lymphadenitis characterized by congested blood vessels, necrosis of lymphocytes and infiltration of neutrophils in cortex and medulla (Goat Kid) H&E 100X



Fig 10: Pale indurated liver with white necrotic areas (arrow) and distended gall bladder (Adult buffalo)



Fig 11: Focal hepatitis characterized by focal coagulative necrosis of hepatocytes (arrow) and infiltration of leucocytes in liver parenchyma (Cattle calf, *Kocuria spp.*) H&E 200X



Fig. 12: Cirrhosis characterized by diffuse proliferation of fibrous tissue (arrow) and atrophy of hepatic chords in liver parenchyma. (Adult buffalo, *Micrococcus luteus* infection) H&E 200X

Microbiological studies in present study revealed isolation of different bacterial organisms from 47 cases. Escherichia coli (33.8%) was the most prominent bacterial species isolated followed by Kocuria spp. (K. kristinae, K. rosea), Sphingomonas paucimobilis, Pseudomonas mendocina, Enterococcus spp. (E. faecalis, E. faecium), Stenotrophomonas multophila, Streptococcus spp. (S. alactolyticus, S. uberis, S. gallolyticus ssp gallolyticus), Staphylococcus spp. (S. lentus, S. warneri, S. hemolyticus, S. aureus), Proteus mirabilis, Salmonella group, Micrococcus luteus, Pseudomonas spp. (P. aeruginosa, P. oleovarans). Most of the newly isolated bacteria in the present study belong to opportunistic pathogen group. The parasitological studies revealed parasitic infestations in 30 cases with maximum cases of coccidiosis (*Eimeria spp.*; 15) followed by mixed parasitic infections (10), *Strongyle spp.* (3), *Strongyloides spp.* (1) and *Buxtonella sulcata* (1). Buffalo carcasses revealed parasitic infections in 63.3% cases, ovine in 23.3% cases, cattle in 6.7% cases and caprine in 6.7% cases.

Parasitic conditions were mainly found affecting young bovine calves as compared to adults; while adult sheep and goat were found to be affected more than the young ones.

It is concluded that mortality due to gastrointestinal disorders among large and small ruminants was found to be maximum in adult of more than two years and calves of up to 6 months of age. Main conditions found were acute necro-hemorrhagic or chronic granulomatous type and were found associated with *Escherichia coli* along with other secondary opportunistic pathogens as well as mixed parasitic conditions.

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