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CLINICAL AND LABORATORY EVALUATION OF RUMINORETICULAR DISORDERS IN CATTLE

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ABSTRACT

Diseases of ruminant's stomach cause huge economic losses. Therefore, the present study was carried out to evaluate various ruminoreticular disorders and their effects on hematology and biochemical changes in blood and ruminal fluid as well as their effects on the ruminal protozoal activity and population in cattle. A total number of 46 cattle of both sexes (31 diseased cattle and 15 healthy control one) were included in the study. Cases were admitted at Veterinary Teaching Hospital, Faculty of Veterinary Medicine, Assuit University, Assuit, Egypt during the period between July 2015 and November 2016. These animals were subjected to thorough clinical, ferroscopic, radiographical examination surgical exploration and laboratory investigation and divided into: TRP (n=7), vagus indigestion (n=5), impaction (n=5), free gas tympany (n=9) and frothy tympany (n=5). The results of this study showed that cases of TRP were suffered from obvious signs of pain, jugular pulsation in some animals. Ferroscopic examination was positive and X-rays showed presence of metal foreign objects. Hematological analysis showed significant decrease ($P<0.01$) in TRBCs count and Hb concentration and significant increase ($P<0.01$) in TWBCs and neutrophils counts. Serum biochemical analysis showed significant increase in total protein ($P<0.01$), globulin ($P<0.05$), and urea ($P<0.05$) concentration while there was significant decrease in the concentration of albumin, calcium ($P<0.01$), inorganic phosphorus ($P<0.05$), sodium ($P<0.05$) and zinc ($P<0.01$). Analysis of ruminal juice showed a dramatic decrease ($P<0.01$) in the activity and total number of protozoa, potassium, sodium and inorganic phosphorus concentrations. Animals with vagus indigestion showed characteristic abdomen distension (L-shape). Metal detector was negative in 2 animals and positive in 3 which were confirmed by X-rays. Hematological analysis showed significant decrease ($P<0.01$) in hemoglobin concentration with significant increase ($P<0.01$) in TWBCs, neutrophils and eosinophils counts. Serum biochemical analysis showed a significant increase ($P<0.01$) in the concentration of total protein globulin and urea and the activities of ALP, GGT with significant decrease in albumin ($P<0.01$) chloride ($P<0.01$) and inorganic phosphorus ($P<0.05$). Ruminal fluid analysis showed sharp decline in number and movement of protozoa and highly significant increase in chloride ($P<0.01$) and calcium ($P<0.05$) and significant decrease (($P<0.01$) inorganic phosphorus concentrations. Animals with Impaction showed ruminal atony and slight decrease in body temperature. Hematological analysis revealed highly significant increase ($P<0.01$) in TWBCs, neutrophils counts and significant increase ($P<0.05$) monocytes count. Serum biochemical analysis showed significant increase ($P<0.05$) in urea, lactate ($P<0.01$) and sodium ($P<0.05$) while there were significant decrease ($P<0.01$) in potassium and inorganic phosphorus. Analysis of ruminal fluid showed highly significant decrease ($P<0.01$) in number and motility of protozoa, sodium, calcium and significant decrease ($P<0.05$) in potassium, and highly significant increase ($P<0.01$) in lactate concentration. Cases with free gas tympany were characterized by severe abdominal distention, anorexia, rapid heart rate and decrease rumen motility. Ferrosopic examination was negative in 6 animals and false positive in 3 animals which is confirmed by using X-rays. Hematological analysis revealed highly significant increase ($P<0.01$) TWBCs, neutrophil and significant increase ($P<0.05$) in eosinophil counts. Serum biochemical analysis showed significant increase in the concentration of globulin ($P<0.05$), urea ($P<0.01$) and sodium ($P<0.05$), and in the activities of ALP, GGT ($P<0.01$). Rumen fluid analysis showed highly significant decrease ($P<0.01$) in the total number and activity of ruminal protozoa and sodium and significant decrease ($P<0.05$) in ruminal potassium. Frothy tympany was characterized by severe abdominal distention and ruminalatony. Examined cases of frothy tympany were negative by metal detector and X-ray. Hematological analysis showed highly significant increase ($P<0.01$) in neutrophils count. Serum biochemical analysis showed highly significant increase ($P<0.01$) in the concentration of globulin and activities of ALP, GGT and significant increases ($P<0.05$) in serum urea and sodium ($P<0.05$). Rumen fluid analysis showed sharp decrease in the number and motility of protozoa. Ruminal biochemical analysis showed significant decrease ($P<0.01$) in sodium, potassium, inorganic phosphorus and calcium levels. In conclusion, diagnosis of ruminoreticular disorders depends collectively on history, clinical and ferroscopic examination. Radiographic examination is an important tool for differential diagnosis between traumatic and non-traumatic ruminoreticular disorders. Hematological and biochemical examination of serum and ruminal juice were of additional values in discriminating between various ruminoreticular disorders in cattle.

Key words: Ruminants, Disease, Rumen, Reticulum, TRP, Vagus, Impaction, Tympany

INTRODUCTION

Ruminants represent one of the most populous and economically important groups of animals in the world (Craig, 1981; Guffra *et al.,* 2000 and Bradford *et al.,* 2003). They are great contributors to the human food chain due to their ability to utilize complex polysaccharides in plant cell walls (cellulose, hemicellulose and pectin) and turn these into meat and milk for human consumption. As the health of ruminants depend critically on the health of their rumen and on efficient ruminal metabolism, therefore, ruminal dysfunctions are responsible for huge economic losses in cattle industry. These losses could largely be attributed to decrease in production and increase in expenditure towards treatment (Enemark, 2008; Plaizier *et al.,* 2009; Steele *et al.,* 2009; Kahn, 2011 and Kirbas *et al.,* 2014).

Various diseased conditions are encountered due to disorders of forestomach. They are classified into traumatic and non-traumatic indigestion (Radostits *et al.,* 2007). Traumatic affection of the bovine forestomach due to ingestion of sharp foreign is one of common ruminoreticular dysfunctions, particularly in developing countries. The bovine species does not have sensitive prehensile organs that discriminating sense of taste. As a consequence, cattle and buffaloes kept in farm yards, stables or at other sites close to human mechanical activities are prone to swallow metallic objects such as nails and pieces of wires that have been carelessly left in their feeding areas (Jones *et al.,* 1997 and Desiye and Mersha, 2012). Ingestion of non- dietary materials causes various problems in rumen and reticulum including ruminitis, vagus indigestion, and traumatic reticuloperitonitis/ traumatic pericarditis (TRP/TP) (Radostits *et al.,* 2007). On the other hand, dietary changes and improper feeding management also cause several forms of indigestion such as tympany and impaction which cause severe economic loss in cattle industry.

Most of ruminoreticular disorders have common history including change in the appetite (inapetance to anorexia), change in milk production (mild to sharp drop of milk production) and altered rumination process (reduction to cessation), thus diagnosis and differential diagnosis of ruminoreticular disorders arecomplex procedure that imply adaptation of various clinical and laboratory skills. Therefore, the present study aimed to evaluate clinically various ruminoreticular disorders and their effects on hematology and biochemical changes in blood and ruminal fluid as well as their effects on the ruminalprotozoal activity and population in cattle.

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MATERIALS AND METHODS

Animals: A total of 31 cattle were admitted to the Veterinary Teaching Hospital (VTH) at Assiut University- Egypt with one or more of the following history: inappetence, anorexia, absence of rumination, tympany, decrease of milk production, and loss of body weight from July 2015 to November 2016. Additionally, 15 healthy animals were included in this study as a control.

Samples

Blood Samples: Blood samples were collected from jugular vein on a clean dry vaccutanier tubes containing EDTA as anticoagulant for hematological analysis. Another blood samples were collected on clean and dry plain centrifuge tubes and allowed to flow freely and gently from the jugular vein over the inner surface of the tube, and kept in the refrigerator at 4°C for 30 min and then centrifuged at 3000 rpm for 15 min for separation of serum (Coles, 1986). The collected sera were transferred into Eppendorff tubes, which were coded and kept at – 20 °C up to the time of analysis.

Ruminal Juice Samples: Ruminal juice samples were collected directly from the bottom of the rumen in a clean and dry flask during rumenotomy or by using stomach tube, with pump and suction. A tranquilizer was administered to excited animals before collection (Hajikolaei *et al*., 2006; Bramley *et al*., 2008 and Abo-Donia *et al*., 2011). Ruminal juice samples were subjected to analysis immediately after collection. Samples were sieved and then divided into four parts. The first part was used to evaluate the physical character of ruminal juice as color, odour, consistency (Dirksen, 1969). The second part was examined microscopically to evaluate motility of microfauna (Misra and Singh, 1974). The third part was used to assess the total protozoal count. The fourth part was centrifuged at 3000 rpm and then the supernatant were transferred into Eppendorff tubes, which were coded and kept at – 20 °C up to the time of biochemical analysis according to Coles (1986).

History and Clinical Examination: Animals were subjected to thorough clinical examination according to Cockcroft (2015). Animal's and owner's data as well as clinical findings were recorded on specific clinical chart. Appetite, milk yield, general attitude, pain expression (grunting, tearing, tongue protrusion), eye appearance (conjunctival mucous membranes and sclera blood vessels), body temperature, heart rate and rhythm, pulsation and filling of jugular vein, abdominal distention and ruminal movement were thoroughly investigated.

Ferroscopic Examination: All admitted cases were screened by guardian metal detector (Hand hold security metal detector, USA) for tentative diagnosis

of swallowed metal foreign body. A metal detector was applied over the ventral and ventrolateral aspect of caudal chest and cranial abdomen for detection of ferromagnetic foreign bodies.

Radiographic Examination: Suspected cases of swallowed metal foreign bodies based on clinical and ferroscopic findings were further subjected to radiographic examination using fixed radiographic apparatus (Philips, Super 80 CP). Radiography was performed as lateral plain radiography to the caudal thoracic and cranial abdominal region in recumbent position.

Surgical Exploration: Exploratory laparotomy and rumenotomy were carried out on suspected cases of TRP with the help of staff members of Animal Surgery Department, Faculty of Veterinary Medicine, Assiut University, Egypt).

Hematological Analysis: Total red blood cell count (TRBCs $x10^6$ /mm^3) and total white blood cell count (TWBCs $x10^3$/mm^3) were determined manually using the Hemocytometer method (Coles 1986). Hemoglobin concentration (g/dl) was determined using Acid Hematin (Sahli's) method. Differential leukocytic count was determined using four field meander method (Kelly, 1984).

Biochemical Analysis of Serum and Ruminal Juice: Serum biochemical analysis including level of total protein (g/dl), and albumin (g/dl), and activities of alkaline phosphatase (ALP, U/L), gamma glutamyl transpeptidase (GGT, U/L) and alanine aminotransferase (ALT, U/L), and concentration of serum urea (mg/dl), creatinine (mg/dl), sodium (mmol/l, potassium (mmol/l), chloride (mmol/l), calcium (mg/dl), inorganic phosphorus (mg/dl), magnesium (mg/dl), copper (µg/dl), zinc (µg/dl) and lactate (mg/dl) levels were determined. Ruminal biochemical analysis including levels of sodium (mmol/l), potassium (mmol/l), chloride (mmol/l), calcium (mg/dl), phosphorus (mg/dl) and lactate (mg/dl) levels. All estimated biochemical parameters were determined spectrophotometrically (Optizen 3220 UV, Mecasys Co. Ltd, Korea) using diagnostic test kits ((Spectrum Diagnostics, Cairo, Egypt and Spinreact, Spain). Manufacturer's instructions were followed. Serum globulins were calculated mathematically by subtracting albumin level from total protein level according to Coles (1986).

Physical examination of ruminal juice: Ruminal juice samples were examined for color, odor and consistency according to Abdel-Salam (1981).

Microscopic examination of ruminal protozoal activity: Microscopic examination of ruminal microfauna was carried out according to Fouda (1995). Briefly, one drop of strained ruminal juice was taken on a clean warmed slide and examined immediately under the microscope. The activity of the protozoa was evaluated as followings: Highly motile and very crowded (+++), motile and crowded (++), sluggish motility and low number (+) and no motility or sporadic alive infusoria (-).

Determination of total ruminal protozoa count: Total count of ruminal protozoa was carried out according to Wang *et al*. (2009). Briefly, ruminal juice sample was thorough mixed and diluted 5 times with 15 ml saline solution and 5 ml lugol's iodine solution. The iodine solution was added to kill and stain the protozoa cell. Immediately after gentle shaking, 1 ml was taken up by plastic pipette. Exactly 0.1 ml was taken on a dry clean slide as quickly as possible. The 0.1 ml volume taken on the slide was carefully spread under a cover glass of 1100 squares millimeter area (22 x 50). Care was needed when fitting the cover on the slide to avoid formation of vacuoles or the escape of some liquid out. Counting was carried out using low power (10 x). The field area of that lens was one square millimeter. In each slide 30 fields were counted. The average counts in 30 fields represented the protozoal count per one square millimeter area of the field. Multiplying this figure by 1100 which is the area of the cover give the protozoal count in 0.1 ml of the diluted samples, and represent 0.02 ml of the original sample. The protozoal count per ml. ruminal content was obtained by multiplying this value by 50.

Overall Total Protozal Count (TPC/ml) = TPC in 30 fields × 1100 × 50

Statistical Analysis: Statistical analysis of the obtained data was done by using statistical package for the Social Sciences for Windows (SPSS, version 16.0) according to Borenstein *et al*. (1997). Data were presented as mean and standard error. Data from diseased cattle were compared with control group using ANOVA. Statistically significant differences were determined at $P \leq 0.01$ (Highly significant) and $P < 0.05$ (Significant).

RESULT

Case History and Clinical Signs: Case history of the admitted cases including the following complains: inappetence, anorexia or depraved appetite, absence of rumination, abdominal distension, regurgitation of food, various degrees of reduction in milk production and loss of body weight. Dietary changes and improper feeding management were also recorded in the history. The recorded body temperature and heart rate showed no significant change however there was slight increase in the body temperature in case of TRP and slight decrease in impaction. There was marked reduction in the rate of ruminal cycle in all admitted case with complete cessation of ruminal movement in impaction. Tachycardia was reported in some cases of TRP and free gas tympany. Pain expression (tearing),

true jugular pulsation/filling and congestion of conjunctival mucous membranes were evident in TRP/TP cases (n=7) (Figures 1-2). Cases of vagus indigestion (n= 5) showed distension of the upper left and lower right of the abdomen (L-shape abdominal distension) (Figure 3) while free gas tympany (n= 9) showed severe left abdominal distension (Figures 4&5). Trocarization of left flank in cases of free gas tympany revealed expulsion of large amount of gas (Figure 6). Animals with frothy typmany (n=5) showed distension of the whole left flank while impaction (n=5) showed mild left flank distension. The recorded clinical signs in animals under investigation were summarized in Table 1.

Ferroscopic and Radiographic Examinations: A total number of 31 animals were examined by guardian metal detector (Hand hold security metal detector, U.S.A. made) and. X- ray (Table 2). Radiographic examination revealed differentiated and non-differentiated radio-opaque foreign bodies within the reticulum (Figure 7) in 10 cases.

Rumenotomy Findings: Operations were performed tosuspected cases of TRP (7 positive X-ray) and vagus indigestion (3 positive X- ray plus 2 negative but suspected) and various foreign bodies (penetrating and floating) including nails, robes, keys, stones, coins, needles and wire pieces were extracted (Figure 8) confirming TRP (n =7) and vagus indigestion (n =5).

Hematological Findings: Cases of TRP showed significant decrease (P<0.01) in total R.B.C.s count and hemoglobin concentration in TRP while there were significant increase (P<0.01) in TWBCs and neutrophils counts. Animals suffered from vagus indigestion, free gas tympany and impaction showed significant increase (P<0.01) in TWBCs and neutrophils counts. Eosinophils count showed significant increase (P<0.01) in cattle suffered from vagus indigestion, free gas tympany and frothy tympany (P<0.05) while monocytes count showed significant increase (P<0.05) in impaction. Lymphocytes count revealed significant decrease in cattle suffered from frothy tympany (P<0.01) and vagus indigestion (P<0.05). Data of the mean values (±SE) of the hematological parameters in admitted cases of ruminal disorders were presented in Table 3.

Serum Biochemical Analysis

Serum total protein level (g/dl): There was a significant increase (P<0.01) in cattle suffered from TRP and vagus indigestion.

Serum globulins level (g/dl): There was a significant increase in cattle suffered from TRP, free gas tympany (P<0.05), frothy tympany and vagus indigestion (P<0.01).

Table 1: Clinical Findings of Ruminal Disorders in Cattle

Parameter	Control (n=15)	T.R.P (n=7)	Vagus indigestion (n=5)	Impaction (n=5)	Free gas tympany (n=9)	Frothy Tympany (n=5)
Temp. (^{o}C)	38.5±0.6	39.2±0.65	39.1±0.5	37.7 ±0.6	39.1 ±1.5	38.9±1.2
Heart rate (Beat/min)	68.4±19.7	67.7±10.5	81.6±34	63.8 ±11	79.6 ±32.5	67.8±18.5
Ruminal motility (cycle /2 min)	3-4	1-2	0-1	0	1-3	1-2
Jugular pulsation	-ve	True +ve	-ve	False +ve	-ve	-ve
Mucous membranes	Bright pink	Congested in 3 cases	Normal	Pale in 1 Icteric in 1	Normal	Congested in 1 case
Defecation	Normal	Scanty in 3 Diarrheic in 1	Scanty in 2 No defecation in 3	Scanty in 3 No defecation in 2	Scanty in 3	Scanty in 1
Dehydration	-ve	Mild to moderate	Mild to moderate	Mild to moderate	-ve	-ve
Abdominal distension	-ve	-ve	L- shape distension	Slight left flank distension	Severe abdominal distension	+ve

Table 2: Ferroscopic and Radiographic Findings of Ruminal Disorders in Cattle**.**

Parameter		T.R.P (n=7)	Vagus indigestion (n=5)	Impaction (n=5)	Free gas tympany (n=9)	Frothy Tympany (n=5)
Ferroscopic examination	positive	6	3	0	3	0
	negative	1	2	5	6	5
Radiographic examination	positive	7	3	0	0	0
	negative	0	2	5	9	5

Table 3: Mean Values (±SE) of the Hematological Parameters in Cattle suffered from Ruminal Disorders.

Parameter	Control (n=15)	T.R.P (n=7)	Vagus indigestion (n=5)	Impaction (n=5)	Free gas tympany (n=9)	Frothy Tympany (n=5)
TRBCs count (10^6/mm^3)	8.62±0.49	6.13±0.71**	9.71±1.1	6.47±0.09	8.38±0.76	8.78±0.52
Hb. Concentration (g/dl)	12.3±0.56	9.5±0.88**	8.8±0.49**	12.9±0.94	10.61±0.77	10.6±0.98
TWBCs count (10^3/mm^3)	7.2±0.56	12.9±1.31**	9.31±1.2**	10.78±1.14**	11.34±1.1**	7.51±1.0
Lymphocytes count (10^3/mm^3)	4.83±0.43	3.76±0.71	2.75±0.55*	4.67±0.89	4.13±0.45	2.15±0.75**
Neutrophils count (10^3/mm^3)	2.1±0.19	8.47±1.3**	4.42±0.68**	5.68±1.17**	6.29±0.95**	4.56±0.44**
Eosinophils count (10^3/mm^3)	0.26±0.04	0.26±0.05	1.25±0.82**	0.20±0.01	0.47±0.08*	0.54±0.21*
Monocytes count (10^3/mm^3)	0.16±0.05	0.17±0.02	0.07±0.03	0.34±0.03*	0.42±0.12	0.21±0.6

Data were presented as Mean ±SE (Standard error), *: Significant (P<0.05), **: Highly Significant (P<0.01)

Table 4: Mean Values (±SE) of Serum Biochemical Parameters in Cattle suffered from Ruminal Disorders.

Parameter	Control (n=15)	T.R.P (n=7)	Vagus indigestion (n=5)	Impaction (n=5)	Free gas tympany (n=9)	Frothy Tympany (n=5)
TP(g/dl)	6.94±0.10	7.75±0.35**	7.75±0.35**	6.47±0.31	6.80±0.16	7.15±0.35
Albumin (g/dl)	3.80±0.85	2.41±0.13**	2.75±0.08**	2.59±1.23	2.92±0.15**	2.78±0.20**
Globulin (g/dl)	3.40±0.10	5.35±0.39**	4.87±0.38**	3.88±0.52	3.88±0.27*	4.37±0.48**
ALT(U/l)	46.03±4.14	48.57±5.3	32.44±6.92	46.52±1.84	53.92±10.53	47.04±1.42
ALP(U/l)	89.72±4.30	98.91±5.12	148.28±16.6**	104.08±17.74	131.2±11.37**	155.34±8.68**
GGT (U/l)	16.77±0.85	17.07±2.71	27.58±2.8**	20.03±2.31	23.70±1.37**	22.61±0.64**
BUN(mg/dl)	21.72±1.33	28.79±3.40*	35.29±2.88 **	34.65±3.87**	33.65±2.13**	28.50±3.22*
Creat.(mg/dl)	1.61±0.11	1.20±0.19	1.74±0.10	1.92±0.22	1.64±0.11	1.65±0.1
Na(mmol/l)	155.16±3.02	142.69±5.44*	154.71±6.65	171.70±6.84*	170.05±5.77*	169.88±3.77*
Cl (mmol/l)	110.80±1.35	115.32±4.63	102.38±3.01**	106.07±3.4	106.11±2.90	113.58±4.65
K(mmol/l)	4.23±0.21	4.14±0.22	2.45±0.22*	3.18±0.48*	3.51±0.38	3.77±0.27
Ca(mg/dl)	10.01±0.30	7.47±0.30**	8.65±0.91	10.07±0.99	9.88±0.37	9.81±0.49
Inorganic P (mg/dl)	7.26±0.42	5.50±0.58*	5.46±0.33*	4.33±0.91**	7.10±0.57	7.69±0.52
Mg (mg/dl)	2.21±0.12	2.09±0.25	2.49±0.47	2.60±0.34	1.93±0.27	2.26±0.19
Cu(µg/dl)	117.26±5.44	102.13±10.14	129.33±13.57	112.43±7.36	131.23±3.9	107.26±15.8
Zn (µg/dl)	108.10±3.02	86.52±2.25**	101.87±9.04	103.36±2.86	102.2±2.4	91.43±5.36*
Lactate (mg/dl)	42.16±0.51	43.01±2.63	44.78±4.14	57.92±4.64**	48.66±6.22	35.60±3.3

Data were presented as Mean ±SE, *: Significant (P<0.05), **: Highly Significant (P<0.01)

Table 5: Mean Values (±SE) of Protozoal Count and Biochemical Parameters of Ruminal Juice in Cattle suffered from Ruminal Disorders.

Parameter	Control (n=15)	T.R.P (n=7)	Vagus indigestion (n=5)	Impaction (n=5)	Free gas tympany (n=9)	Frothy Tympany (n=5)
Protozoal count (x10^6)	98.93±4.7	13.40±5.53**	2.99±0.83**	1.94±0.25**	35.79±9.31**	28.2±12.50**
Na (mmol/l)	148.60±2.94	126.30±5.0**	140.01±0.87	116.04±0.0**	128.05±8.0**	122.26±3.5**
Cl (mmol/l)	24.30±1.9	24.08±2.33	59.16±6.**	30.74±7.21	24.51±3.24	30.85±3.31
K (mmol/l)	36.44±1.93	19.20±1.4**	34.87±5.3	26.65±4.8*	27.34±3.9*	24.21±.98**
Ca (mg/dl)	7.40±0.42	7.41±0.5	10.15±1.9*	4.69±0.83**	8.23±0.37	4.02±0.55**
Inorganic P (mg/dl)	17.35±0.9	13.32±1.0**	8.04±2.8**	18.98±0.62	17.01±0.32	11.51±0.28**
Lactate (mg/dl)	1.39±0.08	1.19±0.24	1.84±0.23	2.58±0.61**	1.13±0.24	1.24±0.04

Data were presented as Mean ±SE, *: Significant (P<0.05), **: Highly Significant (P<0.01)

Figure 1: Sign of pain (tearing) in adult cow with TRP

Figure 2: Jugular distension in adult cow with TRP/TP

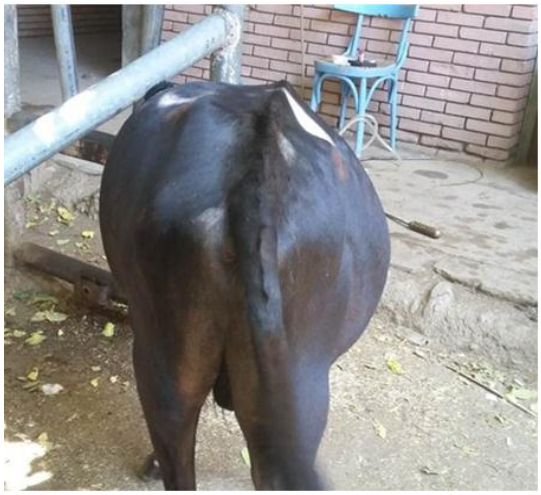

Figure 3: Left and lower right flank distension (L-shape abdomen) in a bull with vagus indigestion

Figure 4: Moderate left flank distension in a yearling calf with free gas tympany

Figure 5: Severe abdominal distension in adult cow with free gas tympany

Figure 6: Trocarization of the rumen in adult cow with free gas tympany

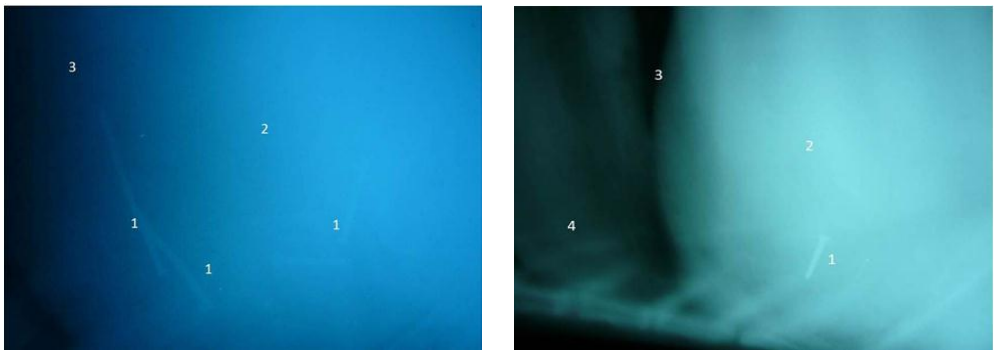

Figure 7: Lateral plain radiography of the cranial abdominal region in adult cow showing radio-opaque metallic foreign bodies-nails (1) within the reticulum (2), diaphragm (3) and heart (4)

Figure 8: Various forms of foreign objects extracted from rumenotomy in cases of TRP including nails, wires, robes, plastic bags, rubber bags, stones, undifferentiated metallic objects**.**

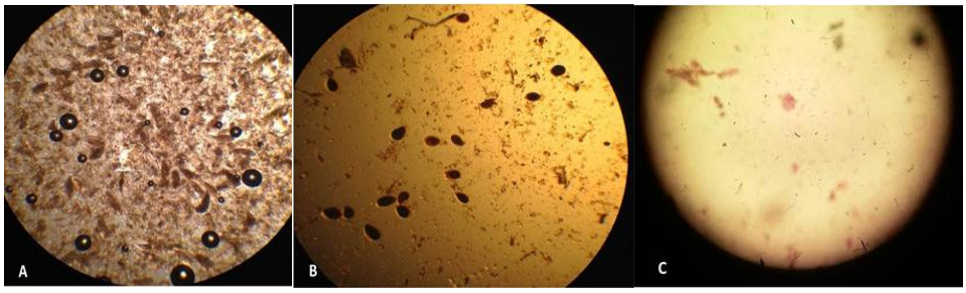

Figure 9: Qualitative evaluation of motility of ruminal protozoa, showing (a) crowded protozoa (++ve), (b) low number of protozoa (+ve), and (c) sporadic ruminal protozoa (-ve) under low power magnification (10x) in adult cow with ruminal disorders.

Liver enzymes: serum alanine aminotransferase (ALT) activity (U/L) showed no significant change ($P<0.05$) in case of TRP, impaction, free gas tympany, frothy tympany and vagus indigestion while serum alkaline phosphatase (ALP) and gamma glutamyl transferase (GGT) activities (U/L) revealed significant increase ($P<0.01$) in cattle with free gas tympany, frothy tympany and vagus indigestion.

Renal function tests: Blood urea nitrogen (BUN) showed significant increase ($P<0.05$) in cattle suffered from TRP and frothy tympany and highly significance increase ($P<0.01$) in impaction, free gas tympany and vagus indigestion while serum creatinine level (mg/dl) revealed no significant change in all cases.

Serum electrolytes Significant decrease in serum sodium ($P<0.05$), chloride ($P<0.01$) and potassium ($P<0.01$) levels (mmol/l) were recorded in cattle with TRP, vagus indigestion and impaction, respectively. Animals with impaction, free gas tympany and frothy tympany showed significant increase ($P<0.05$) in sodium level.

Major elements: Serum calcium level (mg/dl) showed significant decrease ($P<0.01$) in cattle suffered from TRP. Serum inorganic phosphorus

level (mg/dl) revealed significant decrease in cattle suffered from TRP (P<0.05), impaction (P<0.01), and vagus indigestion (P<0.05). Serum magnesium level (mg/dl) showed no significant change.

Minor elements: Serum copper level (µg/dl) showed no significant change while zinc level revealed significant decrease in cattle suffered from TRP (P<0.01) and frothy tympany (P<0.05).

Serum lactate level (mg/dl) showed significant increase in cattle suffered from impaction (P<0.01).
Data of the mean values (±SE) of the biochemical parameters in admitted cases of ruminal disorders were presented in Table 4.

Ruminal Juice Analysis

Physical examination and protozoal count: Physical examination of ruminal juice revealed abnormal consistency and odor in diseased cattle. Examination of ruminal juice for protozoa showed various reductions in the activity and total number of protozoal count. Ruminal impaction showed severe reduction in both activity and number **(Table 5)** compared with control one (Figure 9a-c).

Ruminal electrolytes: There was a significant decrease (P<0.01) in sodium and potassium level in cattle suffered from TRP, impaction, free gas tympany, frothy tympany while there was a significant increase in chloride level in cattle suffered from vagus indigestion only (P<0.01).

Ruminal calcium and inorganic phosphorus level (mg/dl): There was a significant decrease in calciumin cattle suffered from impaction and frothy tympany (P<0.01) while it significantly increased in vagus indigestion (P<0.05). There was a significant decrease in inorganic phosphorus level incattle suffered from TRP, frothy tympany and vagus indigestion (P<0.01).

Ruminal lactate level (mg/dl): There was a significant increase in lactate level in cattle suffered from impaction (P<0.01).

DISCUSSION

Diseases of rumen and reticulum have a great economic importance because of severe losses of animal's productivity, treatment costs and deaths. In the present study, various pathological conditions of the forestomach including traumatic reticulo-peritonitis, impaction, free gas tympany, frothy tympany and vagus indigestion were investigated in cattle.

Clinical findings: Animals with TRP showed signs of pain, tachycardia, jugular pulsation/distension, congestion of mucous membrane and decrease of ruminal motility. Cattle with impaction showed anorexia, absence of rumination, slight abdominal distention and dehydration. Cattle with free gas tympany showed severe distension of left abdomen, decreased ruminal motility and tachycardia. Animal suffered from vagus indigestion showed characteristic inverted "L" shape of abdomen. The clinical signs recorded in this study were in agreement with those reported in the previous studies (Behl *et al*., 1997; Sattler *et al*., 2000; Ramprabhu *et al*., 2002; Singh *et al*., 2003; Radostits *et al*., 2007; Saleh *et al*., 2008; Suthar *et al*., 2011; Abu-seida and Al-abbadi 2014 Sudhakara *et al*., 2014 and Neamat-Allah, 2015 and Mohan *et al*., 2015).

Ferroscopic and radiographic examination: Ferroscopic examination is a simple screening tool for detection of swallowed metal foreign body however it has some limitations (Ramprabhu *et al*., 2002 and El-Hawari, 2013). In the present study, mine detector gave false results in some examined cases (Table 2). On contrary, radiography was best suited for gaining accurate results about foreign body. It enables visualization of metallic foreign bodies and obtains accurate information about their nature and position within reticulum (Braun *et al*., 1993 and Aref and Abdel-Hakiem, 2013).

Hematological examination: Screening of the obtained data of hematology showed that there were significant decrease in TRBCs count and Hb in cases with TRP. A significant decrease in TRBCs could be attributed to the loss of blood during penetration of reticulum or due to decrease proper digestion and absorption of nutrients essential for erythropoiesis. This finding was in agreement with Gokce *et al.* (2007); Ghanem (2010) and Sudhakara *et al*. (2014). On contrary, El-Hawari (2013) reported that there was no significant change in Hb in animals suffered from TRP. Additionally, there were significant increases in TWBCs and neutrophils count in TRP and vagus indigestion. Leukocytosis and neutrophilia were indicative of inflammatory responses that might have been due to infection associated with the penetration of the foreign bodies in reticulum or peritoneal cavity. Similar findings were obtained by Ghanem (2010); Aref and Abdel-Hakiem (2013); Sudhakara *et al*. (2014) and Kirbas *et al*. (2015).

There was a highly significant increase in total leucocytic count, neutrophils and monocytes, which may be attributed to inflammation (ruminitis) due to chronic irritation of the forestomach wall (Hailat *et al*., 1996; Vanitha *et al*., 2010; Behera and Nayak, 2013 and Hussain *et al*., 2013). Leukocytosis was also recorded in free gas and frothytympany which may be due to a reduction in cellular immunity associated with the stress factors. Similar finding was reported in previous studies (Latimer *et al*., 2003; Gokce *et al*., 2007 and Ismael *et al*., 2007).

Biochemical analysis

Serum proteins: Screening of the obtained data of serum proteins revealed hyperprotenamia, hyperglobulinemia and hypoalbuminemia were constant findings in cattle with TRP and vagus indigestion. These findings could be attributed to the production of protein in the liver in response to inflammation, which switched towards increased synthesis of positive acute phase proteins with a concomitant decrease in the synthesis of albumin or selective loss of albumin or sequestration into body spaces or malnutrition (Coles, 1986 and Gokce *et al*., 2007) associated with TRP and vagus indigestion.

Liver enzymes: Liver enzymes in the present study showed significant increase of serum ALP, GGT activities in free gas tympany, frothy tympany and vagus indigestion. A concomitant liver disease could explain the increase level of serum ALP and GGT.

Renal function The current study showed significant increase in BUN level in various forestomach disorders while there was no significant change in creatinine level. Increased BUN level with normal serum creatinine level indicate that there are no inherent kidney diseases (Coles, 1986) and consequently, these finding could be attributed to faulty fermentation and reduced microbial activity associated with disorders of forestomach (Hobson, 1988; Braun *et al*., 2007 and Vanitha *et al*., 2010). It also could be correlated with anorexia and decreased ruminoreticular activity leading to disturbed rumen metabolism (Garry, 2002).

Serum electrolytes Serum electrolytes showed significant decrease in serum potassium level in cases of vagusindestion and impaction. Moreover, there is significant decrease in serum chloride in case of vagus indigestion. These could be attributed to anorexia, ruminalhypomotility and probably abomasal reflux (Hoflund, 1980, Moore, 1997; Ghanem, 2010 and Vanitha *et al.*, 2010) result in sequestration of hydrochloric acid in the rumen causinghypokalemia, hypochloremia and metabolic alkalosis (Kuiper and breukink, 1986; Behl *et al*., 1997 and Latimer, 2003; Ghanem 2010; Hussain *et al*., 2014 and Sudhakara *et al*., 2014).

Serum lactate: increased significantly in case of impaction which may be attributed to increased lactate production in the rumen associated with impaction. These resultswere in agreement with Ghanem (2010) and Sudhakara *et al*. (2014).

Major elements: showed significant decrease in serum calcium and inorganicphosphorus level in cases of TRP, impaction, vagusindigestion which may be due to decreased elements uptake and absorption because of anorexia and gastrointestinal stasis (Moore, 1997; Roussel *et al*., 1997 and Sudhakara *et al*., 2014). These results go in a parallel line with that mentioned by Samad *et al*. (1994); Ward and Ducharme (1994); Gokce *et al*. (2007) and Sudhakara *et al*. (2014).

Minor elements: The significant decreases in zinc in cattle suffered from TRP might be associated with shortage of feeds especially of minerals and reduced ruminal motility due to anorexia (Radostits *et al.*, 2007 and Vanitha *et al.*, 2010).

Rumen fluid analysis: Examination of the rumen fluid is often essential to establish the functional state of the rumen and its contents through examination of color, consistency, odour, pH, chemical examination and microbial status (Khan *et al*., 1999). Abnormalities in the physical characters of the ruminal fluid in various affections of the forestomach were in agreement with the findings of previous studies (El-Amrousi *et al*., 1985; Radwan, 1992; Radostits *et al*., 2007; Turkar and Uppal, 2007 and Mohan *et al*., 2015).

An immense population of various specialized kinds of ciliate protozoa is found in the rumen of domestic ruminants. Their number according to Hungate (1966) may exceed one million per gram of rumen contents and their mass roughly equals that of the bacteria. Rumen protozoa mainly functions to store starch granules thereby help in maintaining pH of rumen.

In cases of TRP and free gas tympany, the motility of protozoa ranged from ++ ve to +++ ve, but there were unexpected decrease in total protozoal count. These results were in agreement with Clarke (1965) and Mahmoud *et al*. (2016). In cases of impaction, frothy tympany and vagus indigestion, the motility of protozoa ranged from +ve to nil (-ve) with dramatic decrease in the total protozoal count. Low rumen pH and high lactate production in impaction leads to severe destruction of ruminal protozoa (Radostits *et al*., 2007).

Results of ruminal biochemistry revealed significant decrease of ruminal K, Ca and P levels in case of impaction and frothy tympany and the significant decrease in ruminal K and Na levels in free gas tympany. These findings could be attributed to anorexia and rumen hypomotility (Turkar and Uppal, 2007, Ismael *et al*., 2007 and Radostits *et al*., 2007). While the increase of lactate concentration in rumen may be attributed to increase of lactic acid production associated with impaction (Radostits *et al*., 2007). In case of vagus indigestion in cattle, there were significant increases in ruminal chloride and calcium levels and significant decrease in ruminal phosphorus level. The increase of ruminal chloride level could be attributed to hypochloremic metabolic alkalosis associated with sequestration of hydrochloric acid in the rumen caused by vagus indigestion and abomasal reflux (Kuiper and breukink, 1986; Behl *et al*., 1997

and Latimer, 2003: Ghanem 2010; Hussain *et al*., 2014 and Sudhakara *et al*., 2014).

Conclusion: Diagnosis and differential diagnosis of different ruminal disorders were collectively depends on history, clinical signs, ferroscopic and radiographic examination. The use of X-ray is very useful and can be used to distinguish between TRP/vagus indigestion and other ruminal disorders. Hematological, biochemical and ruminal juice analysis would assist in diagnosis of ruminant stomach diseases.

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التقييم الإكلينيكي والمعملي لإضطرابات الكرش والشبكية في الابقار

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تمثل أمراض المعدة في المجترات أهمية اقتصادية كبيره، لذا هدفت الدراسة إلي التقييم الإكلينيكي والمعملي لاضطرابات الكرش والشبكية في الأبقار مع دراسة التغيرات الدموية والبيوكيميائية المصاحبة في الدم وسائل الكرش وكذلك تأثيرها علي عدد ونشاط الحيوانات الأولية (البروتوزوا) في الكرش. اشتملت الدراسة على عدد ٤٦ حيوان من ذكور وإناث الأبقار (٣١ مريضة و ١٥ بقرة سليمة كمجموعة ضابطة). تم فحص الحالات المريضة داخل المستشفى البيطري التعليمي بجامعة أسيوط في الفترة من يوليو ٢٠١٥ إلي نوفمبر ٢٠١٦. أُخْضعت هذه الحيوانات للفحوصات الإكلينيكية المختلفة وكذلك فحص الحيوانات باستخدام جهاز كشف المعادن والأشعة السينية والجراحة الاستكشافية العلاجية. كما جري التقييم المعملي لجميع الحيوانات من تحاليل دموية وبيوكيميائية لمصل الدم وسائل الكرش. تم تقسيم حالات إصابات الكرش والشبكية بناءاً على الفحص الإكلينيكي وكشف المعادن والأشعة السينية والجراحة الاستكشافية إلى ٥ مجموعات: التهاب الشبكية البريتونى الوخذي (٧ حيوانات) ، عسر هضم نتيجة إصابة العصب الحائر (٥ حيوانات) ، اللكمة (٥ حيوانات) ، الانتفاخ الغازي (٩ حيوانات) والانتفاخ الرغوي (٥ حيوانات). أظهرت الدراسة أن الحيوانات المصابة بالتهاب الشبكية البريتونى الوخذي تعاني من أعراض واضحة للألم مع وجود نبض في الوريد الوداجى في بعض الحيوانات. وكان الفحص باستخدام جهاز كشف المعادن موجباً كما أظهر الفحص باستخدام الأشعة السينية وجود أجسام معدنية مخترقة الشبكية. أظهرت التحاليل الدموية انخفاض معنوي في عدد كرات الدم الحمراء وتركيز الهيموجلوبين وارتفاع معنوي في عدد كرات الدم البيضاء والخلايا المتعادلة متعددة النواة (النيتروفيل) في الدم، كما أظهرت التحاليل البيوكيميائية لمصل الدم ارتفاع معنوي في مستويات البروتين الكلى والجلوبيولين والبولينا وانخفاض معنوي في مستوى الالبيومين والكالسيوم والفسفور والصوديوم والزنك بينما أظهر تحليل سائل الكرش انخفاض معنوي فى عدد وحركة الحيوانات الأولية (البروتوزوا) وتركيزات البوتاسيوم والصوديوم والفسفور. تميزت الحيوانات التي المصابة بسوء الهضم نتيجة إصابة العصب الحائر بوجود انتفاخ مميز في البطن (يشبه الكمثري). وكان الفحص باستخدام جهاز كشف المعادن سالبا في عدد ٢ حيوان وموجبا في عدد ٣ حيوان وتم التأكد عن طريق الأشعة السينية. وأظهرت التحاليل الدموية انخفاض معنوي في تركيز الهيموجلوبين وارتفاع معنوي في عدد كرات الدم البيضاء والنيتروفيل والايزينوفيل ، كما أظهرت التحاليل البيوكيميائية لمصل الدم ارتفاع معنوي في مستويات البروتين الكلى والجلوبيولين ونشاط الألكالين فوسفاتيز والجاما جلوتاميل ترانسفيريز والبولينا وانخفاض معنوي في مستويات الالبيومين والكلوريد والفسفور. اظهر تحليل سائل الكرش انخفاض معنوي في عدد وحركة البروتوزوا وارتفاع معنوي في مستوى الكلوريد والكالسيوم وانخفاض معنوي في الفسفور. أظهرت الحيوانات المصابة باللكمة توقف تام في حركة الكرش وانخفاض بسيط في درجة حرارة الجسم. وكان الفحص باستخدام جهاز كشف المعادن سالباً. وأظهرت التحاليل الدموية ارتفاع معنوي في عدد كرات الدم البيضاء والنيتروفيل ووحيدات النواة كما أظهرت التحاليل البيوكيميائية لمصل الدم ارتفاع معنوي في مستوى البولينا واللاكتيت والصوديوم انخفاض معنوي في مستوى البوتاسيوم والفسفور. اظهر تحليل سائل الكرش انخفاض معنوي في عدد وحركة البروتوزوا ومستوى البوتاسيوم والصوديوم والكالسيوم ارتفاع معنوي في مستوى اللاكتيت. الحيوانات المصابة بالانتفاخ الغازي الحر تميزت بوجود انتفاخ شديد في الكرش. وكان الفحص باستخدام جهاز كشف المعادن سالبا في ٦ حيوانات و موجب كاذب في ٣ وتم التأكد باستخدام الأشعة السينية. وأظهرت التحاليل الدموية ارتفاع معنوي في عدد كرات الدم البيضاء والنيتروفيل والايزينوفيل، كما أظهرت التحاليل البيوكيميائية لمصل الدم ارتفاع معنوي في تركيزات الجلوبيولين والبولينا والصوديوم ونشاط انزيم الألكالين فوسفاتيز والجاما جلوتاميل ترانسفيريز. اظهر تحليل سائل الكرش انخفاض معنوي في عدد وحركة البروتوزوا ومستوى الصوديوم والبوتاسيوم. تميز الانتفاخ الرغوي بانتفاخ شديد في البطن وتوقف حركة الكرش. وكان الفحص باستخدام جهاز كشف المعادن والأشعة السينية سالباً. وأظهرت التحاليل الدموية ارتفاع معنوي في النيتروفيل، كما أظهرت التحاليل البيوكيميائية لمصل الدم ارتفاع معنوي في مستوى الجلوبيولين ونشاط الألكالين فوسفاتيز والجاما جلوتاميل ترانسفيريز والبولينا والصوديوم. بينما اظهر تحليل سائل الكرش انخفاض معنوي في عدد وحركة البروتوزوا وتركيزات الصوديوم، البوتاسيوم، الفسفور والكالسيوم.

الخلاصة: يعتمد تشخيص أمراض اضطرابات الكرش والشبكية بشكل أساسي على التاريخ المرضي للحالة، العلامات السريرية والفحص بجهاز كاشف المعادن. ويعتبر الفحص بالأشعة السينية أداة فعالة في التشخيص المقارن للتفرقة بين اضطرابات الكرش والشبكية الناتجة عن ابتلاع الأجسام المعدنية والناتجة عن التغذية الغير سليمة. تعتبر التحاليل الدموية والبيوكيميائية وعصارة الكرش ذات قيمة إضافية في تقييم الحالات المختلفة لإصابات الكرش والشبكية في الأبقار.