

Animal Health Research Institute
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**STUDY OF SOME INTESTINAL BACTERIAL
INFECTION AND BLOOD BIOCHEMICAL CHANGES
IN GOATS SUFFERING FROM ENTERITIS
IN ASSIUT GOVERNORATE**
(With 6 Tables)

By

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(Received at 9/10/2010)

دراسة بعض الإصابات البكتيرية والتغيرات البيوكيميائية في الماعز المصابة
بالنزلات المعوية في محافظة أسيوط

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أجريت هذه الدراسة على عدد ٣٣٠ رأساً من الماعز البلدى المصرى من الجنسين تراوحت أعمارهم من ١-٢ سنة من أماكن مختلفة بمحافظة أسيوط. أظهر الفحص الإكلينيكي بأن ٦٠ (١٨,١٨%) من الماعز كانت تعاني من نزلات معوية متمثلة في إسهالات بدرجات متفاوتة لونها مائل للاخضرار ومخاطية في بعض الأحوال مع هزال وضعف عام وقلة الشهية للأكل وجفاف متوسط وباقي الماعز سليمة من الناحية الإكلينيكية الظاهرية. اشتملت الدراسة على أخذ ٦٠ مسحة شرجية من الماعز المريضة إكلينيكياً وذلك لفحصها بكتريولوجياً. هذا وقد أظهر الفحص البكتريولوجى عن عزل ميكروبات القولونى ٥٩ (٩٨,٣%) والسالمونيلا ١ (١,٦٧%). كما تم تصنيف الميكروب القولونى (*E. coli*) سيرولوجياً إلى ١٠ (١٦,٩٤%) *E. coli* O₂₆/B₆ ، ٦ (١٠%) *E. coli* O₁₁₄/B₁₆ ، ٤ (٦,٦%) *E. coli* O₂₆/B₆₀ ، ٣٩ (٦,٦%) لم يتم تصنيفهم. وتم تصنيف العترة المعزولة من ميكروب السالمونيلا سيرولوجياً إلى ١ (١,٦٧%) سالمونيلا إنترديدياس. وتم اختبار حساسية للميكروبات المعزولة لعدد من المضادات الحيوية ووجد أن أكثرهم تأثيراً ريمكتان - كلورمفينكول - جاراميسين. وقد شملت الدراسة أيضاً قياس مستوى كلاً من الجلوكوز والبروتين الكلى في مصل الدم وتبين وجود انخفاض ملحوظ في نسبة الجلوكوز والبروتين الكلى في الحالات المريضة. وقد استهدفت الدراسة معرفة مدى تأثير الإصابات البكتيرية المعوية على الاتزان الحمضى القاعدى وغازات الدم في الدم الغير متجلط، وكذلك الأليكترولايت في مصل الدم للماعز المريضة والسليمة. وأتضح من النتائج وجود نقص معنوى جداً في قيم الأس الهيدروجينى pH ، وانخفاض معنوى في قيم كل من البيكربونات (HCO₃)، ضغط ثانى أكسيد الكربون الكلى (TCO₂) ، الزيادة القاعدية (B.E) ، الصوديوم ، الكلورايد بالإضافة إلى ارتفاع معنوى في قيمة البوتاسيوم في الماعز المريضة عند مقارنتها بالمجموعة السليمة.

SUMMARY

The current study was carried out 330 Egyptian Balady goats from both sex, aged 1-2 years from different localities at Assiut governorate. The clinical examination revealed that 60 (18.18%) have enteritis with signs of diarrhea of different degrees, anorexia, loss of appetite, depression and slight emaciation and the remaining goats were apparently clinically healthy. 60 rectal swabs were taken from clinically diseased goats for bacteriological examination which revealed that the main isolates were *E.coli* 59(98.3%) and *Salmonella* 1(1.67%). The isolated *E.coli* strains were identified serologically into 10 strains and 39(66.10%) untypable strains. The isolated salmonella strain were identified serologically into 1(1.67%) salmonella enteritidis. Antibiogram for the bacterial isolates revealed that the best sensitive antibiotics were rimactan, chloromphenicol and garamycin. Blood serum biochemical analysis in diseased cases showed significant ($p<0.05$) decrease in glucose and total protein levels in diseased cases. Also, the study aimed to know the effect of bacterial enteritis on the blood gases, acid-base balance in heparinized blood and serum electrolytes of both clinically healthy and diseased goats. There were a marked decrease in mean values of blood pH, significant decrease in values of HCO_3 , TCO_2 , B.E. sodium and chloride as well as significant increase in potassium level in clinically diseased goats when comparison with the control group.

Key words: Goat, Bacterial enteritis, Biochemical changes, Assiut Governorate.

INTRODUCTION

Goats are considered one of the most important farm animals and constitute one of the sources of meat for human consumption in Egypt.

Enteritis in young animals is a syndrome of great aetiological complexity. In addition to the influence of varied environmental, managerial, nutritional and physiological factors, but the numerous infectious agents are capable of causing enteritis in small animals (Snodgrass *et al.*, 1986).

Bacterial enteritis is the most important cause of diarrhea in goats and lambs. Enterotoxigenic *Escherichia coli* (ETEC) is considered among the most prevalent causative agent of enteritis in goats (Gerald *et al.*, 1992).

Coliform bacteria are commensals of the alimentary tract and can readily be identified on faecal culture. Enterotoxigenic *E.coli* strains are

significants as a cause of diarrhoea in goats (Matthews, 1999) Enterogenic *E.coli* can also be isolated from both diarrhoeic and non diarrhoeic kids, (Meshram *et al.*, 2009).

E.coli scour is an opportunistic disease associated with sloppy environmental conditions, poor sanitation and managerial practices. (Scott and Gessert, 1996).

Shaw (1971) has described a dysentery associated with *E.coli*. the author stated that the condition is usually seen where ewes and lambs were crowded together.

Causative agent *E.coli* has two different virulent strains, one type is associated with enterotoxigenic *E.coli*, which has two virulen factors responsible for diarrhea. *E.coli* is causative agent of white scour in goat (Bhat *et al.*, 2008). Animals suffering from white scour have severe colitis characterized by abdominal pain, pasty faeces, and severe enteritis which may culminates to death due to severe dehydration (Radostits *et al.*, 1995).

E.coli while as a normal gastro-intestinal inhabitant is also associated with a variety of pathological conditions in animals (Smith, 1965).

Salmonellosis cause acute diarrhoea in older goats and kids (Matthews, 1999).

Salmonella enteridis produces enterotoxins which are invasive to cause inflammatory change within the intestine leading to diarrhoea.

Bacterial enteritis remains the most common clinical problem in the goats (Meshram *et al.*, 2009).

Salmonella typhimurium and salmonella enteritidis were also reported to be associated with some outbreaks of diarrhoea in adult goats and kids (Bulgin and Anderson, 1981).

Also Blackwell (1983) stated that salmonella enteritidis and salmonella typhimurium were frequently reported to be the cause od enteritis in kids, lambs, adult sheep and goats.

Many diseased conditions of which digestive disturbances are of great importance, induce severe changes in serum glucose and total protein elvels which reflect the status of animal body (Kaneko and Cornelius, 1971).

Ruminants subjected to different types of metabolic disorders that originated more or less from digestive origin (Brobest, 1975).

Serious disorders of acid-base balance and serum electrolyte occur in association with some diseased conditions in sheep and goats (Coles, 1980).

The purpose of this study:

- 1 – Isolation and identification of bacteriological causative agents responsible for enteritis in goats located at Assiut Governorate.
- 2 – Study the anti-bacterial susceptibility test of isolated bacteria.
- 3 – Study the effect of enteritis on some biochemical changes in blood serum of diseased goats and the changes in values of acid-base balance, blood gases and electrolytes in goats.

MATERIALS and METHODS

A total of 330 balady goats of both sex aged (1-2) years were used in this study located in different localities at Assiut Governorate. 60(18.18%) out of them showed clinical signs of enteritis.

Samples for bacteriological examination:

60 rectal swabs were taken from clinical and diarrhoeic goats for bacteriological studies.

The rectal swabs were inoculated aseptically into nutrient broth at 37°C for 24 h. and then subculture into the following media. Difco nutrient agar, 5% sheep blood agar, MacConkey agar, SS agar and XID agar.

The isolates were identified according to the colonial morphology, the pigment production, microscopically by gram stain and biochemically according to Baily and Scott, (1974); Quinn *et al.* (1994).

Serological characterization of isolated strains of *E.coli* and salmonella were done.

Antibiotic sensitivity testing: The bacterial isolates were tested for their resistance using disc diffusion method according to Bauer *et al.* (1966), while zones of inhibition were determined according to National Committee for clinical Laboratory Standards (NCCLS, 2002). The antibiotic discs used were chloramphenicol (30 µg), amoxicillin (25 µg), nalidixic acid (30 µg), erythromycin (15 µg), garamycin (30 µg), netilmicin (30 µg), tetracycline (30 µg), rimactan (30 µg), colistin (10 µg) and neomycin (30 µg). The diameter of inhibition zones were measured (in mm) for each plate compared with the standard break points.

Blood samples with anti-coagulant (EDTA) were collected from control group and clinically diseased goats for blood gases and acid-base balance measurements using corning pH-blood gas analyzer Model 168. The analyses directly measured at 37°C, blood pH, carbon dioxide tension (PCO₂ mm Hg), oxygen tension (PO₂ mm Hg), bicarbonate (HCO₃ mmol/L), total carbon dioxide (TCO₂ mmol/L) and base excess (B.E mmol/L).

Another blood serum samples were collected for determination of glucose mg/dL (Werner and Wielinger, 1970) and total proteins g/dL (Weichselbum, 1946). These levels were estimated spectrophotometrically by using test kits supplied from Bio-Merieux/Lab. Reag. Prodi, France.

Serum Na and K were determined by using flame photometer (Corning Model 400) while chloride level was determined by using chloride meter analyzer (Corning Model 925 England).

Statistical analysis of obtained serum biochemical data was performed according to method of Kalton (1967) using t-test.

N.B.I would like to express my deep thanks to the department of biochemistry and microbiology, faculty of medicine Assiut El-Azhar University.

RESULTS

The most prominent clinical signs of diseased goats were loss of appetite, diarrhoea, depression and general weakness.

The obtained results were shown in Tables 1-6.

Table 1: Localities and percentages of clinically diarrhoeic goats.

Locality	Sex		No. of examined goats	No. of clinically diseased goats	
	Male	Female		No	%
Bani-Adi	30	80	110	28	25.45
Abnob	50	70	120	20	16.67
Bani-Mor	40	60	100	12	12
Total	120	210	330	60	18.18

Table 2: Isolated pathogens from bacteriologically examined rectal swabs from diarrhoeic goats.

Bacterial isolates	No	%
<i>E.coli</i>	59	98.3
Salmonella	1	1.67
Total	60	100%

Table 3: Serological identification of isolated *E.coli* and Salmonella strains of diarrhoeic goats.

Enteropathogenic	No	%
<i>E.coli</i> O ₂₆ /B ₆	10	16.94
<i>E.coli</i> O ₁₁₄ /B ₁₆	6	10
<i>E.coli</i> O ₂ /B ₆₀	4	6.6
Untypable <i>E.coli</i> strains	39	66.10
Salmonella enteritidis D ₁	1	1.67

Table 4: Inhibition Zone diameter (IZD) of test antibiotics against isolates of *E.coli* and Salmonella.

Antimicrobial agents	Isolation	
	<i>E.coli</i>	Salmonella
Amoxicillin (10µg)	R(<13 mm)	R(<13 mm)
Naladixic acid (30 µg)	R(<13 mm)	R(<13 mm)
Colistin (10 µg)	R(<8 mm)	R(<8 mm)
Erythromycin (15 µg)	R(<13 mm)	R(<13 mm)
Neomycin (30 µg)	R(< 10 mm)	I(<15 mm)
Garamycin (30 µg)	S(>15mm)	S(>15 mm)
Teracycline (30 µg)	R(<14 mm)	R(<14 mm)
Chloramphenicol (30 µg)	S(>18 mm)	S(>18 mm)
Netilimicin (30 µg)	S(>22 mm)	S(>22 mm)
Rimactan (30 µg)	S(>20 mm)	S(>20 mm)

R: Resistance

I: Intermediate inhibition zone

S: Sensitive

Table 5: Blood serum biochemical values in apparently healthy (Control) and clinically diseased goats.

Parameters	Healthy goats (10) mean ±SE	Diseased goats (20) mean±SE
Glucose (mg/dL)	60.98 ± 0.5	58.66 ± 1.25*
Total protein (g/dL)	6.37 ± 0.64	5.83 ± 1.03*

*: Significant (P < 0.05)

**: Highly significant (P < 0.01)

Table 6: Values of blood gases, acid-base balance and electrolytes in clinically healthy and diarrhoeic goats.

Parameters	Clinically healthy mean ± S.E	Diseased goats (10) mean ± S.E
pH	7.441 ± 0.040	7.250 ± 0.071 **
PCo ₂ mmHg	38.243 ± 3.987	35.532 ± 3.817
Po ₂ mmHg	32.236 ± 6.763	33.573 ± 6.175
HCO ₃ mmol/L	26.573 ± 2.813	19.620 ± 4.003*
TCO ₂ mmol/L	28.180 ± 3.296	21.163 ± 4.256*
B.E mmol/L	3.140 ± 2.933	-2.240 ± 5.575*
Na ⁺ mmol/L	154.710 ± 6.867	139.530 ± 2.351*
K ⁺ mmol/L	5.43 ± 0.363	6.610 ± 0.126*
CL mmol/L	106.600 ± 357	97.400 ± 2.675*

*: Significant (P < 0.05)

**: Highly significant (P < 0.01)

DISCUSSION

Various degrees of diarrhoea, depression, weakness and loss of weight were noticed during clinical investigation of diarrhoeic goats. Diarrhoea in some cases was offensive, semifluid, watery, yellowish, containing mucous and sometimes tinged with blood. In others, faeces were foeted, profuse, caly to yellowish or grayish. These findings support many of previous investigations in a similar condition (El-Daml *et al.*, 2001 and Meshram *et al.*, 2009). Mean while Abd-El-Mawla and Ahmed (2002) recorded that growth rate is reduced and weight gain loss could occur in older goats and sheep but diarrhoea is a common condition affecting newly born lambs and goats.

Table 1 recorded 60(18.18%) goats out of 330 had diarrhoeic signs while El-Daml *et al.* (2001) mentioned that 124(66.66%) of goats from total 186 showed clinical signs of diarrhoea. Meanwhile Abd-El-Mawla and Ahmed, (2002) recorded that 77(77%) diarrhoeic goats from 100 clinically healthy ones.

The result in table 2 recorded that *E.coli* is the dominant bacterial infection among diarrhoeic goats. This came in agreement with data obtained by El Daml *et al.* (2001). Also, Samaha *et al.* (2002) who recorded that *E.coli* and salmonella were the most common bacterial isolates from diarrhoeic goats. The authors mentioned that 29(58%) *E.coli* and 1(2%) salmonella sp. from total 50 rectal swabs of goats with diarrhoeic signs. On the other hand, El Daml *et al.* (2001) mentioned that

E.coli was isolated from 22 cases of diarrhoeic goats (17.74%) out of 124 cases of goats of clinically signs of diarrhoea and 7(5.64%) salmonella sp. were isolated.

Abd-El-Mawla and Ahmed, (2002) recorded that *E.coli* was isolated 32(32%) from 100 diarrhoeic goats and 2(2%) salmonella typhimurium. The results recorded in Table 3 revealed that the enteropathogenic *E.coli* O₂₆/B₆; O₁₁₄/B₁₆; O₂₆/K₆₀) were the main microbiological cause of enteric infections in diseased goats. This result agreed with that previously reported by Amer *et al.* (1985) and Snodgrass *et al.* (1986). The mechanism by which the organisms may produce the disease is the toxins production. These toxins may be entro or endotoxins. The entro-toxin is produced by entero-pathogenic *E.coli* proliferating in the anterior small intestine causing hypersecretion through intestinal epithelium. Some pathological changes in the jejunum and ileum, and diarrhoea. Endotoxin is thought to act either directly or indirectly by means of an anaphylactic reaction which could produce diarrhoea, various forms of enteritis and oedema disease (Sojka, 1971).

However there are some entero-pathogenic *E.coli* which do not produce toxin, but they adhere to surface of the enterocytes of large intestine, and the affected calves pass bright red blood in diarrhoeic faeces (Hail, 1985). In this study salmonella infection formed the second main cause of enteritis in diseased goats (Table 3). Our results agreed with Meshram *et al.* (2009) who recorded that bacteriological examinations of goats suffering from enteritis were found to be positive for either Salmonella enteritidis and Escherichia coli (*E.coli*). Johnston *et al.* (1976) mentioned that Salmonella causes the main pathological lesion in the from of inflammation of the ileum, while the main clinical manifestations were loss of appetite, depression, dullness and diarrhoea. The faeces was watery and contained mucous and clots of blood.

Antibiogram for the isolates of enterotoxigenic *E.coli* and salmonella revealed that the best sensitive antibiotics are rimactan, chloramphenicol, garamycin and netilimicin (Table 4). This result agreed with previously reported by Monica (1985) and Amar *et al.* (1993). On the other hand, Sayed *et al.* (2001) recorded that all *E.coli* strains in enteric sheep were sensitive and susceptible to both tobramycin and enrofloxacin. Also El-Daml, (2001) mentioned that all strains of *E.coli* and salmonella in diarrhoeic kids were sensitive to ciprofloxacin and Enrofloxacin, gentamycin and chloramphenicol. These results did not agree with data obtained by Abd-El-Mawla and Ahmed (2002). Those authors mentioned that streptomycin, gentamycin and neomycin were the best drugs for

treatment of *E.coli* in diarrhoeic lambs and goats while gentamycin, ampicillin and chloramphenicol were the best drugs for salmonellosis.

Blood serum biochemical analysis (Table 5) declared that values of glucose and total protein levels in clinically healthy goats agreed with the data obtained by Meshram *et al.* (2009). In addition there were significant decrease ($p < 0.05$) in glucose and total protein in goats suffering from enteritis when compared with clinically healthy ones. These data were generally supported by Meshram *et al.* (2009). The obtained hypoglycaemia may be due to the lack of intestinal absorption in case of enteritis (Coles, 1967). Mottelib, (1972) attributed such decrease in serum glucose level found in calves suffering from *E.coli* enteritis is due to the alteration in tissue metabolism caused by decreased blood flow and oxygenation. Another explanation has been given by Madison (1964) which attributed it to be due to the elevation of the ketone bodies in case of gastro-enteritis, and such elevation in blood ketones which consequently led to depression of the blood glucose. There was a significant decrease ($p < 0.05$) in total protein in diarrhoeic goats when comparison with control healthy ones. These obtained data were in agreement with Meshram *et al.* (2009). Coles (1986) attributed the reduction of proteins to the anorexia of the diseased goats and the inability of their liver to synthesize protein.

Also, West *et al.* (1968) and Doxey (1971) suggested that certain bacteria or bacterial toxins causes increased capillary permeability and permit the escape of plasma proteins in tissues, so the osmotic pressure of proteins is increased in the tissue fluids and at the same time was decreased in the blood. Meanwhile Amer *et al.* (1993) reported that there is a significant decrease in glucose level but there is no significant change in total protein in calves with bacterial enteritis.

Amer *et al.* (1985) and Hassan *et al.* (1985) detected the predominance of entero pathogenic *E.coli* as the main cause in the pathogenesis of enteritis in calves at Upper Egypt. Salmonella sp. are considered mainly as primary organisms in case of enteritis in calves.

Evaluation of acid-base status and serum electrolytes level of the patient, are of great importance, as they provide the necessary information that will lead to greater understanding of the nature of the disease process, and guide line to the way of therapy. Serious disorders of acid-base balance occur in association with some diseased conditions in goats and sheep (Radostits *et al.*, 1995).

In Table 6 the values of pH, PCO_2 , PO_2 , HCO_3^- , TCO_2 and B.E. as well as Na^+ , K^+ and CL in clinically healthy goats are nearly similar to results obtained by Ali, (1985). In clinically diarrhoeic goats where highly significant ($p < 0.05$) decrease in values of pH and significant decrease

($p < 0.05$) in values of HCO_3 , TCO_2 and excess (B.E.). On the other hand, there were significant decrease ($p < 0.05$) in both Na^+ and Cl^- while significant increase ($p < 0.05$) in K^+ level in diseased goat. These results agreed with the results obtained by El-Sebaie *et al.* (1988). The drop of pH and base excess indicated more or less the occurrence of metabolic acidosis (Haskins, 1983). The significant drop in the HCO_3 value in enteric group is mainly due to the loss of HCO_3 with intestinal secretion through faeces. It was a constant finding that any alteration in HCO_3^- was usually accompanied with the change in the concentration of TCO_2 because the TCO_2 values were calculation dependant.

The results of Na^+ , K^+ and Cl^- are nearly similar to results recorded by El-Sebaie *et al.* (1988) who mentioned that there were significant drop in Na^+ , Cl^- values and significant increase in K^+ value in calves suffering from enteritis and such variations due to great loss of sodium (Na^+) and chloride (Cl^-) from body in enteric calves. While the obtained data about electrolytes did not agree with results obtained by Abou-El-Ela (2008), where in significant changes in both potassium and chloride occurred in enteritis of goats. On the other hand, primary metabolic acidosis may occur in animals during enteritis (Duks, 1964). Metabolic acidosis varying from moderate to severe was observed by Robert *et al.* (1990) in most calves suffering from diarrhoea concomitant with acidosis variations which were observed in serum chloride, sodium and potassium levels. In general our obtained data was in agreement with values previously recorded by Donawich and Baue, (1968), Saleh, (1983), El-Sebaie and Hassan (1984) and El-Sebaie *et al.* (1984).

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