## GOATS RUMENAL BACTERIA, THEIR COUNT AND ANTIBACTERIAL EFFECT AGAINST-GRAM POSITIVE BACTERIA

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### **ABSTRACT**

The bacterial and fungal population in the rumen fluid was measured by using different cultures media and incubation temperature. The higher mean of mesophils and staphylococci was found in the rumenal fluid of goat. While higher mean of psachrophils, coliform and fungi count was observed in the rumenal fluid of cow. Significant mean difference among microbial population in the rumenal fluid was observed among cow, goat and buffalo concerning the mesophils bacteria. Also significant mean difference was observed in Escherchia coli mean count among goat, buffalo and cow rumenal fluid microbial population. There was no significant difference in the mean count of mesophils, coliform, staphylococci and fungi. A freshly isolated E.coli from rumenal fluid of goat had antibacterial activity against Streptococcus spp. and Staphylococcus aureus.

#### INTRODUCTION

The rumen microbial ecosystems comprises at least 30 predominant bacterial species at a total concentration of 10<sup>10</sup> to 10<sup>11</sup> cells/ml of rumenal fluid, some 40 species of protozoa(10<sup>5</sup> to 10<sup>7</sup> cells/ml), and five species of fungi(10<sup>5</sup> cells/ml).

Bacterial species of the rumen are considered more important than protozoa and fungi in determined the extent and rate of feed degradation, and utilization for the production of microbial protein<sup>(10)</sup>. Further more ,some bacteria produce antibacterial compounds like bacteriocins. Bacteriocins-Like inhibitors (BLI) are heterogeneous group of antibacterial peptides and protein characterized by their ability to inhibit closely related and some times more distantly, strains of bacteria<sup>(1)</sup>. It has been proposed that bacteriocins may play a key role in bacteria population dynamics<sup>(8)</sup>. In particular, the bacteriocin may give the producing strain a competitive advantage by killing bacteria in the some environment competing for the resources<sup>(6)</sup>. The diversity and density of the microbial population of the rumen suggest that this environment might favor the evolution of bacteriocins as competitive factor<sup>(3,4)</sup>. If this can be confirmed, bacteriocins would have

potential applications as agents for the modification of rumen microbial population(11).

The aim of this study is to count rumenal bacteria in different animals species and determination of antimicrobial agent of rumen bacteria against other bacteria.

# MATERIALS AND METHODS

Samples

A total of 45 rumenal fluid were randomly obtained for the microbial analysis. Fifteen samples were collected from each of slaughtered goat, buffalo and cow seen in Basrah slaughtered house.

Microbial analysis

All media used were obtained from oxoid limited, London. The following test were conducted according to method of Russel and mantovani <sup>(9)</sup>. Rumenal fluid diluted 1:100 in basal media was streaked on to nutrient agar, a total aerobic plate (APC)count with incubation at 37C° for 48hr *mesophiles* and at 4C° for 10 days for *psaychrophiles* (PPC). Total coliform (TC) and Escherchia coli were determined by using MacConkey agar and eosine methylen blue (EMB) agar with the incubation at 37 C° for 48hr and at 4C°, respectively. Positive MacConkey plates were used to calculate TC and EMB plates for E. coli count. The isolated from EMB plates were tested for indol production, methyle red, voges-proskaur reaction and citrate utilization ,*Staphylococcus* aureus count on mannitol salt agar at 35 C° for 48 hr.

Typical *Staph. aureus* colonies were counted and randomly picked up and incubated in to brain heart infusion (BHI) broth for 24hr at 35 °C and subjected to coagulase test .Fungi(molds and yeasts) were enumerated on saburud dextrose agar (SDA)and incubated at 22 °C for 5 days. Plates in all cases were incubated in triplicated and the microbial count were expressed as mean colony forming units per gram(CFU/G).

### Detection of bacteriocins of rumenal bacteria:

The bacteiocins activity against bacterial growth were determined according to methods of Perez et al(1990)  $^{(7)}$ . The colonies of *E. coli* from rumenal fluid that grown on (EMB) were picked and transferred to broth, 0.1 ml were taken from the broth to the wells (with 0.5 mm in diameter) on the plates of mullor hinton agar (MHA) seeded with approximately 106 cell/ ml of each *Streptococcus spp* and *Staphylococcus aureus* which were obtained from microbiology laboratory of Biology Department/college of science. The plates were re-incubated at  $37C^{\circ}$  for 24hr and each isolate was scored for its ability to create a distinct zone of clearing ( $\geq 3$ mm) in the agar overlay statistical analysis.

Statistical analysis:

The result were analysis by one-way ANOVA test using statistical package for the social sciences (SPPS) version 9.0 .All data were expressed as mean standard error. Differences between data were compared by least significant difference.

# RESULTS AND DISCUSSION

Fourty-five ruminal fluid samples were using to measure the bacterial and fungal population on different culture media and incubation temperature. The statistical analysis of these population in the rumen of cow ,buffalo and goat were displayed in (tables-1,2). In table(1) the higher rate of ruminal bacteria relate to the total /bacterial population occur in mesophils of goat, cow and buffalo. Also in this table the fungi are a relatively have small rate in compare to bacteria population. Table 2 show the mean and STD of ruminal bacteria in different culture media in goat ,cow and buffalo ,the high mean of mesophils and Staphylococci count was found in the ruminal fluid of goat while higher mean of psaychrophils , coliform and fungi count was found in the ruminal fluid of cow. The significant mean difference among microbial population in the ruminal fluid of cow, buffalo and goat was observed in case of mesophils ruminal bacteria between goat and buffalo. Also significant difference was found in E.coli mean count among goat, buffalo and cow ruminal fluid. The significant difference was observed in psaychrophils mean count among cow, buffalo and goat ruminal fluid, also the significant difference was found in the coliform mean count between buffalo and goat. There was no significant difference in the mean count of Staphylococci and fungi. The result reported here are consistent with other previous studies who find that the microbial population in the rumen consist of bacteria ,protozoa and fungi. The majority of the concentration is as bacteria which can number  $10^{10}$  to  $10^{11}$  cells/gram of rumen content<sup>(5)</sup>. The diet fed to ruminate animals influences the number and relative proportion of the different microbial species in the rumen. Consideration of microbial reproduction rate is essential when making dieting changes in any ruminant .Major changes in the diet require a period of transition to allow for shifts in the population of different microbial species ,this adaptation may take several days. One of the most common problems encountered in nutrition mandement is sudden change in the ruminant diet(6). The antimicrobial compounds produced by the population of E.coli isolated from the rumenal fluid of the goat had antimicrobial activity against gram positive bacteria species like Staphylococcus aureus fig.1 and Streptococcus spp fig.2. These results are in agreement with other studies which reported that some ruminal bacteria produce bacteriocins, there were speculation that this compound had effects on ruminal fermention that were similar to the ionophore, monensin<sup>(9)</sup>. However, cow are often fed silages, and silage fermentation is a batch culture system that favours rapidly growing lactic acid bacteria, silage fermentation could be a vehicle for delivering bacteriocins to the rumen<sup>(9)</sup>.

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Table-1-The rate of ruminal bacteria of goat, buffalo and cow.

Animal species	No.of	MESO	PSACHRO	масс.	MANN.	ЕМВ	SDA	Total
Goat	15	2514.5 (45.98 %)	202 (3.69%)	777.5 (14.21%)	834 (15.25%)	964 (17.62%)	176 (3.21%)	5468
Buffalo	15	1432.5 (30.71 %)	590.5 (12.65%)	595 (12.27%)	846.5 (18.14%)	1042.5 (22.34%)	1575 (3.37%)	4664. 5
Cow	15	1970 (38.13 %)	679 (13.14%)	1115.5 (21.15%)	715 (13.48%)	412 (7.97%)	247 (5.30%)	5165. 5

Table-2-The statistical analysis of ruminal bacteria of goat, buffalo and cow.

		No. Of sample	MEAN	Std. Deviation	Std. Error
MESO.	1.0	15	167.633	124.407	32.121
	2.0	15	95.500	65.086	16.805
	3.0	15	116.66	62.975	16.260
	Total	45	126.600	92.041	13.720
PSACHRO.	1.0	15	12.266	10.743	2.774
j	2.0	15	39.366	25.501	6.584
	3.0	15	45.266	35.687	9.214
	Total	45	32.300	29.326	4.3717
MACC.	0.1	15	51.866	37.733	9.7427
	2.0	15	39.666	29.069	7.5038
	3.0	15	67.700	32.415	8.3696
	Total	45	53.077	34.507	5.1441
MANN.	1.0	15	55.600	55.699	14.381
	2.0	15 .	56.433	38.374	9.908
	3.0	15	47.666	24.603	6.352
	Total	i45	53.233	40.7950	6.081
EMB	1.0 2.0 3.0 Total	15 15 15 45	64.266 69.500 27.500 52.755	38.933 61.189 21.398 6.954	10.052 15.799 5.525 6.954
		43	32.733	0.934	0.934
SDA	1.0 2.0 3.0 Total	15 15 15 45	11.733 10.500 18.266 13.500	8.6207 6.0415 21.045 13.714	2.225 1.559 5.434 2.044

<sup>1=</sup>goat 2=buffalo

<sup>3=</sup>cow

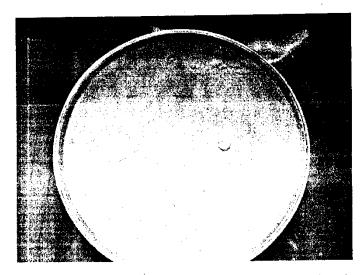


Fig1; Sensitivities of Streptococcus spp to BLIS produced by ruminal E.col.

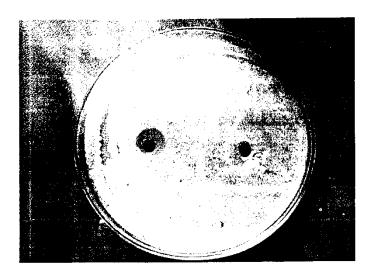


Fig 2; Sensitivities of Staphylococcus aureus to BLIS produced by ruminal E.coli.

### **CONCLUSIONS:**

The bacterial and fungal population in the rumen was different according to differences in the species of animals and different culture media and temperature. Ruminal bacteria has antimicrobial effect against other bacteria.

# بكتريا كرش المعز إعدادها وتأثيرها المضاد للجراثيم مقارنه مع الحيوانات المجترة الأخرى

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### الخلاصة

تم حساب مجاميع الجراثيم والغطريات الموجودة في سائل الكرش باستخدام أوساط زر عيه مختلفة وبدرجات حضن مختلفة, إن أعلى معدل لعدد الجراثيم المحبة لدرجات الحرارة المعتدلة وجراثيم المكورات العنقودية لوحظ في في سائل كرش المعز, بينما لوحظ أعلى معدل للجراثيم المحبة للبرودة والجراثيم القولونية والفطريات لوحظ في سائل كرش الأبقار ولوحظ وجود فرق معنوي في معدل عدد الجراثيم المحبة للبرودة بين الأبقار والجاموس والمماعز والجاموس وكذلك لوحظ فرق معنوي في معدل عدد الإسرش القولونية بين الماعز والأبقار من جهة والجراثيم القولونية بين معدل عدد المكورات العنقودية والجراثيم القولونية والغطريات من جهة أخرى وجد أن الأشرش القولونية المعزولة من سائل كرش الماعز أظهرت فعالية مضادة المكورات السبحية والمكورات العنقودية

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