



Comparative Study between Synovial Fluid and Serum Metabolites in Sudanese Cattle

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Abstract

This study was conducted to establish Normal values for Total protein (TP),Albumin (Alb), Globulin (Gl), Glucose(G) and Alanine aminotransferase (ALT) from synovial fluids in Baggara Sudanese cattle , compared with the corresponding values in serum and to investigate the correlation and regression among serum and synovial fluid of these metabolites. Eleven blood samples and twenty two synovial fluid samples were collected from carpal and tarsal joints of clinically healthy Baggara bulls, during the period from January to March 2018. The colorometric methods were used to determine the concentration of these metabolites using commercial kits which were analysed using Spectrophotometer (UV mini-1240-Japan). The data was analysed by ANOVA and LSD using SPSS programme (version 16). The glucose concentration of carpal joint was significantly ($p=0.024$) lower than that of the tarsal joint and serum, while the other metabolites were significantly lower ($p=0.000$) than serum. The correlation (between total protein, albumin, globulin, and ALT, between albumin and globulin and between globulin and ALT) and the subsequent simple linear equations of synovial fluid metabolites were established. In conclusion, data obtained will be useful as diagnostic tool to differentiate between normal and abnormal joints of cattle which will help in the diagnosis of joint disorders and diseases. The study recommends that, other studies of synovial fluid should be made in large number of cattle and comparison between synovial fluid metabolites and other body fluid metabolites such as cerebrospinal fluid should be done.

Keywords: Total protein, Glucose, ALT, Carpal joint, Tarsal joint.

Introduction

Synovial fluid is the dialysate of plasma, to which hyaluronic acid is added, its synthesis takes place in joint cavity by synovial membrane (Swenson and Reece,1993). It is considered one of the important transcellular fluid (Coles,1986). It is viscous, transparent, colourless to light yellow fluid (Altintas *et al.*, 2010 ; AbdEllah *et al.*, 2012), which contains in addition to hyaluronic acid, proteins, glucose, electrolytes, enzymes, cells (Coles,1986 ; Latimer, 2011) and antioxidant vitamins (Chalmeh *et al.*, 2016). Determination of physical, chemical and cytological characteristics of synovial fluid are considered as diagnostic valuable tools in diseases (Latimer, 2011). Several studies in camels (Al-Rukibat *et al.*, 2006), horses , donkeys , buffaloes (AbdEllah *et al.*, 2012) ,sheep (Ameri and Gharib, 2005) and rodents (Brombini *et al.*, 2017) analysed the synovial fluid in one joint, but there is scarce data in cattle concerning the synovial fluid analysis in different joints .

This study was designed to determine normal values of some metabolites in synovial fluid (carpal and tarsal joint) and serum in normal local Sudanese Baggara cattle breed.

Materials and Methods

Animals

Eleven apparently clinically healthy local Sudanese Baggara bulls were used in this study, they were selected from the herd of the Animal Production Research Centre - Khartoum north –Hillat kuku. They aged (28-49) months and their weights were ranged from 220 to 370 kg.

Collection of blood

Six mls of blood were collected from the jugular vein using 10 ml disposable syringe. Immediately, 2.5 ml of blood was transferred to clean dry test tubes containing sodium fluoride (Na F) as anticoagulant for

glucose test. The separated plasma was used for glucose determination. The rest of the blood was allowed to stay for 2hrs at room temperature and then centrifuged at 3000 r.p.m for 10 minutes to separate serum. Haemolysed free serum samples were harvested into clean vials and immediately frozen at -20C° for subsequent analysis.

Collection of synovial fluid (Arthrocentesis)

The synovial fluid of carpal and tarsal joints was collected immediately after the animals were slaughtered. It was collected under a septical condition, using 18G needle according to Chauhan and Agrawal ,(2006).

Biochemical analysis

Colorimetric methods were adopted for determining total protein, albumin, glucose concentration and activity of Alanine amino transferase using commercial kits (Biosystems-Spain), while globulin is obtained by subtraction of total protein from albumin. The analysis was done using spectrophotometer (UV mini -1240-Japan).

Statistical analysis

The generated data was analysed by ANOVA and means were separated using LSD. The correlations between the studied parameters were done and when a significant correlation was obtained simple linear regression was done according to the following model $y = a + b x$. The statistical analysis was done using SPSS programme (version 16).

Results

Metabolites

The mean values of serum, carpal and tarsal joint of total protein (TP), albumin, globulin (Table1). Glucose in plasma, carpal and tarsal joints and alanine aminotransferase (ALT) in serum, carpal and tarsal joints were

displayed in Table 2. Serum showed significantly ($p=0.000$) higher concentration of TP, albumin, globulin and ALT compared to carpal and tarsal joints, while no significant variation was found between the two joints. Carpal joint synovial fluid showed significantly ($p= 0.024$) lower concentration of glucose than tarsal joint and serum, while no significant variation was observed between serum and tarsal joint with regard to glucose.

Correlation

A highly correlation between serum protein and globulin was observed. Also significant

correlation among synovial fluid metabolites (between total protein, albumin, globulin and ALT, between albumin and globulin and between globulin and ALT) was obtained (Table 3).

Regression

The linear regression equations of serum and synovial fluid metabolites were designed. The association between serum and synovial fluid metabolites were observed .The results indicated that, the association of synovial fluid protein with globulin is higher than that of serum (Table 4).

Table (1) Normal values (g/dl) of protein, albumin and globulin of serum and synovial fluids

Mean \pm SE				
Metabolites	Serum	Carpal joint	Tarsal joint	Sign.
T. protein	7.03 \pm 0.22 ^a (6.53 - 7.52)	0.87 \pm 0.14 ^b (0.56 - 1.18)	0.57 \pm 0.09 ^b (0.36 - 0.79)	.000
Albumin	2.50 \pm 0.12 ^a (2.24 - 2.77)	0.37 \pm 0.05 ^b (0.25 - 0.48)	0.27 \pm 0.03 ^b (0.21 - 0.33)	.000
Globulin	4.52 \pm 0.26 ^a (3.9 - 5.1)	0.50 \pm 0.11 ^b (0.25 - 0.75)	0.26 \pm 0.07 ^b (0.10 - 0.42)	.000

Table (2) Normal values of Plasma glucose and ALT of serum and synovial fluids.

Mean \pm SE				
Metabolites	serum	Carpal joint	Tarsal joint	Sign.
Plasma glucose (mg/dl)	40.56 \pm 4.5 ^a (30.5 - 50.6)	27.43 \pm 2.5 ^b (21.8 - 33.04)	38.29 \pm 2.9 ^a (31.8 - 44.8)	.024
ALT (U/L)	15.77 \pm 1.13 ^a (13.3 - 18.3)	8.94 \pm 1.77 ^b (4.99 - 12.89)	5.45 \pm 0.96 ^b (3.32 - 7.58)	.000

a ,b: means within the same row followed by different superscripts are significantly ($p<0.05$)different.

Table 3 Correlations between some serum (lower triangle) and synovial fluid (upper triangle) metabolites.

Metabolites	T. protein	Albumin	Globulin	Glucose	ALT
T. protein		0.693**	0.933**	0.136	0.787**
Albumin	-0.024-		0.444*	-0.126-	0.374
Globulin	0.894**	-0.467-		0.155	0.816**
Glucose	-0.117-	0.287	-0.219-		0.086
ALT	-0.007-	-0.126-	0.056	0.376	

** correlation is significant at the 0.01 level.

* correlation is significant at the 0.05 level.

Table 4 Regression equations of some serum and synovial fluid metabolites.

Serum	Synovial fluids
Globulin (g/dl) = - 2.887 + 1.054 T. protein(g/dl) (r ² = 0.80)	Albumin (g/dl) = 0.154 + 0.237 T. protein(g/dl) (r ² = 0.48)
	Globulin (g/dl) = -0.155 + 0.742 T. protein(g/dl) (r ² = 0.87)
	ALT (U/L) = 0.399 + 9.422 T. protein(g/dl) (r ² = 0.62)
	ALT (U/L) = 2.520 + 12.279 globulin(g/dl) (r ² = 0.67)
	Globulin (g/dl) = 0.047 + 1.046 albumin(g/dl) (r ² = 0.20)

a , b: means within the same row followed by different superscripts are significantly (p<0.05) different

Discussion

In this study, the mean concentration of total protein, albumin, globulin, plasma glucose and Alanine amino transferase (ALT) were analyzed in serum, carpal and tarsal synovial fluid. The serum metabolites of the present work lie within the normal range of cattle (Radostits *et al.*, 2007). The total protein percentage in carpal joint and tarsal joint were **12.4%** , **8.11%** respectively, to that of serum. Also in cattle, the protein concentration was obtained in synovial fluid, which is less than 2g/100 ml (Chauhan and Agarwal, 2006). This finding is in close agreement with our results of carpal (0.87 ± 0.14 g/dl) and tarsal (0.57 ± 0.09 g/dl) total protein concentration. Similar findings were obtained in carpal joint of buffalo (0.89 ± 0.058 g/dl) (Baniadam and Razi Jalali, 2005), which belongs to the same family of cattle.

The albumin concentration in metacarpophalangeal joint of cattle was obtained (0.45 ± 0.17 g/dl) (AbdEllah *et al.*, 2012) which accords with the range of this work with regard to the carpal and tarsal joints, these observations concluded that there is no difference in albumin concentration between different joints of cattle.

According to results obtained in the current study, no significant difference between plasma glucose and tarsal synovial fluid glucose. Similar findings were observed between serums and stifle joint of cattle (S.A.Omer, April.14.2019. Personal. Communication.). In the present work, the glucose concentration range of carpal joint is (21.8 - 33.04 mg/dl). The glucose concentration obtained by Nazifi *et al.* (2012) in DIJ of cattle (32.55 ± 0.52 mg/dl) lie within our range . In the current work, the significant differences observed between plasma glucose and carpal synovial fluid on one hand and between tarsal and carpal synovial fluid on the other hand, may be due

to limitation of carpal joint movement against tarsal joint movement.

The maximum value of the ALT mean concentration (3.97 ± 1.09 unites/L) (Nazifi *et al.*, 2012) in DSB of cattle is in accordance with the range of the present work.

The minimum value of globulin mean concentration (1.80 ± 1.18 g/dl) in metacarpophalangeal joint of cattle (AbdEllah *et al.*, 2012) is in accordance with the carpal and tarsal joint range of the present work. Also the accordance of protein concentration in the carpal joint of buffaloes (Baniadam and RaziJalali, 2005), with our study lead to agreement of globulin concentration of this work with ours.

In this study, the positive correlations were made between T. protein, albumin globulin and ALT, between albumin and globulin and between globulin and ALT of synovial fluid; this may be due to the fact that all the correlated metabolites are proteins. The subsequent linear regression equations were designed for the first time, thus no comparable results were found.

Conclusion

Normal values of synovial fluid in carpal and tarsal joints were determined. The total protein percentage of carpal and tarsal joint against serum was obtained .Also the correlation and linear regression equations among serum and synovial fluid metabolites were established. These equations will be useful in estimation the concentration of one metabolite from other. All these information will help in diagnosis of various joint disorders and diseases.

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دراسة مقارنة بين مستقلبات السائل الزليلي ومصل الدم في الماشية السودانية

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المستخلص

أجريت هذه الدراسة للحصول على القيم الطبيعية للبروتين الكلي، الألبومين، القلوبولين، الجلوكوز وناقل الامين (ALT) من السائل الزليلي لأبقار البقارة السودانية، مقارنة هذه النتائج مع القيم المناظرة لها في مصل الدم، وللكشف عن الارتباط والإعتماد لهذه المستقلبات في مصل الدم والسائل الزليلي. إحدى عشرة عينة دم وإحدى وعشرون عينة سائل زليلي تم جمعها من مفصلي الرسغ والعرقوب لثيران البقارة الصحيحة إكلينيكيًا، في الفترة ما بين شهري يناير ومارس من العام 2018 . تم استخدام طرق القياس اللوني لتحديد تركيز هذه المستقلبات باستخدام محاليل تجارية تم تحليلها بجهاز مقياس الطيف الضوئي(UV mini-1240-Japan) . تم تحليل البيانات بإختباري تحليل التباين و اقل إختلاف معنوي باستخدام برنامج التحليل الإحصائي SPSS (النسخة 16) . تركيز الجلوكوز في مفصل الرسغ كان أقل معنويًا (قيمة إحصائية = 0.024) مقارنة مع مفصل العرقوب ومصل الدم، وكانت المستقلبات الأخرى أقل معنويًا (قيمة إحصائية = 0.000) من مصل الدم . تم الحصول على الارتباط (بين البروتين الكلي ، الألبومين، القلوبولين و ALT ، بين الألبومين والقلوبولين وما بين القلوبولين و ALT) و المعادلات الخطية البسيطة الملحقة له لمستقلبات السائل الزليلي. خلصت الدراسة إلي أن هذه البيانات المتحصل عليها ستكون ذات فائدة للتفرقة بين المفاصل الطبيعية وغير الطبيعية للماشية السودانية والتي ستساعد في تشخيص إضطرابات وأمراض المفاصل، وتوصي الدراسة بإجراء دراسات أخرى للسائل الزليلي في عدد كبير من الأبقار، ومقارنة مستقلبات السائل الزليلي مع مستقلبات سائل الجسم الأخرى كالسائل النخاعي الشوكي.