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ORIGINAL ARTICLE

Comparative studies on biochemical and cytological constituents of synovial fluids in some farm animals

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Abstract The goal of the present study was to evaluate the difference in the synovial fluid constituents in cattle, buffaloes, camels, and donkeys. A total number of 20 clinically healthy adult male animals (cattle (N=5), buffaloes (N=5), camels (N=5), and donkeys (N=5) were subjected to study. Synovial fluid samples were collected from the metacarpophalangeal joint under complete aseptic conditions. The samples were examined physically, cytologically, and biochemically. Synovial fluid analysis revealed significant variations in specific gravity, total leukocyte counts, total proteins, albumin, globulins, glucose levels, and alkaline phosphatase activity among investigated animal species.

Keywords Synovia · Buffaloes · Cattle · Camel · Donkey

Introduction

Normally, synovial fluid (SF) is clear, pale yellow, viscid, and does not form a clot. Synovial fluid is believed to have two main functions: to aid in the nutrition of articular cartilage by acting as a transport medium for nutritional substances, such as glucose, and to aid in the mechanical function of joints by lubrication of the articulating surfaces (O'Farrel and Costello 1980).

Studies of mammalian synovial fluid have found considerable similarities among species, although notable differences do exist. The majority of investigative work determining the composition of synovial fluid had been performed on bovine synovia because large quantities of it are available (Lipowitz 1985). The major difference between synovial fluid and other body fluids derived from plasma is the high content of hyaluronic acid (hyaluronan) in synovial fluid (Lipowitz 1985; Mcllwarith et al. 2001). The source of hyaluronan is the synovial membrane. Hyaluronan provides the synovial fluid with a number of unique properties. It imparts a high viscosity to the fluid. It acts as a boundary lubricant for the synovial membrane and influence the further composition of the fluid (O'Farrel and Costello 1980). Therefore, the decrease in viscosity may be due to a change in the overall relationship of hyaluronan and other molecules in addition to simple depolymerization (Swan 1978).

To obtain synovial fluid, arthrocentesis is usually done. Arthrocentesis is the aspiration of synovial fluid from a joint cavity. Arthrocentesis is used to diagnose and make treatment decisions regarding a joint. Obtaining synovial fluid is safe, simple, relatively pain-free, inexpensive, and extremely beneficial to the patient (Werner 1979).

Synovial fluid analysis should be an integral part of any diagnostic evaluation of an animal with lameness, especially the animal with joint effusion (Werner 1979), as it provides unique and valuable information about the affected joint and helps differentiate inflammatory from noninflammatory causes of joint disease (Reichman and Waddell 2004). It is the only method to diagnose or rule out a septic arthritis (Hasselbacher 1993; Mcllwarith et al. 2001). The fluid should be analyzed for the presence of

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crystals. A white blood cell count and differential may help identify the causes of an effusion (Reichman and Waddell 2004). Synovial fluid has been used as an indicator of articular pathology for many years (Van Pelt 1962).

Vascular permeability and synovial membrane permeability are altered by inflammation, which accounts for protein content changes in diseased synovial fluid. Immunoglobulins, immune complexes, and complement are produced by cells accumulating in the inflamed synovial membrane and periarticular lymph nodes and find their way to the synovial fluid (Bunch et al. 1974). A decreased amount of synovial fluid glucose may be associated with articular diseases, particularly septic and immune-mediated arthritis (Cohen et al. 1975). In particular, the activities of several enzymes in SF have been determined for evaluating the extent and type of tissue damage in various joint diseases in the horse (Lohmander 1977). Several investigations of the synovial composition in normal and abnormal joints have been published. The differences in the cytological and biochemical constituents of synovial fluid in different animal species have not been clearly shown. The goal of the present study was evaluate the difference in the synovial fluid constituents in cattle, buffaloes, camels, and donkeys.

Materials and methods

A total number of 20 clinically healthy adult male animals (cattle (N=5), buffaloes (N=5), camels (N=5), and donkeys (N=5)), which belong to the Veterinary Teaching Hospital, Assiut University, Egypt, were subjected to the study. Synovial fluid samples were collected from the metacarpophalangeal joint under complete aseptic conditions.

Collection of synovial fluid

The synovial fluid samples were collected using sterile needle and syringes. The site for arthrocentesis was prepared aseptically. The joint was flexed to a 45° angle, and a sterile needle was inserted on the latero-palmar aspect between the lateral branch of the interosseous medius and large metacarpus at a point of 1 cm proximal to the joint (Reichman and Waddell 2004). The synovial fluid samples were divided into two parts; one part was collected on a plain tube and stored at 4°C for 2 days to test clot formation, and the other part was collected on 3.8% sodium citrate (0.1 ml/ml of synovial fluid) and mixed properly to prevent coagulation and used for physical, cytological, and biochemical analyses.

Examination of synovial fluid

Synovial fluid was examined physically, chemically, and cytologically according to Chauhan and Chandra (2007)

Physical examination

Physical examination included color, viscosity, coagulation, odor, transparency, specific gravity, and clot formation. Specific gravity was measured using test strep (Medi-Test Combi 10® SGL, Macherey-Nagel, Germany). Samples for testing clot formation was kept in a refrigerator up to 2 days.

Cytological examination

Total synovial leukocyte counts were measured in synovial fluid using hemocytometer method according to Coles (1986), and then synovial fluid samples were centrifuged; the supernatant fluid was collected in Eppendorf tube and kept for biochemical analysis. A drop from the sediment was applied on a microscopic slide and examined for number of leukocytes/high power field (leukocytes/HPF).

Chemical examination

The following biochemical constituents were measured in synovial fluid samples: total proteins (g/dl), albumin (g/dl), globulins (g/dl), glucose (mg/dl), chloride (mmol/l), creatine phosphokinase (CPK, units per liter), gamma glutamyl transferase (GGT, units per liter), alkaline phosphatase (ALP, units per liter), and lactate dehydrogenase (LDH, units per liter) using commercial test kits supplied by Spectrum Diagnostics (Spectrum Diagnostics, Cairo, Egypt) and by means of digital VIS/ultraviolet spectrophotometer (Cecil instruments, Cambridge, England, Series No. 52.232). Synovial fluid pH was measured using test strip (Medi-Test Combi 10® SGL, Macherey-Nagel, Germany). Mucin test was performed according to the method described by Chauhan and Chandra (2007). Briefly, in a small test tube, 0.8 ml distilled water and one drop of 7N acetic acid were mixed thoroughly. To this, 0.2 ml synovial fluid was added in such a way that the fluid does not come in contact with glass of test tube, and the fluid was mixed thoroughly by swirling and allowed to stand for 1 h at room temperature.

Statistical analysis

Data are presented as mean and standard deviation (mean ± SD). Statistical significance was determined by the analysis of variance using Statistical Package for the Social Sciences for Windows (SPSS, version 10.0, Chicago, IL, USA). Data from studied groups were tested for difference using least significant difference test. Statistically significant differences were determined at $p \leq 0.05$.

Table 1 Specific gravity, pH, and total leukocyte counts in synovial fluid

	Cattle $N=5$	Buffaloes $N=5$	Camels $N=5$	Donkeys $N=5$
Specific gravity	1.009±2.23bc	1.007±2.73ac	1.006±2.23a	1.010±0.0b
pH	8.00±0.00a	8.20±0.44a	8.00±0.0a	8.00±0.0a
WBCs/HPF	4.60±4.09a	3.00±1.41a	16.33±5.03b	1.60±0.89a
Total leukocyte counts/mm^3	193.2±68.51c	78.60±32.09a	460.33±56.58b	127.60±42.6ac

Data expressed as mean ± SD. Values followed by different lowercase letters are significant

Results

Physical analysis

Synovial fluids from animals under investigation were colorless, odorless, transparent, and viscous. No coagulation was detected in synovial fluid stored at 4°C for 2 days.

Synovial fluid specific gravity (1.010±0.0) was significantly higher in donkeys than buffaloes (1.007±2.73) and camels (1.006±2.23). On the other hand, specific gravity for synovial fluid from cattle (1.009±2.23) was significantly higher than that of camels (Table 1).

Cytological analysis

There were significantly higher leukocytes/HPF and synovial total leucocytic count in camels' synovial fluid ($p<0.01$) than those from other animals. Synovial total leucocytic counts in cattle were significantly higher in cattle ($p<0.01$) than in buffaloes.

Biochemical constituents of synovial fluids

Biochemical analysis of SF (Table 2) revealed significantly higher synovial total proteins in donkeys than that of cattle ($p<0.05$), buffaloes ($p<0.01$), and camels ($p<0.01$). Synovial albumin was significantly higher in camels ($p<0.01$) than other animal species.

Globulins were significantly higher in donkeys than that of cattle ($p<0.05$), buffaloes ($p<0.01$), and camels ($p<0.01$). Furthermore, synovial globulins were significantly higher in cattle ($p<0.05$) than in camels.

There was a significantly higher ($p<0.05$) glucose level in buffaloes' synovial fluid than in cattle. Chloride level ($p<0.01$) was significantly higher in cattle than in buffaloes, camels, and donkeys.

There were no significant differences in synovial CPK, GGT, and LDH activities in different animal species. Synovial ALP activity was significantly higher ($p<0.01$) in cattle than buffaloes ($p<0.05$), camels ($p<0.01$), and donkeys ($p<0.05$).

Results for the mucin test were the same in different animal species and appear as a tight ropy clump in a clear solution.

Discussion

Arthrocentesis is indicated to evaluate the cause of arthritis or a joint effusion. All patients presenting with an acute monoarthritis or an acute nontraumatic effusion should undergo arthrocentesis. The current study aimed to compare the physical, biochemical, and cytological constituents of a normal SF in different animal species. Except for SF specific gravity, physical characters of synovial fluids agreed with previous studies (Chauhan and Chandra 2007). Specific gravity was reported to be from 1.010 to 1.015 (Chauhan and Chandra 2007) in different animals. In the current study, the lowest mean value for synovial specific gravity was in camels (1.006±2.23) and

Table 2 Synovial biochemical constituents in different animal species

	Cattle $N=5$	Buffaloes $N=5$	Camels $N=5$	Donkeys $N=5$
Total proteins (g/dl)	2.25±1.15a	1.43±0.32a	1.64±0.29a	3.39±0.88b
Albumin (g/dl)	0.45±0.17a	0.50±0.23a	0.97±0.17b	0.39±0.14a
Globulins (g/dl)	1.80±1.18a	0.92±0.15ab	0.66±0.31b	2.99±1.00c
Glucose (mg/dl)	59.84±24.61a	101.96±37.65b	78.87±31.25ab	–
Chloride (mmol/l)	348.1±151.9a	177.48±34.35b	79.99±45.09b	101.79±8.15b
CPK (U/l)	76.66±22.97a	92.65±52.24a	105.32±45.31a	113.32±57.0a
GGT (U/l)	10.42±4.97a	15.04±5.72a	11.69±9.94a	17.07±5.11a
LDH (U/l)	65.92±33.61a	102.17±56.03a	91.56±41.74a	68.66±17.18a
ALP (U/l)	135.37±56.85a	69.84±36.40b	60.34±38.87b	62.11±18.06b

Data expressed as mean ± SD. Values followed by different lowercase letters are significant

the highest was in donkeys (1.010±0.0). Failure of clot formation in synovial fluid up to 2 days indicated that normal synovial fluid can be collected without using an anticoagulant.

The highest significant increase in leukocytes/HPF and in total leukocytes counts was in camels, which indicated that the number of leukocytes/HPF in SF sediment can be used as an indicator for synovial total leukocyte counts. The total leukocyte count for donkeys was 127.60±42.6, which is less than that reported by Chauhan and Chandra (2007) in equine synovial fluid. In buffaloes, it was significantly higher than that of cattle (Table 1). Results for total leukocyte counts in cattle agreed with that reported by Chauhan and Chandra (2007). Normally, SF contains few leukocytes, which is varied according to animal species (Todhunter 1996).

In the present study, synovial total proteins and globulins were significantly higher in donkeys; albumin was significantly higher in camels than other investigated animals. Total protein in donkeys was 3.39±0.88, which is higher than its content in previous reported value in horse SF (1.08 g/dl; Korenek et al. 1992). In buffaloes, total proteins and glucose levels were 1.43±0.32 g/dl and 101.96± 37.65 mg/dl, respectively. Their levels had been reported to be 0.89±0.055 g/dl and 59.25±3.39 mg/dl, respectively (Baniadam and Jalali 2005), and 1.02 g/dl and 0.9 g/dl respectively (Krishnamurthy and Tyagi 1973; Soliman et al. 1975). Synovial total proteins and glucose levels in investigated cattle were 2.25±1.15 g/dl and 59.84± 24.61 mg/dl, respectively. Their levels had been reported to be 0.91±0.17 and 61.59±6.06 mg/dl, respectively (Mojabi et al. 1991).

No significant variations were observed in enzyme activities of CPK, GGT, and LDH in SF from different animal species. Synovial ALP activity and chloride level were significantly higher in cattle than buffaloes ($p<0.05$), camels ($p<0.01$), and donkeys ($p<0.05$). Cattle synovial fluid was reported as a good source for ALP (Dabich and Neuhaus 1966), which agreed with results of the present study. In conclusion, synovial fluid constituents showed significant variations in total leukocyte counts, total proteins, albumin, globulins, chloride, and glucose levels among investigated animals. Arthrocentesis is an easy, safe, inexpensive technique which allows analysis of the synovial fluid and recognizes the different constituents of the synovial fluid, which could be used in diagnosis and differential diagnosis of different joint lesions.

References

Baniadam A, Jalali RM (2005) Studies on normal synovial fluid from the carpal joints of Iranian water buffaloes: physical and biochemical parameters. Iran J Vet Res 6:17–20

Bunch TW, Hunder GG, Offord K (1974) Synovial fluid complement: usefulness in diagnosis and classification of rheumatoid arthritis. Ann Intern Med 81:32

Chauhan RS, Chandra D (2007) Veterinary laboratory diagnosis, 2nd edn. International Book Distributing Company, Lucknow

Cohen AS, Brandt KD, Krey PK (1975) Synovial fluid in laboratory diagnostic procedures. In: Cohen AS (ed) The rheumatic disease, 2nd edn. Brown & Co, Boston

Coles EH (1986) Veterinary clinical pathology, 4th edn. Saunders, Philadelphia

Dabich D, Neuhaus OW (1966) Purification and properties of bovine synovial fluid alkaline phosphatase. J Biol Chem 24:415–420

Hasselbacher P (1993) Arthrocentesis, synovial fluid analysis, and synovial biopsy. In: Schumaker HR Jr, Klippel JH, Koopman WJ (eds) Primer on rheumatic diseases, 10th edn. Arthritis Foundation, Atlanta, pp 67–72

Korenek NL, Andrews FM, Maddux JM, Saunders WL, Faulk DL (1992) Determination of total protein concentration and viscosity of synovial fluid for the tibiotarsal joint of horses. Am J Vet Res 53:781–784

Krishnamurthy D, Tyagi RPS (1973) Studies on stifle synovial fluid of bovines. II. Certain biochemical parameters of the fluid in normal buffalo calves. Indian Vet J 50:79–82

Lipowitz AJ (1985) Synovial fluid (chapter 86). In: Newton CD, Nunamaker DM (eds) Textbook of small animal orthopaedics. International Veterinary Information Service, Ithaca

Lohmander L (1977) What is the current status of biochemical markers in the diagnosis, prognosis and monitoring of osteoarthritis? Baillières Clin Rheumatol 11:711–726

Mcllwarith WC, Billinghurst RC, Frisbie DD (2001) Current and future diagnostic means to better characterize osteoarthritis in the horse—routine synovial fluid analysis and synovial fluid and serum markers. Proc Amer Assoc Equine Practnr 47:171–179

Mojabi A, Nowrouzain I, Safi S (1991) Evaluation of LDH, ALT and AST levels in normal synovial fluid of cattle. J Fac Vet Med Univ Teh 46:81–100

O'Farrel T, Costello BG (1980) Osteochondritis dissecans of the talus: the late results of surgical treatment. J Bone Jt Surg 62B:131

Reichman EF, Waddell R (2004) Arthrocentesis. Chapter 65, In: Reichman E, Simon R (eds) Emergency Medicine Procedures. McGraw-Hill Medical Publishing Division, New York, Chicago, Sydney, Toronto, pp. 559–584.

Soliman MK, Elamrosi S, Youssef LB (1975) TI: studies on the normal synovial fluid from the tibio-tarsal joint of buffaloes. Indian Vet J 52:19–23

Swan DA (1978) Macromolecules of synovial fluid. In: Sokoloff L (ed) The joints and synovial fluid. Academic, New York

Todhunter RJ (1996) Anatomy and physiology of synovial joints. In: McIlwraith CW, Trotter GW (eds) Joint disease in the horse. Saunders, Philadelphia, pp 1–28

Van Pelt R (1962) Properties of equine synovial fluid. J Am Vet Med Assoc 143:738–748

Werner LL (1979) Arthrocentesis and joint fluid correlates with synovial analysis: diagnostic applications in joint diseases of small animals. Compendium Contemp Ed 1:855