

Effect of iron dextran injections on some synovia markers of Iraqi sport horses

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Abstract

In the past decades, many veterinarians and trainers had been tried to use iron preparations as performance enhancers for racing, or for treatments such as "anemiä. The study was conducted on (12) Iraqi sport horses. The principal aims of the study were to investigate the effect of iron dextran on some synovial fluid constituents at the rest and the endurance exercise of Iraqi sports horses. The physical markers of synovial fluids of the sports horses which are included in this study, shows: (TG-1) (gallop and iron dextran treatment) are mostly slight turbid, dark yellow, very low viscid, and normal mucin clot formation. (TG-2) (Iron dextran only) are mostly clear, yellow, viscid and normal mucin clot formation. While (CG-3) (distil water only) are mostly clear, pale yellow, viscid and fair mucin clot formation. The effect of iron dextran on some biochemical markers of the synovial fluid constituents of the sports horses which included in the study were shows as in table-3: the PH value of (TG-1) was very low than normal 6.15 ± 0.85 which was significantly different at $P \leq 0.05$ with the PH value of the (TG-2) 6.64 ± 0.36 while the PH of (CG-3) was within normal value 7.57 ± 0.85 and there was no significant different at $P \leq 0.05$ with (TG-2). The WBC value of (TG-1) was 8.61 ± 0.38 (WBC/mm³) which was different significantly at $P \leq 0.05$ with (TG-2) 6.95 ± 0.60 (WBC/mm³) but (TG-2) was not different significantly at $P \leq 0.05$ with (CG-3) 5.45 ± 0.20 (WBC/mm³). The glucose value of (TG-1) was reduced than normal 52.89 ± 1.58 (mg/dl), which was different significantly, at ($P \leq 0.05$) with (TG-2) 51.48 ± 1.12 (mg/dl), but (TG-2) was not different significantly at ($P \leq 0.05$) with (CG-3) 53.14 ± 1.04 (mg/dl). The total protein value of (TG-1) was increased than normal 3.68 ± 0.38 (g/dl) which was different significantly at $P \leq 0.05$ with (TG-2) 2.45 ± 0.23 (g/dl) but (TG-2) was not different significantly at ($P \leq 0.05$) with (CG-3) 1.52 ± 0.21 (g/dl). The hyaluronic acid values were significantly different at $P \leq 0.05$ between all groups, (TG-1) 0.33 ± 0.03 (mg/ml), (TG-2) 0.21 ± 0.03 (mg/ml), and (CG-3) 0.37 ± 0.03 (mg/ml) respectively. The alkaline phosphatase values also were significantly different at $P \leq 0.05$ between all groups, (TG-1) 38.44 ± 1.61 (IU/L), (TG-2) 27.25 ± 0.22 (IU/L), and (CG-3) 25.31 ± 1.79 (IU/L), respectively. In conclusion, iron injections had no any improvement to the sport horses' synovia.

Key words: Iron, synovial fluid, Iraqi, sport horses.

Introduction:

In the past decades, many veterinarians and trainers have been try iron preparations as performance enhancers for racing, or for treatments such as "anemiä. Since exercise induce physiological tension and can be considered a form of stress on the body which result to a number of chemical (hormonal) and cellular changes a part from physical changes as increase oxygen intake, blood pressure, body temperature (1). The load transmission which participate on all parts of articulating joints, and failure of the

bone articular cartilage, muscles, ligament/ tendons or nerves of a joint may lead to exercise induced damage (2). The normal synovial joints contain a fluid called synovial fluid (SF) comprise of a unique plasma filtrate augmented with hyaluronate and other biomolecules produce by type- B synoviocytes (3). The former, less sensitive and specific approach consists of evaluations of viscosity, color, clot formation, volume, total protein and cytological examination (4). Pale yellow, clear and free of flocculent

debris and does not clot are the physical markers of normal synovial fluid (5). Due to efficient iron incorporation into hemoglobin, iron preparations are usually administered to racing horses by intravenous route. They have shown good results in racing horses as they have a fast effect (6). Due to the fact that, iron losses are gradual iron deficiency develops slowly. The magnitude of the consequences of iron deficiency depends on the duration and on the available iron pool present in the body depot degree of the negative iron balance (6). According to (7), the clarity of the synovial fluid is found turbid at gallop in six out of ten non-trained Iraqi Arabian horses, the color is dark yellow in two horses at gallop, the viscosity is very low in all horses at gallop and the mucin clot formation is very poor in all horses at gallop. While there is decrease of PH 5.25 ± 0.09 , glucose 32.08 ± 0.98 (mg/dl). But there is an increase of WBC counts 10.57 ± 0.25 (WBC/mm³), total protein 3.12 ± 0.13 (g/dl) and alkaline phosphatase 39.17 ± 0.49 (IU/L). To the author's knowledge, no study evaluating the effect of iron dextran on the synovial fluid properties of Iraqi Arabian Sports horses is available. Study of (8) mentions that, the deposition of the iron load within the decreased joint and determines the effect of intravenous iron dextran, infused during the acute and chronic phases of the arthritis in rabbits. Little is known of the fate of iron-dextran in the rheumatoid joint, particularly with respect to its influence on disease processes. The effect of iron dextran on inflammatory synovitis is therefore investigated using an experimentally induced rabbit model of monarticular arthritis (9) which, in terms of histological appearance, gross pathology, and drug responsiveness, closely resembles the human rheumatoid disease. The principal aims of the study are to investigate the effect of iron dextran on some synovial fluid constituents at the rest and the endurance exercise of Iraqi sports horses.

Materials and Methods:

In Al-Herrah city/ Al-Najaf province at January 2016, (12) private sports Iraqi horses

were included in this study. These horses were (3-6.5) years old, (325-450) kg of body weights, (5 stallions and 7 mares), were housed in individual boxes and were fed grains, alfalfa hay, mineral blocks with free access to clean water. These horses were under private veterinary check with history of antihelmintic treatments. All horses were ridden in a regularly maintained on soil tracks. Each horse exercise includes walking, trotting, and cantering for the duration of 1 to 1.5 hours for 5 days/week.

Experimental design:

The experimental horses were divided equally and randomly into three groups: treatment group-1 (TG-1), treatment group-2 (TG-2), and control group-3 (CG-3). (TG-1) and (TG-2) had been injected, iron dextran (Kontam Pharmaceuticals, Zhongshan, CO., LTD, China), (400mg/kg B.W. I/V) dose per a week for three months, while (CG-3) had been injected intravenously, 5ml. of distal water. Only (TG-1) runs on soil track at gallop speed (70km/hr.) which is calculated by sports clock for (5) km. While (TG-2) had no endurance running. Under strict sterile conditions, arthrocentesis of mid-carpal joint was done for all groups (directly for TG-1). Synovial samples (2-3ml.) had been transferred to vacutainers tubes and sent in icebox to laboratory analysis. The physical markers were recorded, clarity, color, viscosity by dropping the synovial fluid from the end of the syringe and notice the stringing out as much as 5 cm. before separating. Mucin clot formation was measured by adding 0.5 ml. of synovial fluid to 2 ml. of 2% acetic acid and mixing it rapidly with glass rod. PH was measured by PH meter. Total white blood cells (WBC/mm³) were measured by hemocytometer counts method, while glucose (mg/dl), total protein (g/dl), hyaluronic acid (mg/ml.) and alkaline phosphatase (IU/L) were measured by spectrophotometer (Chrom Tech, V-1100 Spectrophotometer MED & Lab. Instrument, USA). Statistical analysis of the data was done for the mean values and standard errors by one way ANOVA, the P values were significant statistically at ≤ 0.05 .

Results:

The physical markers of synovial fluids of the sports horses which were included in this study, as in table-1 shown: (TG-1) (gallop and iron dextran treatment) were mostly slight turbid, dark yellow, very low viscid, and normal mucin clot formation. (TG-2)

(Iron dextran only) were mostly clear, yellow, viscid and normal mucin clot formation. While (CG-3) (distil water only) were mostly clear, pale yellow, viscid and fair mucin clot formation.

Table (1): The physical markers of synovial fluids of the experimental horses (n=12)

Groups	No.	Age (years)	Gender	Clarity	Color	Viscosity	Mucin clot formation
TG-1	1	4	female	Slight turbid	Dark yellow	Very low viscid	Fair
	2	5.5	female	Slight turbid	Yellow	Very low viscid	Normal
	3	6	male	Clear	Dark yellow	Very low viscid	Fair
	4	3.5	male	Slight turbid	Dark yellow	Viscid	Normal
TG-2	1	3	female	Clear	Yellow	Viscid	Normal
	2	4.5	male	Clear	Yellow	Viscid	Normal
	3	5	female	Slight turbid	Dark yellow	Very viscid	Fair
	4	6.5	female	Clear	Yellow	Viscid	Normal
CG-3	1	4.5	male	Clear	Pale yellow	Viscid	Fair
	2	5.5	female	Clear	Pale yellow	Low viscid	Fair
	3	6	female	Clear	Pale yellow	Viscid	Poor
	4	3.5	male	Clear	Pale yellow	Viscid	Fair

To compare the results, table-2 shows the normal values of PH, WBC, glucose, total protein, hyaluronic acid, and alkaline phosphatase and their references.

Table (2): The normal values of the biochemical markers of the synovial fluid of horses

Markers	Values	References
PH	7.34±0.12	(10)
WBC/ 10 ³ /μl	2.10±0.12	(10)
Glucose(mg/dl)	67.22±3.11	(10)
Total protein(g/dl)	4.73±0.82	(10)
Hyaluronic acid(mg/ml.)	0.33-1.5	(11)
Alkaline phosphatase(IU/L)	39.32±8.34	(10)

The effect of iron dextran on some biochemical markers of the synovial fluid constituents of the sports horses which included in the study were shows as in table-3: the PH value of (TG-1) was very low than normal 6.15±0.85 which was significantly different at $P \leq 0.05$ with the PH value of the (TG-2) 6.64±0.36 while the PH of (CG-3) was within normal value 7.57±0.85 and there was no significant different at $P \leq 0.05$ with (TG-2). The WBC value of (TG-1) was 8.61±0.38 (WBC/mm³) which was different

significantly at ($P \leq 0.05$) with (TG-2) 6.95±0.60 (WBC/mm³) but (TG-2) was not different significantly at ($P \leq 0.05$) with (CG-3) 5.45±0.20. (WBC/mm³). The glucose value of (TG-1) was reduced than normal 52.89±1.58 (mg/dl), which was different significantly, at ($P \leq 0.05$) with (TG-2) 51.48±1.12 (mg/dl), but (TG-2) was not different significantly at $P \leq 0.05$ with (CG-3) 53.14±1.04 (mg/dl). The total protein value of (TG-1) was increased than normal 3.68±0.38 (g/dl) which was different significantly at $P \leq 0.05$ with (TG-2) 2.45±0.23 (g/dl) but (TG-2) was not different significantly at $P \leq 0.05$ with (CG-3) 1.52±0.21. (g/dl). The hyaluronic acid values were significantly different at $P \leq 0.05$ between all groups, (TG-1) 0.33±0.03 (mg/ml), (TG-2) 0.21±0.03 (mg/ml), and (CG-3) 0.37±0.03 (mg/ml) respectively. The alkaline phosphatase values also were significantly different at $P \leq 0.05$ between all groups, (TG-1) 38.44±1.61 (IU/L), (TG-2) 27.25 ± 0.22(IU/L), and (CG-3) 25.31 ± 1.79 (IU/L), respectively.

Table (3): The effect of iron dextran on the biochemical markers of the synovial fluid constituents of the experimental horses: values represent means \pm SE (n=12)

Groups	PH	WBC/mm ³	Glucose (mg/dl)	Total protein (g/dl)	Hyaluronic acid (mg/ml.)	Alkaline phosphatase (IU/L)
TG-1	6.15 \pm 0.85 A	8.61 \pm 0.38 A	52.89 \pm 1.58 A	3.68 \pm 0.38 A	0.33 \pm 0.03 A	38.44 \pm 1.61 A
TG-2	6.64 \pm 0.36 B	6.95 \pm 0.60 B	51.48 \pm 1.12 B	2.45 \pm 0.23 B	0.21 \pm 0.03 B	27.25 \pm 0.22 B
CG-3	7.57 \pm 0.85 B	5.45 \pm 0.20 B	53.14 \pm 1.04 B	1.52 \pm 0.21 B	0.37 \pm 0.03 C	25.31 \pm 1.79 C

* Different letters mean significant different at $P \leq 0.05$.

Discussion:

High performance of sports horses is still the goal of many researchers in many countries, which have interest in equine industry. They try many preparations, the most famous are, iron preparations, which have a strong relationship with carrying the oxygen by red blood cells to the body cells. According to our knowledge, only (7, 12) studied the effect of exercise on some different markers of synovial fluid and blood markers in non- training Iraqi Arabian horses. Therefore, we designed this research, to study the effect of iron injections on some physical and biochemical markers of synovial fluid of Iraqi sport horses. It is worthwhile to mention that, synovial fluid play an essential role in lubricating the joints during the motion. So its constituents must be affected specially with high speed (gallop), these facts are accompanied with the physical markers results of (7), which were turbid, dark yellow, very low viscosity, and very poor mucin clot formation. The study shows that, the synovial fluids of (TG-1) will be slight turbid, dark yellow, very viscid, and normal mucin clot formation. We believe that, these changes of the physical markers are due to the effect of the high speed (gallop) which changes the chemical ratios of the synovial fluid constituents. In addition, these results indicate that, iron dextran reduce the effect of the high speed as in the study of (7). The iron dextran has better effect on the physical markers of (TG-2), the synovial fluids are clear, yellow, viscid, and normal mucin clot formation. Results shows, the biochemical changes of the synovial fluid constituents which may be related to the permeability changes of the

synovial membranes of the joints due to the high speed (gallop). The PH value was very low than normal 6.15 \pm 0.85 in (TG-1), while in (TG-2) is 6.64 \pm 0.36, these results may be due to the formation of uric acid by the effect of the high speed. The WBC counts are increased significantly in (TG-1) 8.61 \pm 0.38 (WBC/mm³), and in (TG-2) 6.95 \pm 0.60 (WBC/mm³) which is not significant. These increases explain clearly the role of the high-speed endurance exercise as a stress harmful factor to the joints. The results also shows significant reducing of glucose than normal in (TG-1) 52.89 \pm 1.58 (mg/dl), that may be indicate the breakdown of glucose molecules and releasing of energy. The total protein values are increased significantly in (TG-1) 3.68 \pm 0.38 (g/dl), we believe, may be due to the high damage of the lining cells of the joint. In addition, the alkaline phosphatase values increase significantly in (TG-1) 38.44 \pm 1.61 (IU/L), which explain the effect of exercise (high speed) as a stress on the joints. The results also shows clearly the significant reducing of hyaluronic acid values, 0.33 \pm 0.03 (mg/ml.) in (TG-1) which is in our believe due to the high destruction of the hyaluronic secretion cells which are lined the inner surface of the joint capsule and cover the joint cartilage and the viscosity of synovial is reduced. According to our results, the iron injections will not improve the synovial quality, so the most important goals to increase the physical activity and get better performance of our Iraqi sport horses are, must have many requirements like good nutrition, and routine exercises. We believe our results are very useful for veterinarians and trainers.

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