Effect of iron injections and exercise on some blood parameters and wound bleeding in Iraqi Arabian horses

Amir I. Towfik / Arabian Horses Center / University of Al- Qadisyah*
Hayder Gazi Abdul – Shaheed / Agriculture College / University of Al- Qadisyah
Ahmad Kadhim Munahi / Veterinary Medicine College / University of Al- Qadisyah
Sameer Sabri Kadhim / Arabian Horses Center / University of Al- Qadisyah

Abstract
Iron is one of the elements which are very important for the performance of the healthy horses. It play an essential role in the hemopoitic processes.

The study is conducted on (9) Iraqi Arabian horses in the Arabian horses center / Al-Qadisyah University.
The goals of the study are to focus on the effect of exercise and iron dextrin on some blood parameters and some wound markers of Iraqi Arabian horses.
The results show high increase of the blood parameters which include in the study, these parameters especially are more when exercise combine with iron injections than iron injections only.

Keywords: iron, blood, parameters, wound, bleeding, Arabian, horses.

Introduction
Iron was one of the elements which were very important for the performance of the healthy horses. It played an essential role in the hemopoitic process.

Exercise could be considered a form of stress since it induced physiological
tension on the body which resulted to a number of chemical (hormonal) and cellular changes a part from physical change as increased blood pressure, body temperature, and oxygen intake (1).

The hypothesis that a high hematocrit value and high hemoglobin concentrations were the most important conditions which had to be fulfilled in order to achieve to racing results had resulted in a massive use of iron preparations in healthy horses. This specially implied in racing horses during intensive training aimed at the prevention of anemia (2).

(3) was reported that the leukocytes responded to exercise was believed to be caused by mobilization of WBC from the marginal pool in response to catecholamines and also had been associated with the release of blood rich in lymphocytes from the spleen in horses.

Iron was playing an indispensable role in various physiological processes due to its capacity to easily accepted and donated electrons when converting between the ferrous (Fe^{2+}) and ferric(Fe^{3+}) forms (4).

Hematologic parameters, particularly the erythrocyte count, hemoglobin concentration and packed cell volume(PCV) had been use as indexes of readiness to race and performance potential. It had been recognize that when these parameters were abnormal, either too high or too low, horses often perform below expectation. Iron supplementation in the diet and or by parenteral injection as an almost universal practice for Thoroughbred horses in race training in North America (5).

(6) studied the effects of Fe-saccharate (Hippiron) (Jessen- Cilagsa Vifor Inc., St. Galen) in the English Thoroughbred and Yugslav Trotter, with a special in sight into the negative side effects of this preparation. In 60% of the studied horses there was a sharp increase in erythrocyte count and hemoglobin concentration. This showed a positive effect on the racing performances of the studied horses. About 10% of those horses did not show any significant changes, in 31.5% after an initial rise the Hb concentration and erythrocyte count decreased by 30% compared to the initial values, accompanying marked hyper-bilirubinemia was recorded.

Leukocytes protected the body against infection while the erythrocytes function in caring oxygen from the lungs to the rest of the body. Thrombocytes, the smallest type of blood cells, functioned in blood clotting (7).

Iron deficiency developed slowly due the fact that iron losses were gradual. The magnitude of the consequences of iron deficiency depended on the duration and degree of the negative iron balance, as well as on the available iron pool presented in the body depot. Severe iron deficiency with clinical manifestations could occur in cases of massive hemorrhage(8).

Efficient energy metabolism was possible only in the presence of oxygen, hence the need for a high hemoglobin concentration as a prerequisite for efficient oxygen binding and transport (8).

The aims of this research are to study the effects of iron injections and exercise on some blood parameters and some wound markers of Iraqi Arabian horses as a model.
Materials and methods
Nine Iraqi Arabian horses (4-5 years), (350-450±30 kg. of body weight), (2 stallions and 7 mares) in the Arabian Horses Center/Al-Qadisya University are used. They are housed individually in boxes and are fed grain, alfalfa hay with free access water.
All horses have daily clinical examinations for one week to check their healthy conditions with history of anthelmintic treatment.
Blood samples are collected by jugular venipuncture in EDTA tubes for blood analysis the normal blood parameters by use commercial kits special for each parameter. Also under routine surgery, all horses have cutaneous incisions (10 cm) at the left side of the neck region to estimate their bleeding time (Dukes method) (9) and clot formation time (Lee-White method) (9). Their specific oxygen content (SPO$_2$) are measured by Vet Pulse Oximeter (RPO-60V, Brandnew, Beijing, China) at their tongues.
After (24) hours, the horses are divided randomly and equally into (3) groups, treatment group-1 (TG-1), treatment group-2 (TG-2) and control group-3 (CG-3). (TG-1) have injected iron dextran (Kontam Pharmaceuticals, Zhongshan, CO., LTD, China). (400 mg/kg B.W. I/V) daily for one week before canter running at speed 40-50 km/hour by sports clock for 500 meter distance, then blood samples are taken for blood parameters analysis, under routine surgery, cutaneous incisions at the right side of their neck regions are done to estimate bleeding time and clotting time. While (TG-2) are dialed at the same matter but without running. At the same period (CG-3) are injected (10 ml.) distilled water and are dialed as other treatment groups.
The blood parameters which are measured: PH value, Hb, ESR, platelets count, RBC count, WBC count, PCV, glucose, blood urea nitrogen, creatinine, uric acid, albumin, total serum protein, alkaline phosphatase, creatine phosphokinase, SPO$_2$, bleeding time, clot formation time.
Statistical analysis is done after determining the mean values and standard deviations of the studied parameters. The significant of differences is calculated using (T-test) at level P≤0.05 (10).

Table-1: Pre and post-treatment values of blood parameters of the experimental horses

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</thead>
<tbody>
<tr>
<td>PH values</td>
<td>7.23±0.4 A</td>
<td>7.52±0.9 A</td>
<td>7.32±0.8 A</td>
<td>7.51±0.9 A</td>
<td>7.37±0.8 A</td>
<td>7.34±0.07 A</td>
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<tr>
<td>Hb g/dl</td>
<td>14.7±0.5 B</td>
<td>18.4±0.7 A</td>
<td>15.63±0.5 A</td>
<td>17.16±0.6 A</td>
<td>13.1±0.5 A</td>
<td>13.0±0.3 A</td>
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<tr>
<td>ESR mm/hr</td>
<td>35.83±9.1 A</td>
<td>43.06±9.7 A</td>
<td>35.45±8.9 A</td>
<td>40.86±9.0 A</td>
<td>44.8±1.1 A</td>
<td>43.68±1.4 A</td>
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<tr>
<td>Platelets count g/l</td>
<td>329±20.7 B</td>
<td>632±25.8 A</td>
<td>318.66±29.3B</td>
<td>492.66±38.6 A</td>
<td>298±37.2 A</td>
<td>281.6±29.8 A</td>
<td></td>
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<tr>
<td>RBC 10$^6$/µl</td>
<td>9.71±0.3 B</td>
<td>13.21±0.6 A</td>
<td>9.65±0.4 B</td>
<td>5.66±1.8 A</td>
<td>9.53±0.1 A</td>
<td>9.55±0.1 A</td>
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</tbody>
</table>
### Table-2: Pre and post-treatment values of the wound bleeding parameters of the experimental horses (mean ± standard error)

<table>
<thead>
<tr>
<th>Parameter</th>
<th>TG-1</th>
<th>TG-2</th>
<th>CG-3</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bleeding time (minutes)</td>
<td>2.66±0.1</td>
<td>B</td>
<td>7.5±0.7</td>
</tr>
<tr>
<td></td>
<td>5.63±0.4</td>
<td>A</td>
<td>3.7±0.1</td>
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<tr>
<td>Clotting time (minutes)</td>
<td>4.13±0.2</td>
<td>B</td>
<td>9±0.7</td>
</tr>
<tr>
<td></td>
<td>7.66±0.7</td>
<td>A</td>
<td>4.9±0.4</td>
</tr>
</tbody>
</table>

*Similar letters: no significant variations.

**Different letters: significant variations at P≤ 0.05.
Discussion

Blood plays an essential role during exercise. Its components provide transport of oxygen, electrolytes, hormones, water and nutrients to the exercising muscle, and at the same time provide removal of CO$_2$ and metabolism waste products.

To increase the physical activity and get good performance of the horses, they must have many requirements. Routine exercise and iron preparations are the most important for these goals.

Many articles in the equine sport medicine literature have studied different training schedules in order to prepare and improve the athletic condition of horses, and to avoid muscular damage(11). Hemodynamic changes have occurred in the horses that have attended, which have been evidenced by significant difference between pre-treatment and post-treatment concentration of the variables analyzed in the present study.

Knowledge of exercise induced inflammatory reaction is rather limited in equine sport medicine. There are several studies that have demonstrated that exercise alter the immune response (12). Table-1 show an significant increase of WBC of group-1 (exercise plus iron parenteral) and group-2(iron parenteral only).

Increased serum total protein and serum albumin concentrations in post samples (table-1) can be explained by increased plasmatic concentrations due a loss of part of the water from plasma, which is commonly encountered in athletes.

Our results show that increments of serum albumin of (TG-1:4.41±0.08) and (TG-2:3.73±0.04) and serum total protein of (TG-1:6.44±0.1) and (TG-2:5.29±0.2).This increase, however, cannot be attributed only to plasma volume contraction and fluid shifts.

Equine sport medicine studies report that a large number of erythrocytes are stored in the spleen. During exercise catecholamine-induced spleen contraction occurs and splenic pool erythrocytes are released into circulation.

Other relevant finding evidenced by our results is represented by increase of serum creatinine concentration (TG-1:1.75±0.05) and (TG-2:1.20±0.2).

During exercise β-adrenergic secretion will influence renal vascular smooth muscle determining the contraction of afferent arterioles. Renal perfusion is decreased and blood flow is directed to support other organs like brain, heart and muscles. In equine athletes has been evidenced that renal blood flow can be decreased by exercise up to 70% (13).

Table-1 show an increase of Hb values, RBC count, and SPO$_2$%, we believe that there is a strong relationship between these parameters. Exercise will stimulate releasing of hemoglobin from RBCs and the hemoglobin will carry more oxygen to the body tissues under the effect of iron.

Hemostasis topic in equine sport medicine has not been extensively studied as in human athletes. Nevertheless, several studies performed on horses in training and on horses performing different types of exercises (15), (16), (17), (18),(19). Table-2 show high increase of post bleeding time of (TG-1:7.5±0.7) and
(TG-2:5.63±0.4), and high increase of clotting time of (TG-1:9±0.7) and (TG-2:7.66±0.7), these results are accompanied with (20).

Some studies have evidenced that exercise can influence platelet function in horses. The mechanisms proposed for exercise-induced platelet aggregation are spontaneous platelet aggregation, shear stress induced platelet activation (21). Other studies, using different type of exercise and training sessions have evidenced increased blood coagulation following exercise(15),(19). Hemostatic alterations in horses following exercise are mainly attributed to endothelial activation of the factors involved coagulation-fibrinolysis cascades. We regard that our research is the first study conclude the Iraqi Arabian horses. Its results are very important for veterinarians and trainers of racing horses in our country.

References
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