Sudan University of Science and Technology

College of Veterinary Medicine

Studies on Effects of Exercise on Complete Blood Count of Horses (*Equus caballus)* in Khartoum State

Graduation Research Project
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بسم الله الرحمن الرحيم

﴿وَالْخَيْلَ وَالْبِغَالَ وَالْحَمِيرَ لِتَرْكَبُوهَا وَزِينَةً ۚ وَيَخْلُقُ مَا لَا تَعْلَمُونَ﴾

سوره النحل (8)

Dedication

We dedicate this work to our Mothers, Fathers, Sisters, Brothers and Friends with sincere love and loyalty.

Acknowledgements

We thank our Allah who gave us the strength and patience to conduct and finish this work. We are grateful to professor/ Shadia Abdelatti for her support, supervision of this work and motherly compassion.
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Abstract

This study was conducted at El khawjia Farm –Khartoum North during the period may 2017 to evaluate the effect of exercise on the complete blood count (CBC) of ***Equus caballus***. Four mature non pregnant, non lactating and not on estrous mares were used. The exercise program was 30 minutes moderate running, in an area of 1600 m^2. Blood samples were taken from the jugular vein in tubes containing EDTA. Blood samples were taken before exercise, immediately post exercise and an hour post exercise.CBC was determined by an automated blood cell counter. The collected data were analyzed by ANOVA test using **S**tatistical Package for **S**ocial **S**cience (SPSS16.0).

The result showed that horses, immediately post exercise, had significantly higher ($P \leq 0.05$) values than before exercise for erythrocytes count (7.27±1.40 vs. 6.44±0.69) ($x10^{12}$/L), hemoglobin concentration (Hb g/dl), (13.46±1.50 vs .11.58 ±1.22), Hematocrit (HCT%), (38.61±7.68 vs. 34.12±4.72), total leucocytes count (10.36±2.09 vs. 9.56±1.21) ($x10^{9}$/L), lymphocytes percentage(8.26±4.86 vs.6.68±3.63) ($x10^{9}$/L) and lower values for platelets count (75.82±34.49 vs. 93.797±31.92) ($x10^{9}$/L) respectively. The animals, at an hour post exercise, registered the following mean values for RBCs(6.42±0.63),Hb(11.34±1.38),HCT(34.19±4.98), WBCs(9.17±1.33), lymphocytes (5.83±2.50)% and platelets coun(91.50±29.86) which are significantly different from both the values that were recorded before exercise and immediately post exercise. However exercise did not cause any significant variations in the rest of the studied CBC parameters that is mean corposcular volume (MCV fl), mean corposcular hemoglobin (MCH pg), mean corposcular hemoglobin concentration , red cell distribution width standard deviation (RDW-SD fl), red distribution width coefficient (RDW-CV %), lymphocytes (LYM %), maximum immature ditribution (MID %), granulocyte (GRAN %), plateletcrit (PCT %),

platelet distribution width (PDW fl), platelets large cell ratio (PLCR %).

It is concluded that moderate exercise causes some changes in horses CBC and they will not return to the values before exercise after having an hour of rest.

ملخص البحث

أجريت هذه الدراسة في مزرعة الخواجية – شمال الخرطوم خلال الفترة شهرخمسه لتقييم تأثير التمرين على مكونات الدم (CBC) من الفصيله الخيليه أربع فرسات ناضجة غير حامل، غير مرضعة و وليس على فرس استروس استخدمت. أخذت عينات الدم من الوريد الوداجي في الأنابيب التي تحتوي على إدتا. أخذت عينات الدم قبل التمرين مباشرة بعد التمرين وبعد ساعة من التمرين.تم تحديد الكابينة من خلال عداد خلايا الدم الآلي. تم تحليل البيانات التي تم جمعها من قبل ANOVA Test باستخدام الحزمة الإحصائية للعلوم الاجتماعية (SPSS16.0). سجلت الخيول، مباشرة بعد التمرين، قيم أعلى بكثير ($P\leq0.05$) من قبل ممارسة العد الكريات الدم الحمراء (7.27 ± 0.691.40 ± 6.44 vs)($x10^{12}/L$)، تركيز الهيموغلوبين ,(Hb g/dl) ، (13.46 ± 1.221.50 ± vs11.58) ، الهيماتوكريت (HCT٪)، (38.61 ± 4.727.68 ± 34.12 مقارنة)، مجموع عدد كريات الدم البيضاء (10.36 ± 2.09 مقارنة1.21 ± 9.56) ($x10^9/L$) ، نسبة اللمفاويات (8.26 ± 4.86 75.82) (3.63 ± 6.68 34.49 ± مقارنة 93.797 ± 31.92) ($x10^{12}/L$) على التوالي. سجلت الحيوانات بعد ساعة من التمرين القيم المتوسطة التالية ل كرات الدم الحمراء (6.42 ± 0.63)($x10^{12}/L$) ، Hb (11.34 ± 1.38)(g/dl) ، HCT(34.19 ± 4.98)%، WBCs (9.17 ± 1.33)($x10^9/L$) الخلايا الليمفاوية (5.83 ± 2.50) ($x10^9/L$) وعدد الصفائح الدموية (91.50 ± 29.86) ($x10^9/L$) التي تختلف اختلافا كبيرا عن كل من القيم التي تم تسجيلها قبل ممارسة وبعد مباشرة ممارسة الرياضة. لم يتسبب التمرين في حدوث أي اختلافات معنوية في بقية مكونات الدم (CBC) المدروسة التي تعني متوسط حجم الكريات (MCV fl) ومتوسط الهيموغلوبين في الكريه (MCH pg)، ومتوسط تركيز الهيموغلوبين في الكريه ، والانحراف المعياري لمقياس كرات الدم الحمراء ,(RDW-SD fl) ,(RDW-CV %)اللمفاويات ,($x10^9/L$) (LYM) الحد الأقصى للنزيف غير الناضج (MID %),المحببة ,($x10^9/L$) (GRAN)بلاتكريت ,(PCT %)عرض توزيع الصفائح الدموية ,(PDW fl)، نسبة الخلايا الكبيرة للصفائح الدموية (PLCR %)والصفائح الدموية خلية ($PLCC\ x10^9/L$)ويستنتج أن ممارسة التمارين الرياضية المعتدلة تسبب بعض التغييرات على مكونات دم (CBC) الخيول وأنها لن تعود إلى القيم قبل ممارسة التمرين وبعد ساعة من الراحة بعد التمرين.

Abbreviations

Abbreviations	Meaning
CBC	Complete blood count
RBCs	Red blood cells
Hb	Hemoglobin
HCT	Hematocrit
PCV	Packed cell volume
MCV	Mean corpuscular volume
MCH	Mean corpuscular hemoglobin
MCHC	Mean corpuscular hemoglobin concentration
RDW-SD	Red cell distribution width stander deviation
RDW-CV	Red cell distribution width conversation
WBCs	White blood cells
LYM	Lymphocytes
GRAN	Granulocytes
MID	Maximum immature distribution
PLT	Platelets
MPV	Mean platelet volume
PCT	Plateletcrit
PDW	Platelet distribution width
P-LCR	Platelet large cell ratio
RDW	Red cell distribution width
Fl	Femito litter
g/dl	Gram per deciliter
SPSS	Statistical Package for Social Science

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Introduction

Horses are the fastest animals among the four main athletic species including humans, camel and greyhounds. Horse racing has been great industry in many countries all over the world. Therefore horses attracted great attention in study of athletic science only next to that humans. The performance of a horse during competition is the result of a combination of many complex interactions. These interactions include the horse age, gender, breed, genetic potential, gait, diet, psychology, strength and neuromascular coordination, as well as the capacity for work or fitness. The first factor that influences athletic performance of horse is its genetic background. It is obvious that the major focus of the horse racing industry was on the animals past, i.e., in examining the breeding history of the horse. As a result, progress in equine exercise science research lags behind that in human by decades. Little attention, if any, has been paid to its future, which should involve the application of scientific knowledge and techniques to the study of equine science and/or horse racing. Training would be one of the critical factors that enable a potent animal to achieve its full performance. In previous work, we initiated the investigation of the blood cell parameters in Sudan. (Juet, 1993-2002). In continuation of this work, we looked a further into the changes parameters during exercise and after an hour of moderate training. This information would be of value for understanding the exercise physiology and training of mixed breed horses in Sudan.

General Objectives:

To evaluate the effects of exercise on complete blood count of horse.

Specific objectives

Effect of exercise on erythrocytes, leukocyte and blood platelets

Chapter 1

Literature Review

1.1.Equines:

1.1.1. Classification of equids:

Prior to the beginning of domestication, in the late Pleistocene, long-term geographic isolation of equid populations occurred.1 This led to the distinct species that exist today. True horses (***Equus caballus***) occupied the Eurasian lowlands north of the great mountain ranges, while the asses occupied the arid zones of Asia. Crosses with asses, zebras and onagers are possible but the hybrids are normally sterile.

Order: perissodactyla (odd-toed ungulates)
Suborder: hippomorpha
Super-family: equoidea
Family: equidae (Rodenhuis, 2004).

1.1.2. Domestication:

French and Spanish cave paintings from around 15 000 years ago, depicting hunting for food and hides, represent the earliest record of human use of horses. While early horses would eventually provide their keepers with unprecedented mobility and power, the herding of horses probably had its origins in the consumption of horseflesh.11 Hunters favor meat, such as that from horses, that has a high glycogen content. As a source of dietary sugar, this facilitates endurance, which is important for members of hunting cultures. (Rodenhuis, 2004)..

1.1.3. Horses in Sudan:

Horses in Africa are divided into five types, in Sudan there is only two types:
1-Westren Sudan horses

2-Aldnqlawiyeh horses
The scientists of animal production think, that the horses in the Sudan can be divided into three types:
1-Westren Sudan horses
2-Aldnqlawiyeh horses
3-Hybrid horses (Mason and mule)
The total equine population in Sudan is 79087, and their distribution around the country is in the table below. (Ministry of animal wealth, center of information, Khartoum Sudan 2016).

1.1.4. Uses of horse:

There are several uses of horses.

a. Ride
b. Truction
c. Agricultural work
d. Racing (Naim, 2010).

1.1.5. Equine population and statics in Sudan:

Table (1.1) The equine population in Sudan is 79087,

States	Number	Percentage (%)
North Kordofan	83749	10.6%
South Kordofan	15802	2.0%
West Kordofan	52936	6.7%
North Darfour state	33974	4.30
South Darfour state	122305	15.48%
East Darfour state	149484	18.92%
Center Darfour state	92440	11.70%
West Darfour state	112982	14.30%
El Gedarif state	11851	1.50%
Kassala state	9481	1.20%
Red sea state	11061	1.40%
Blue Nile state	15802	2.00%
Senar state	11851	1.50%
El Gezira state	26863	3.40%
White Nile state	33974	4.30%
Northren state	1580	0.20%
River nile state	2370	0.30%
Khartoum state	1580	0.20%
Total	**79087**	**100%**

(Ministry of animal wealth, center of information, Khartoum Sudan,2016).

1.1.6. Hemogram:

1.1.6.1. Table (1.2). Normal erythrocytes count in horse and indicies:

Total RBC (×106/μL)	9.0
Diameter of RBC (μm)	5.5
Hct (%)	41.0
Hemoglobin (g/dL)	14.4
Mean corpuscular volume (MCV; fL)	45.5
Mean corpuscular hemoglobin (MCH; pg)	15.9
Mean corpuscular hemoglobin concentration	35.0

(Reece, *et al*, 2015).

1.1.6.2. Table (1.3) Normal luekocyte count in horse and differential count.

WBCs	Total leukocyte count (per μL)	Neutrophil (%)	Lymphocyte (%)	Monocyte (%)	Eosinophil (%)	Basophil (%)
Horse	8000-11,000	50-60	30–40	5–6	2–5	<1

(Reece, *et al*, 2015).

1.1.6.3. Table (1.4) platelet count in horses:

Platelets count in horses (10^{14}/µl)	100,000 - 350,000

(Feldman, *et al* 2000)

1.2.Blood

Blood is a fluid connective tissue that flows throughout the entire body. Whole blood is the blood contained in the cardiovascular system. Peripheral blood is whole blood circulating in blood vessels. (Colvilli and Bassert, 2008).

Blood consists of cells and other cell-like formed elements suspended in fluid called plasma. (Franson, *et al* 2009).

Blood has three main functions, they are transportation (eg. Nutrients, oxygen, carbon dioxide, west products, hormones, heat, and immune bodies), regulation (maintain fluid balance, PH equilibrium in the body) and defense (defense against foreign invaders through the phagocytosis or development of the immunity) (Reece, *et al*, 2015).

1.2.1. Plasma:

Plasma is primarily water in which various solutes are dissolved,

When a blood sample is centrifuged, the heavier formed elements are backed into the bottom of the tube, leaving plasma at the top. The formed elements constitute approximately 45% of the total blood volume while the plasma accounts the remaining 55%.

The color of plasma varies from colorless to straw to yellow–orange depending on the species, diet and any pathologic condition present. Normal plasma is transparent. (Colvilli and Bassert, 2008).

When a sample of blood is treated with an anticoagulant to prevent clotting and permitted to stand in a tube undisturbed, the cells gradually settle to the bottom, leaving a straw colored fluid above. This fluid portion of the blood is plasma. When blood is allowed to clot, the cells are trapped in a meshwork of clotting proteins, leaving a yellow fluid, the serum. Essentially, serum is plasma minus the plasma proteins responsible for producing the clot. (Franson, *et al* 2009)

The blood plasma contains proteins and factors necessary for the integrity of the organism. Some of these factors are involved in coagulation, and others function as transport proteins, or

immuneglobulins. Most of the plasma proteins are synthesized in the liver except of immunoglobulin. The major protein of plasma or serum is albumin. Albumin maintains the colloidal osmotic pressure of the serum and is a transport protein for many hormones, ions, vitamins, and other factors, (Schiller, 2007)

1.2.2. Erythrocytes:

1.2.2.1. Shape, life span and size:

Erythrocytes are generally considered to be discocytes, with is some degree of concavity. Mammalian erythrocytes lack of nuclei and are non motile. Erythrocytes vary in diameter and thickness according to species and nutritional status of the animal. Life span of erythrocytes varies with species, reported value for horses are 140-150 days. (Reece, *et al* 2015). Erythrocytes have main function is transport and carry oxygen from the lung to the tissue, by circulating of it's hemoglobin content (Misra, 2014).

1.2.2.2. Hemoglobin

The principal component of erythrocytes is hemoglobin (Hb), which makes up about one-third of the erythrocyte content, The hemoglobin molecule has a molecular weight of about 67,000 and is composed of four heme groups combined with one molecule of globin (the protein component). Globin is composed of four polypeptide chains, each containing one of the heme groups. Each heme group contains an iron atom that combines loosely and reversibly with one oxygen which facilitate function of oxygen transport. The iron atom of heme has a valence of +2 (Fe^{2+}, ferrous) regardless of whether molecular oxygen is combined with it. (Reece, *et al* 2015).

1.2.2.3.Packed cell volume (PCV):

Is expressed as a percent, volume of packed cells in whole blood after centrifugation (Reece, *et al* 2015).

1.2.2.4. Erythrocyte indices

The erythrocyte indices are determinations that are calculated after the erythrocytes (RBCs) have been enumerated and Hct and Hb concentration determined. There are three indices, and each relates to a value for a single RBC. Accordingly, the units are small and are shown for each as follows.

Derivations of values are as follows (exponent manipulations completed but not included):

Mean corpuscular volume refers to the average volume of the individual red cells and is expressed in cubic micrometer per red cell or femto litters(fl) calculated as follow ;

$MCV = (Hct/RBC) \times 10$

Mean corpuscular hemoglobin is refers to hemoglobin content per red cell and is expressed in picogram per red cell and calculated as follow;

$MCH = ([Hb]/RBC) \times 10$

Mean corpuscular hemoglobin concentration is refer to hemoglobin concentration of the red cell and expressed in gram per deci litter and calculated as follow ;

$MCHC = ([Hb]/Hct) \times 100$. (Reece, *et al*, 2015).

Now these equations are determined with use of an Automatic Blood analeyzer.

1.2.2.5. Factors which effect the erythrocytes count and hemoglobin concentration and other blood constituent:

A. Age.
B. Sex.
C. Exercise
D. Nutritional status
E. Lactation
F. Pregnancy

G. Egg production
H. Excitement
I. Blood volume
J. Environmental temperature
K. Altitude and other clinical factors. (Mishra, 2014).

1.2.3. leukocytes:

Leukocytes are colorless and nucleated elements of blood. They are the mobile unit of the body's defense mechanism (Mishra, 2014).

Leukocytes are classified into two categories granulocytes that have granule into their cytoplasm, named by how this granules stain, they are neutrophil, esonophil (red granule) and basophil (blu granules). Agranulocyte do not have granule in their cytoplasm they are monocyte and lymphocyte. The function of all white blood cells is to provide defense for the body against foreign invaders. Each type of white blood cell has its own unique role in this defense. If all the white blood cells are functioning properly, an animal has a good chance of remaining healthy. (Colvilli and Bassert, 2008).

1.2.3.1. Granulocytes:

1.2.3.1.1. Neutrophils:

Neutrophils are the most common white blood cells found in the peripheral blood of all the common domestic species, except ruminants, neutrophils are typically 10–12 μm in diameter and have single nuclei with several indentations, resulting in the nucleus being divided into multiple lobes. It is present normally in GIT, urinary, respiratory and reproductive tract. Functionally it helps prevent entrance of microorganisms or foreign particles (Reagan, *et al.*, 2008)

It has life span in the blood 6-20h and they are constantly leave in the blood into the tissue for varies period. (Reece, *et al* 2015)

1.2.3.1.2 Eosinophils:

Eosinophils are absent or present in very low numbers in normal animals. These cells are typically similar in size of neutrophils but are often slightly larger. The nuclei are very similar to those of neutrophils in that they are segmented, The cytoplasm stains faint blue and has multiple reddish to reddish-orange granules.. (Reagan, *et al.*, 2008).
After their development, leukocytes are circulated in the blood
until the time (relatively short) they leave the circulation to perform their extra vascular function. (Reece, *et al* 2015)
They are found normally in allergic condition and parasitic infection.

1.2.3.1.3. Basophiles:

In blood are similar to the tissue mast cells located immediately outside many of the capillaries in body. Both basophiles and mast cells secrete heparin that prevents blood coagulation and also histamine (Mishra, 2014).
Basophiles are rarely seen in the peripheral blood of all the common domestic species. They are most commonly seen in horses. Basophiles are similar in size or slightly larger than neutrophils, and the cytoplasm is light purple. The nucleus is segmented but often not to the degree of the mature neutrophil. (Reagan, *et al.*, 2008)
.
Life span as esinophils

1.2.3.2. Agranulocytes:

1.2.3.2.1. Monocystes:

They play an important role in defense of body along with nuetrophills, Monocytes conistitute the first line of defense. Like nuetrophils monocytes are motile and phagocytic (Mishra, 2014).
Monocytes have a circulation time of 24 hours or less, but can remain in the tissues for several months. Many monocytes become fixed macrophages in the sinusoids of the liver, spleen, bone marrow, and

lymph nodes; in this way they continue to function in the blood and lymph. (Reece, *et al* 2015).

1.2.3.2.2. Lymphocytes:

Most of the lymphocytes seen in blood are small lymphocytes that can be recognized by their round or oval nucleus and minimal amount of clear, almost colorless cytoplasm. Small lymphocytes are smaller than neutrophils when viewed on a stained blood smear. Medium and large lymphocytes are sometimes seen in peripheral blood. Most of the lymphocytes in the body actually live in what are called lymphoid tissues and constantly circulate between these tissues and blood. Lymphocytosis and lymphocytopenia refer to an increase and a decrease in the number of circulating lymphocytes, respectively.

T cells are processed in the thymus before going to peripheral lymphoid tissue. T cells are responsible for cell-mediated immunity (no antibody production involved) and for activating B cells. Most of the lymphocytes in peripheral blood are T cells.

B cells. Inactive B cells travel through lymph nodes, the spleen, and other lymphoid structures, but rarely circulate in peripheral blood. B cells are ultimately responsible for humoral immunity (antibody production is involved), (Colvilli and Bassert, 2008).
Their lifespan varies, depending on classification. Generally T cells are long-lived (100–200 days), B cells are short-lived (2–4 days), and memory T and B cells are very long-lived (years) (Reece, *et al* 2015).

1.2.4. Platelets:

Blood platelets, also called thrombocytes, are fragments of megakaryocytes, large cells formed and residing in the bone marrow. Thrombocytes are the smallest of the formed elements in the blood at 2–

4 μm. They are surrounded by a plasma membrane and contain some organelles, but not nuclei. (Frandson, *et al* 2009).
On a blood smear, platelets appear non-nucleated, round to oval in shape with clear cytoplasm that contains small blue to purple granules. The granules contain clotting factors and calcium, which are necessary for blood to clot. The appearance of platelets in a stained smear may be considerably different from their actual appearance in circulating blood, where they are oval disks. In smears they may appear as circular disks, star-shaped fragments, or clumps of irregular shape.
Platelets have many functions in the body, but they are most important for normal hemostasis. Hemostasis is the process by which blood is prevented from leaking out of damaged blood vessels. Platelets have specific roles in the clotting process, along with endothelial cells in the blood vessel wall and coagulation factors. The two specific functions of platelets in hemostasis are the formation of a platelet plug and stabilization of the plug, making it irreversible. (Colvilli and Bassert, 2008).

1.2.4.1. Platelets Indices:

Platelet indices are biomarkers of platelet activation. They allow extensive clinical investigations focusing on the diagnostic and prognostic values in a variety of settings without bringing extra costs. Among these platelet indices, plateletcrit (PCT), mean platelet volume (MPV), and platelet distribution width (PDW) are group of platelet parameters determined together in automatic CBC profile; they are related the platelets morphology and prolifration kinetics.
MPV is the mode of the measured platelet volume. Increased MPV indicates increased platelet diameter which can be used as a marker of production rate and platelet activation.
PDW is an indicator of volume variability in platelets size and the physiological conditions, there is direct relationship between MPV and PDW; both usually change in the same direction.
PCT is the volume occupied by platelets in the blood as a percentage and calculated according to the formula PCT= platelet count × MPV/10.000 (25-27). Genetic and acquired factors, such as race, age, smoking status,

alcohol consumption and physical activity, modify blood platelet counted MPV.

Platelet larger cell ratio (P-LCR) is an indicator of circulating larger platelets (> 12 fl), which is presented as percentage (Zhang, *et al* 2015).

1.2.4.2. Platelet count:

Thrombocytes range from 150,000 to 500,000 per micro liter of blood in most mammalian species. (Frandson, *et al* 2009).

Platelets count in horses: 100,000 to 350,000 (Feldman, *et al* 2000)

Platelets circulate in the blood for approximately 5 to 7 days. The liver produces thrombopoietin, which regulates the number of platelets circulating in the body. Much like erythrocytes, platelets are removed from the circulation by macrophages because of old age or damage. (Colvilli and Bassert, 2008).

1.3.Effect of exercise on blood count:

Regular physical exercise or exercise training produce a coordinated pattern of structural and functional adaptation within the cardiovascular and muscular system. (Hinchcliff, 2014). Plasma viscosity and fibrinogen levels are normally unaffected by training. Limits are imposed on muscular performance by the capacity to deliver oxygen and metabolic substrates to the working muscles and the efficiency of removal of waste products from the muscles. Blood is the pathway by which oxygen and substrates are supplied to the musculature and by which waste products, including heat, are removed. When an animal exercises, the changes observed in circulating blood are remarkably rapid. Most notable is a sharp increase in the unit volume of erythrocytes, leucocytes , and platelets. . (Reece, *et al,* 2015).

1.3.1. Effect of exercise on erythrocytes:

The potential of the spleen to increase the circulating red cell volume is impressive in both the dog and horse. At rest, about one-third to half of the erythrocytes are stored in the spleen. The increase in hematocrit is a

function of exercise intensity; a linear relationship between hematocrit and speed exists up to a hematocrit of approximately 60–70%. This auto transfusion of erythrocytes during exercise boosts the oxygen-carrying capacity of the blood and is thought to be a significant factor contributing to the very high, maximal, oxygen consumption of the horse and dog compared with other species. Total blood volume therefore increases dramatically during exercise, as a result of the contribution of the splenic reservoir (Reece, *et al,* 2015).

Physical training induces adaptations to increased metabolic demands in several respects. One limiting factor for fitness and endurance is the oxygen transport capacity of the blood. This capacity is enhanced during training by an increase in the total volume of red cells. A relationship between state of training, cell volume, and other erythrocyte indices is well established in both human and the horse. When training is prolonged, however, the increase in red cell mass may become excessive. This increase in hematocrit results in reduced racing performance and has been attributed to overtraining. (Reece, *et al,* 2015).

1.3.2. Effect of exercise on leukocytes

Stress and corticosteroid leukogram are characterized by moderate neutrophilia, usually no left shift, lymphopenia, eosinopenia and variable monocyte counts. This response is mediated by glococorticocoids of either endogenous or exogenous origin. Neutrophilia is caused predominantly by early release of nuetrophilis from the bone marrow storage pool and to a lesser extent by decreased margination and immigration from the blood, causing neutrophilis to circulate longer. Sometimes hypersegmented neutrophilis are observed on the blood smear. These cells have five or more nuclear lobes and represent older cells that have remained within the vasculature longer than normally expected. Lymphopenia is caused by temporary sequestration of cells in lymphoid tissue whereas eosinopenia is caused by temporary sequestration of cells in the bone marrow. Lymphocytes and eosinophilis generally are not killed at the concentration of glucocorticoids optained

physiologically or achieved through most therapeutic uses of corticosteroids in horses. This response pattern may be seen in horses secondary to endogenous corticosteroids release in acute and severe diseases, pituitary adenoma, or administration of exogenous corticosteroids. (Feldman, *et al* 2006).

1.3.3. Effect of exercise on platelets:

Platelets counts are increase after exercise and which decrease within exercise. (Reece, *et al* 2015).

Chapter Two

Materials and Methods

2.1. Study design, area and duration:

This is an analytical case control study performed at El khawaja Farm-Khartoum North during the period 5/5 017-22/5/2017

2.2. Animals housing and feed:

Four healthy non pregnant, non lactating, not on estrus mature mares aged 15-24 years were used in this study. The animals are used for the treatment of disabled children.

Animals were kept individually in stables (3.5 x 3.5)m ,they are fed twice a day with stalks of corn, fresh clover, bean, peanut shell and water is available all the time.

2.3. The exercise program:

The exercise program was 30 minutes moderate running, in an area of 1600 m^2.

2.4. Blood collection:

Blood samples were taken before exercise, immediately post exercise and an hour post exercise using standard aseptic measures and great care was taken to avoid subjecting the animals to pain or stress .Blood was drawn from the jugular vein using plastic disposable syringes and were immediately transferred to capped test tubes containing EDTA as an anti-coagulant for complete blood count (CBC), which was done by an automated blood analyzer

2.5. Statistical analysis:

The effect of exercise on CBC was performed by one way ANOVA Test. It was performed using **S**tatistical Package for **S**ocial **S**cience (SPSS16.0). All reported P values were considered significant at a level of $P < 0.05$. 95% Confidence intervals

Chapter Three

Results

Tables (3.5) showed that the effect of exercise in RBCs, Hb, HCT were ($P \leq 0.05$) significantly raised from (6.44 ± 0.69) to (7.27 ± 1.40, (34.12 ± 4.72) to (38.61 ± 7.68) respectively immediately post exercise compared with the values before exercise, and an hour post exercise these parameters were decreased but still they were significantly different from the values obtained before and immediately post exercise.

In table (3.6) showed that the effect of exercise in WBCs count were ($P \leq 0.05$) significantly rised from, (9.56 ± 1.21) to (10.36 ± 2.09), LYM (6.68 ± 3.63) to (8.26 ± 4.86). Immediately post exercise compared with the values before exercise, and an hour post exercise these parameters were decreased but still they were significantly different from the values obtained before and immediately post exercise

Table (3.7) shows that Platelets count was significantly decreased by exercise from (93.79 ± 31.92) to (75.82 ± 34.49), while platelets indices did not show any variation with the exercise.

No significant effect due to exercise was detected in the other parameters that is MCV, MCH, MCHC, RDW-SD, RDW-CV, MID, GRAN

Table (3.5) Effect of exercise on RBCs count and indices.

Treatment / **Parameters**	Rest M ± SD	Exercise M ± SD	After exercise M ± SD
RBC ($x10^{12}$/L)	6.44 ± 0.69^{b}	7.27 ± 1.40^{a}	6.42 ± 0.63^{c}
Hb g/dl	11.58 ± 1.22^{b}	13.46 ± 1.50^{a}	11.34 ± 1.38^{c}
HCT (%)	34.12 ± 4.72^{b}	38.61 ± 7.68^{a}	34.19 ± 4.98^{c}
MCV (fl)	53.01 ± 3.75	53.29 ± 3.72	53.13 ± 3.82
MCH (pg)	18.10 ± 2.86	19.49 ± 6.73	17.56 ± 0.710
MCHC (g/dl)	34.41 ± 4.88	36.64 ± 11.84	33.39 ± 1.51
RDW-SD (fl)	16.50 ± 0.631	16.67 ± 0.834	16.71 ± 0.839
RDW-CV (%)	29.87 ± 2.81	29.95 ± 2.73	29.90 ± 2.69

Table (3.6) Effect of exercise on total and differential WBCs count

Treatment / **Parameters**	Rest M ± SD	Exercise M ± SD	After exercise M ± SD
WBC (10^9/L)	9.56 ±1.21[b]	10.36 ± 2.09[a]	9.17 ± 1.33[c]
LYM (10^9/L)	6.68 ± 3.63[b]	8.26 ± 4.86[a]	5.83 ± 2.50[c]
MID (10^9/L)	20.87 ± 8.24	22.25 ± 6.17	22.35 ± 7.16
GRAN (10^9/L)	72.45 ± 8.41	69.49 ± 5.33	71.82 ± 6.93

Table (3.7) Effect of exercise on platelets count and indices.

Treatment / **Parameters**	Rest M ± SD	Exercise M ± SD	After exercise M ± SD
PLT ($x10^9$/L)	93.79 ± 31.92 [a]	75.82 ± 34.49 [c]	91.50 ± 29.86 [b]
MPV (fl)	11.86 ± 1.81	12.03 ± 3.28	12.06 ± 1.58
PCT (%)	0.102 ± 0.831	0.191 ± 0.552	0.102 ± 0.031
PDW (fl)	12.84 ± 1.64	13.18 ± 2.08	13.21 ± 1.92
P-LCR (%)	8.83 ± 6.52	6.78 ± 8.80	9.76 ± 6.92
P-LCC ($x10^9$/L)	8.18 ± 7.01	5.32 ± 6.99	9.14 ± 7.74

M – Mean, SD-stander diviation.

a, b, c, Means within the same row followed by different superscripts are significantly different at ($P \leq 0.05$)

Chapter Four

Discussion

In this study there is a significant increase in red blood cells count, hemoglobin concentration, hematocrit, white blood cells and lymphocytes and a significant decrease in platelets count. This accords with (Reece *et al*2015), who reported rapid and remarkable changes in circulating blood of exercising animals. They found the most notable is a sharp increase in the unit volume of erythrocytes, leucocytes, and platelets and they attributed these alterations to spleenic contraction. Blood cells stored in the spleen can be mobilized into the circulation when there is an increased demand to carry a lot of oxygen. The release of stored erythrocytes from the spleen into the systemic circulation is under the influence of the sympathetic nervous system and circulating catecholamines from the adrenal medulla. The smooth muscle capsule of the spleen is innervated by postganglionic sympathetic neurons. Any factor that increases sympathetic nervous activity or plasma catecholamines, such as exercise, will result in splenic contraction and increase the number of circulating erythrocytes. Consequently, exercise, as well as excitation, causes an increase in the circulating erythrocyte volume at an essentially unchanged or reduced plasma volume, resulting in an increase in the packed cell volume or hematocrit, hemoglobin concentration, and red blood cell count.

(Paladino *et al* 2005) reported that the after 30 minute of post exercise rest RBCs, Hb and WBCs value were the same as that recorded at rest ,this contradicts the findings of this work as these values did not return to the rest values an hour post exercise,and they attributed their findings to the good training degree of the study subject . (Hinchcliff *et al* 2014), reported that the response of exercise depend on the intensity and duration of exercise which may explain the discrepsncy in the findings of this study with that of (Paladino *et al*. 2005).

Conclusion

1.Moderate exercise of horses caused :

- An increase in RBCs,Hb concentration ,HCT.and WBCs.
- A reduction in platelets count.

2.An hour post exercise the values were decreased below the normal, and was not considered scientifical difference just numerical

3.The following parmeters were not affected by moderate exercise Mean corpuscular volume (MCV fl), Mean corpuscular hemoglobin (MCH pg), Mean corpuscular hemoglobin concentration (MCHC g/dl), red distribution width standard deviation (RDW-SD %), red distribution width coefficient (RDW-CV fl), maximum immature ditribution(MID %) , granulocyte (GRAN %)

Recommendations

More studies are needed to study the :

1. Effect of exercise on the stress hormones.

2. Effect of exercise on some blood metabolites.

3. Effect of exercise on some clinical parameters.

4. Effect of the season on the physiological responses of the exercising animals.

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