

Comparative assessment the serum values of Iron, phosphorous and cobalt in both healthy and horses with strangles

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ABSTRACT

*Strangles caused by the bacterium *Streptococcus equi*, the signs of equine strangles include fever, nasal discharge and depression, and abscesses can also develop around the head and neck. This study was conducted to evaluate serum values of these elements in horses with strangles and compare with safe animals. Horses were recognized based on clinical and laboratory findings (culturing of the nasal and lymph node discharges). After taking history and recording it, blood samples from jugular vein was obtained and sera were separated. In that time, we also sampled from healthy animals with the same situation. Serum values of cobalt, iron and phosphorous was measured by spectrophotometry and biochemical available kits, respectively. The mean value of iron in the horses with strangles was 104.11 ± 2.21 $\mu\text{g/dl}$ and in control group was 113.10 ± 1.91 $\mu\text{g/dl}$, so, there was significant difference among groups ($P=0.004$). The mean value phosphorous in ill animals insignificantly was lesser than control group ($P=0.113$). Its value was 4.21 ± 0.16 and 4.61 ± 0.19 mg/dl , respectively. The mean value of cobalt in the horses with strangles was 1.36 ± 0.15 $\mu\text{g/dl}$ and in control group was 1.75 ± 0.14 $\mu\text{g/dl}$, so, there was significant difference among groups ($P=0.062$). Finally, in horses with strangles, the decrease in serum values of iron, cobalt and copper showed is not significant. So, considering the role of these elements in hematopoiesis and immune system, the administration of supplements containing these elements (oral or injection) is suggested.*

INTRODUCTION

Strangles is a highly contagious and serious infection of horses and other equids caused by the bacterium, *Streptococcus equi*. The disease is characterized by severe inflammation of the mucosa of the head and throat with extensive swelling and often ruptures of the lymph nodes, which produces large amounts of thick, creamy pus. Strangles is caused by *Streptococcus equi* subspecies *equi*, better known as *Streptococcus equi* (*S. equi*). The organism can be isolated from the nose or lymph nodes of affected animals, and is usually readily identified in the laboratory by simple sugar tests. The disease is worldwide but its importance has been decreased because of decrease in number of horses and progress in the curing methods. Its prevalence has been decreased among military and freighter horses significantly. Nowadays, it may be seen in the small units or as sporadic [7,15]. Horses of all ages are susceptible, though strangles is most common in animals less than 5 years of age and especially in groups of weanling foals or yearlings. Foals under 4 months of age are usually protected by colostrum-derived passive immunity [22]. *S. equi* is main-tained in the horse population by carrier horses but does not survive for more than 6 weeks in the environment. Although the organism is not very robust, the infection is highly contagious.

Transmission is either by direct or indirect contact of susceptible animals with a diseased horse. Direct contact includes contact with a horse that is incubating strangles or has just recovered from the infection, or with an apparently clinically unaffected long-term carrier. Indirect contact occurs when an animal comes in contact with a contaminated stable (buckets, feed, walls, doors) or pasture environment (grass, fences, but almost always the water troughs), or through flies [23]. Susceptible horses develop strangles within 3–14 days of exposure [23]. Animals show typical signs of a generalized infectious process (depression, inappetence, and fever of 39°C–39.5°C. More typically of strangles, horses develop a nasal discharge (initially mucoid, rapidly thickening and purulent), a soft cough and slight but painful swelling between the mandibles, with swelling of the submandibular lymph node. Horses are often seen positioning their heads low and extended, so as to relieve the throat and lymph node pain [12,18,22]. Infection of upper respiratory tract is associated with purulent discharges of nose, and enlargement of sublingual lymph node. The disease may be fall into mistake with viral inflammation of nasal and lungs, viral arteritis, and laryngitis but it must be noted that lymph nodes aren't enlarged like strangles [9,15]. The role of iron, cobalt and phosphorus in hematopoiesis has been known and in this disease animal seems to be insufficiency because of anorexia [13,25]. This study was conducted to evaluate serum values of these elements in horses with strangles and compare with safe animals.

MATERIALS AND METHODS

This study was carried out on 21 suffered horses in stables around Tabriz for one year (2012). Horses were recognized based on clinical and laboratory findings (culturing of the nasal and lymph node discharges). After taking history and recording it, blood samples from jugular vein was obtained and sera were separated. In that time, we also sampled from healthy animals with the same situation. Serum values of cobalt, iron and phosphorous was measured by spectrophotometry and biochemical available kits, respectively.

Statistical analysis:

SPSS used for statistical analysis and for showing the difference between groups we used the T-test and for comparison the data we used the ANOVA and correlation between groups we used the Pearson correlation index.

RESULTS

The mean value of iron in the horses with strangles was 104.11 ± 2.21 $\mu\text{g/dl}$ and in control group was 113.10 ± 1.91 $\mu\text{g/dl}$, so, there was significant difference among groups ($P=0.004$) see the table and diagram 1.

In the diseased group, there was no significant difference among different age groups in term of serum values of elements.

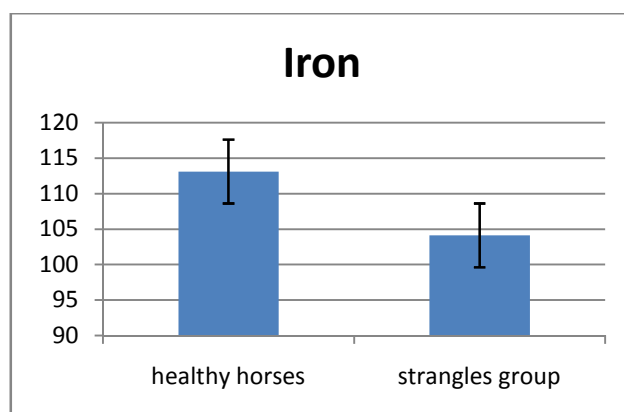


Diagram 1: serum values of iron in animals of both groups

The mean value phosphorous in ill animals insignificantly was lesser than control group ($P=0.113$). Its value was 4.21 ± 0.16 and 4.61 ± 0.19 mg/dl , respectively. See the table 1 and diagram 2.

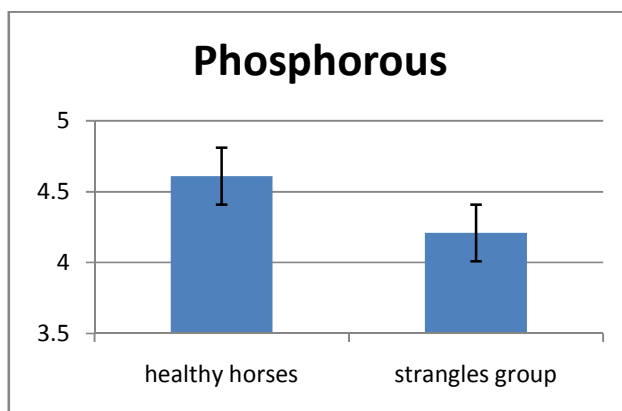


Diagram 2: serum values of phosphorous in animals of both groups

The mean value of cobalt in the horses with strangles was 1.36 ± 0.15 µg/dl and in control group was 1.75 ± 0.14 µg/dl, so, there was significant difference among groups ($P=0.062$) see the table 1 and diagram 3.

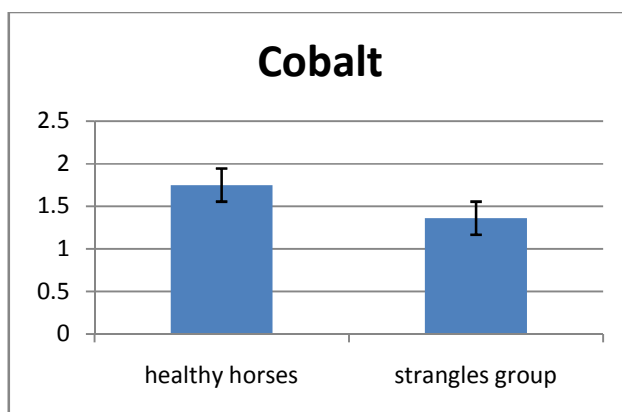


Diagram 3: serum values of cobalt in animals of both groups

Table 1: comparison of mean values of iron, cobalt and phosphorous in both groups

Parameter	Group	No.	Mean	SD	P-value
Iron	Ill	21	104.11	2.21	0.004
	Control	18	113.10	1.91	
Phosphorous	Ill	21	4.21	0.16	0.113
	Control	18	4.61	0.19	
Cobalt	Ill	21	1.36	0.15	0.062
	Control	18	1.75	0.14	

Ill horses were in 3 age groups including 1-3 (8 horses), 4-6 (9 horses) and more than 6 (4 horses) year-old. The mean value of iron, phosphorous and cobalt in these age groups were 102.53 ± 4.49 , 104.44 ± 3.01 and 106.51 ± 4.40 µg/dl in term of iron, 4.33 ± 0.027 , 4.25 ± 0.26 and 3.91 ± 0.022 mg/dl in term of phosphorous and 1.44 ± 0.29 , 1.35 ± 0.21 and 1.36 ± 0.15 µg/dl in term of cobalt. Statistical analysis showed no significance difference among them ($P=0.823$, 0.641 , 0.122 respectively). See table 2 and diagrams 4,5,6.

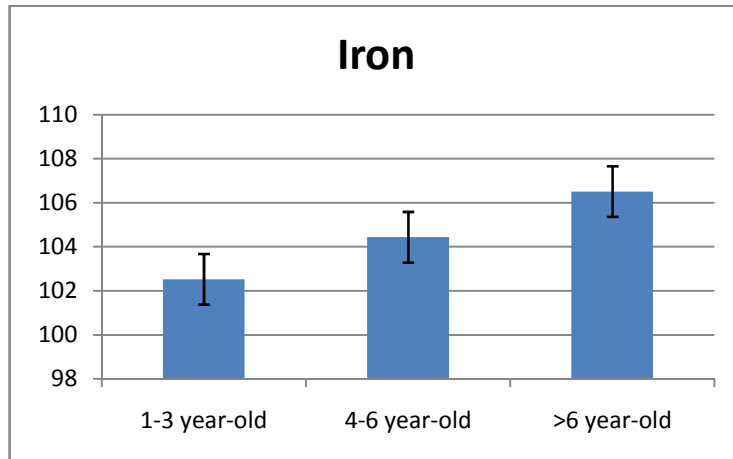


Diagram 4: serum values of iron in ill animals in term of age group

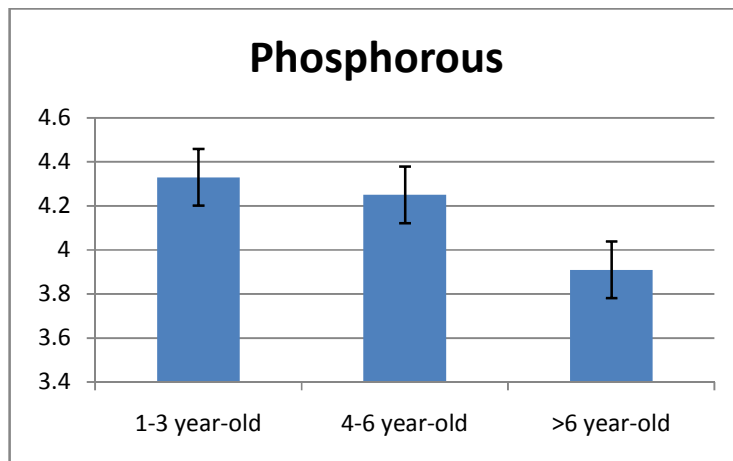


Diagram 5: serum values of phosphorous in ill animals in term of age group

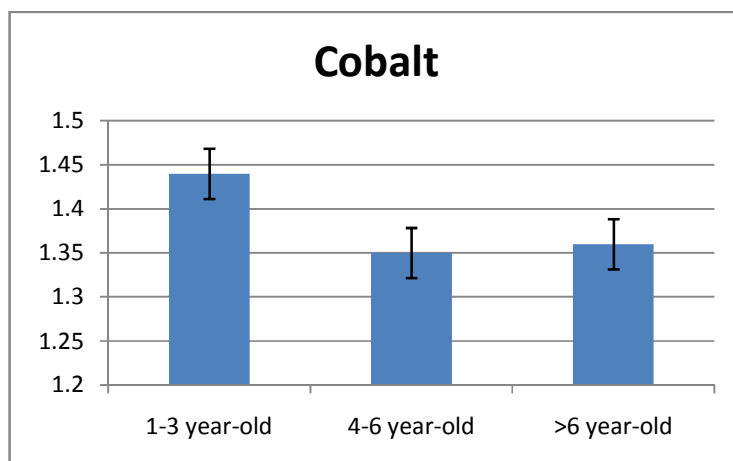


Diagram 6: serum values of cobalt in ill animals in term of age group

Table 2: comparison of mean values of iron, cobalt and phosphorous in ill horses in term of age group

Parameter	Group	No.	Mean	SD	P-value
Iron	1-3 year-old	8	102.53	4.49	0.823
	4-6 year-old	9	104.44	3.01	
	>6 year-old	4	106.51	4.40	
Phosphorous	1-3 year-old	8	4.33	0.27	0.641
	4-6 year-old	9	4.25	0.26	
	>6 year-old	4	3.91	0.22	
Cobalt	1-3 year-old	8	1.44	0.29	0.122
	4-6 year-old	9	1.35	0.21	
	>6 year-old	4	1.36	0.15	

DISCUSSION AND CONCLUSION

Based on obtained data it revealed that serum values of iron, phosphorous and cobalt decrease. Also, it has been revealed that decrease in iron values was significant but in term of phosphorous and cobalt was not significant. Iron is abundant in biology. Iron-proteins are found in all living organisms, ranging from the evolutionarily primitive archaea to humans. The color of blood is due to the hemoglobin, an iron-containing protein. As illustrated by hemoglobin, iron often is bound to cofactors, e.g. in hemes. The iron-sulfur clusters are pervasive and include nitrogenase, the enzymes responsible for biological nitrogen fixation. Influential theories of evolution have invoked a role for iron sulfides in the iron-sulfur world theory. Structure of Heme b, in the protein additional ligand(s) would be attached to Fe. Iron is a necessary trace element found in nearly all living organisms. Iron-containing enzymes and proteins, often containing heme prosthetic groups, participate in many biological oxidations and in transport. Examples of proteins found in higher organisms include hemoglobin, cytochrome, and catalase [1]. Many of the symptoms of iron deficiency are related to the consequences of poor oxygen and carbon dioxide transport. Anaemia may lead to lack of exercise which, in turn affects fitness and other body systems. Physiological actions of iron include take a part in the cellular oxidative mechanisms, structure of hemoglobin, myoglobin, enzymes such as oxygen-carrying cytochromes and flavoproteins[5].

Inorganic phosphorus in the form of the phosphate PO_3^{-4} is required for all known forms of life, playing a major role in biological molecules such as DNA and RNA where it forms part of the structural framework of these molecules. Living cells also use phosphate to transport cellular energy in the form of adenosine triphosphate (ATP). Nearly every cellular process that uses energy obtains it in the form of ATP. ATP is also important for phosphorylation, a key regulatory event in cells. Phospholipids are the main structural components of all cellular membranes. Calcium phosphate salts assist in stiffening bones.

Living cells are defined by a membrane that separates it from its surroundings. Biological membranes are made from a phospholipid matrix and proteins, typically in the form of a bilayer. Phospholipids are derived from glycerol, such that two of the glycerol hydroxyl (OH) protons have been replaced with fatty acids as an ester, and the third hydroxyl proton has been replaced with phosphate bonded to another alcohol.

An average adult human contains about 0.7 kg of phosphorus, about 85–90% of which is present in bones and teeth in the form of apatite, and the remainder in soft tissues and extracellular fluids (~1%). The phosphorus content increases from about 0.5 weight% in infancy to 0.65–1.1 weight% in adults. Average phosphorus concentration in the blood is about 0.4 g/L, about 70% of that is organic and 30% inorganic phosphates. A well-fed adult in the industrialized world consumes and excretes about 1–3 grams of phosphorus per day, with consumption in the form of inorganic phosphate and phosphorus-containing biomolecules such as nucleic acids and phospholipids; and excretion almost exclusively in the form of phosphate ions such as $\text{H}_2\text{PO}_2^{-4}$ and HPO_2^{-4} . Only about 0.1% of body phosphate circulates in the blood, and this amount reflects the amount of phosphate available to soft tissue cells[20]. Cobalt is essential to all animals. It is a key constituent of cobalamin, also known as vitamin B_{12} , which is the primary biological reservoir of cobalt as an "ultra-trace" element. Bacteria in the guts of ruminant animals convert cobalt salts into vitamin B_{12} , a compound which can only be produced by bacteria or archaea. The minimum presence of cobalt in soils therefore markedly improves the health of grazing animals, and an uptake of 0.20 mg/kg a day is recommended for them, as they can obtain vitamin B_{12} in no other way. In the early 20th century during the development for farming of the North Island Volcanic Plateau of New Zealand, cattle suffered from what was termed "bush sickness". It was discovered that the volcanic soils lacked cobalt salts, which was necessary for cattle. The ailment was cured by adding small amounts of cobalt to fertilizers. Non-ruminant herbivores produce vitamin

B₁₂ from bacteria in their colons which again make the vitamin from simple cobalt salts[3,6]. However the vitamin cannot be absorbed from the colon, and thus non-ruminants must ingest feces to obtain the nutrient. Animals that do not follow these methods of getting vitamin B₁₂ from their own gastrointestinal bacteria or that of other animals, must obtain the vitamin pre-made in other animal products in their diet, and they cannot benefit from ingesting simple cobalt salts[21].

It is important to take a balanced and holistic approach to treating micronutrient deficiencies in practical farm situations. It may be that providing multiple routes to solving a problem is appropriate, such as using fertilizers or feed blocks during the grazing season and feed supplementation over winter, with boluses used to 'top-up' the most at risk (highly productive) animals. However, the farmer must be assured that all the deficiencies are being addressed in the most efficient and cost effective manner[2]. In one study on horses infected with equine herpes virus type I, the mean value of zinc and copper decreased but iron increased. Also, cobalt had not significant difference between normal and ill horses [25]. The mean value of copper in patients with hepatic fibrosis has been reported is more than normal ones [8,19]. Also, decrease in serum value of zinc has been reported in some cases with viral hepatitis [4,14] and AIDS [11,24]. In cattle with pneumonia, decrease in zinc and copper has been reported [16]. The results of our study are compatible with above mentioned studies. In one study, feeding pregnant mares with supplements containing copper, zinc, iron, manganese, cobalt, iodine and selenium resulted in increase the serum values of these elements in circulation and milk. They also showed that neonates of these horses also have high levels of these elements [10]. In one research in Brazil on bufflehead with infectious disease, the serum values of copper and zinc were significantly more than normal ones. Also, the serum values of cobalt and phosphorous were not changed significantly [17]. Finally, in horses with strangles, the decrease in serum values of iron, cobalt and copper showed is not significant. So, considering the role of these elements in hematopoiesis and immune system, the administration of supplements containing these elements (oral or injection) is suggested.

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