

Correlation of F2 Isoprostanes and antioxidants of serum in the horses with strangles

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Abstract: This study was conducted on horses with strangles for evaluation serum concentration of F2 Isoprostanes (F2IP) and antioxidants (Glutathione Peroxidase (GPx), Superoxide Dismutase (SOD), Catalase (CAT), Total Antioxidant Capacity (TAC), Vitamin E (Vit E), Vitamin C (Vit C) and Selenium (Se)). 10 horses confirmed on the bases of clinical and laboratory signs of strangles. Blood samples tacked from jugular vein and separated serum. Sampling performed from 8 healthy horses with similar ages, feed and condition, too. Serum concentrations of HB, F2IP, Vit E, Vit C, Se, TAC and serum's activity of GPx, SOD and CAT enzymes measured in the samples. There was a negative correlation between F2IP and serum's antioxidants in two groups, which antioxidants decreased in the serum with increasing of F2IP. The correlation between MDA with GPx, CAT, TAC, Vit E, Se and HB was significant in the patient group ($p < 0.05$) and was not significant with SOD and Vit C. In the healthy group these correlations were significant in GPx, SOD, CAT and TAC ($p < 0.05$) but no significant in Vit E, Vit C, Se and HB. In conclusion, there was an oxidative stress in the horses with strangles and oxidants use in compare antioxidant markers in the serum and this result must be observe in treatment them.

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1. Introduction

Upper respiratory tract infection with purulent discharge from the nose and throat, enlarged lymph node are main sign of Strangles's disease. In the early stages of the disease there should be mistaken with nose and lungs viral inflammation, viral arteritis and bronchitis but in these disease lymph node size is not as big as Strangles (Timoney et al. 2008; Weese et al. 2009). Some changes may be creating in the serum profiles in strangles. Some changes may be creating in the serum profiles in strangles and Evaluation of these changes will help in preventing and controlling the disease. Evaluation of oxidant-antioxidant balance is important in this disease. F2Isoprostanes (F2IP) is an indicator of oxidative stress as oxidant marker and and serum antioxidants include: Hemoglobin (HB), Glutathione Peroxidase (GPx), Superoxide Dismutase (SOD), Catalase (CAT), Total Antioxidant Capacity (TAC), Vitamin E (Vit E), Vitamin C (Vit C) and Selenium (Se) (Abd Ellah et al. 2009; Alberta 2010; Antonic-Svetina et al. 2010; Clarkson et al. 2000; Konda et al. 1998). F2Isoprostanes as a marker is a derivatives of arachidonic acid, which is derived from peroxidation of lipid and is structurally similar to F_{2α} prostaglandin, so why named F2-Isoprostane (Meydani 1997; Montuschi; et al. 2004). With measuring these serum metabolites, produced

antioxidant levels in the stress disease could be predicted.

This study was conducted on horses with strangles for evaluation of correlation between serum concentration of F2Isoprostanes (F2IP) as an oxidative marker and serum antioxidants (Glutathione Peroxidase (GPx), Superoxide Dismutase (SOD), Catalase (CAT), Total Antioxidant Capacity (TAC), Vitamin E (Vit E), Vitamin C (Vit C), Selenium (se)) and Hemoglobin (HB).

2. Materials and Methods

This study was conducted on 10 horses that suffering from strangles in Tabriz area in Iran. Patients confirmed on the bases of clinical and laboratory signs of strangles (culturing of nasal discharge and sampling from lymph nodes for confirmation of *Streptococcus equi*). Blood samples tacked from jugular vein and separated serum. Sampling performed from 8 healthy horses, too. Serum concentrations of HB measured by Ziest chemia kit, serum's F2IP with ELISA with Cayman chemical Company kit, serum's activity of GPx, SOD enzymes and TAC by Randox kit, serum's activity of CAT enzyme by Aebi biochemical method, Se by atomic absorption, Vit C by Lorry biochemical method, and Vit E by gas chromatography (Chiaradia et al. 1998). SPSS13 statistical package used for analysis of data. To comparison means between

groups T-test used. For relation between serum parameters correlation was corrected. The level of statistical significance was set at $P < 0.05$.

3. Results

The means of F2IP and HB were high in patient group than healthy group significantly ($p=0.001$ and $p=0.003$, respectively). Serum activity of GPx, SOD and CAT enzymes decreased in horses with strangles significantly ($p=0.002$, $p=0.000$ and $p=0.000$, respectively). So TAC, Se, Vit E and Vit C decreased in patient group, the difference of means between two groups was significant without Vit C ($P=0.004$, $P=0.011$, $P=0.005$ and $P=0.635$,

respectively)(table 1). There was a negative correlation between F2IP and serum's antioxidants in two groups, which antioxidants decreased in the serum with increasing of F2IP. The correlation between F2IP with GPx, CAT, TAC, Vit E, Se and HB was significant in the patient group ($p<0.05$) ($r=-0.972$, $r=-0.951$, $r=-0.853$, $r=-0.803$, $r=-0.684$ and $r=0.979$, respectively) and was not significant with SOD and Vit C. In the healthy group these correlations were significant in GPx, SOD, CAT and TAC ($p<0.05$) ($r=-0.764$, $r=-0.856$, $r=-0.903$ and $r=-0.853$ respectively), but was not significant in, Vit E, Vit C, Se and HB.(table 2).

Table 1: Comparison of oxidant and antioxidant means in the serum of horses with strangles and healthy horses

Serum parameters	Group	Number	Mean	SD	P Value
F2IP($\mu\text{mol/l}$)	Patient	10	131.39	7.56	0.001
	Healthy	8	109.27	8.99	
GPx(U/gHB)	Patient	10	45.32	6.14	0.002
	Healthy	8	60.24	3.28	
SOD(U/gHB)	Patient	10	2414.20	78.48	0.000
	Healthy	8	8690.12	492.88	
CAT(U/gHB)	Patient	10	9743.10	583.09	0.000
	Healthy	8	18987.50	789.54	
TAC(mmoll)	Patient	10	0.25	0.04	0.004
	Healthy	8	0.81	0.04	
Vit E($\mu\text{g/ml}$)	Patient	10	2.35	0.16	0.005
	Healthy	8	3.32	0.24	
Se(ng/l)	Patient	10	125.98	1.67	0.011
	Healthy	8	145.38	2.15	
Vit C(mg/dl)	Patient	10	0.28	0.014	0.635
	Healthy	8	0.27	0.015	
HB(g/dl)	Patient	10	20.64	1.57	0.003
	Healthy	8	12.54	2.43	

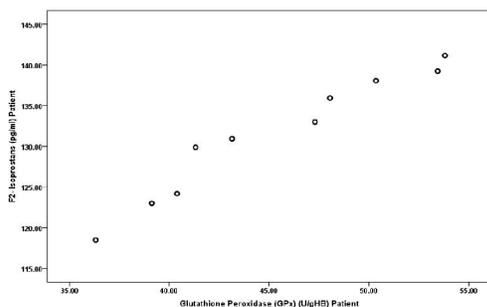
P<0.05 is significant

F2Isoprostanes(F2IP), Glutathione Peroxidase (GPx), Superoxide Dismutase (SOD), Catalase(CAT), Total Antioxidant Capacity (TAC), Vitamin E (Vit E), Vitamin C (Vit C), Selenium (se) and Hemoglobin (HB).

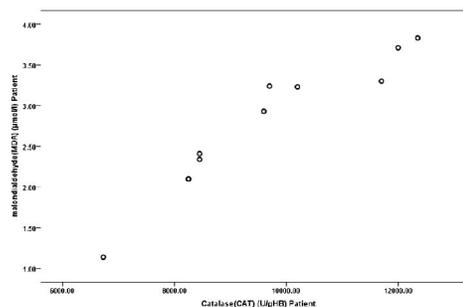
Table 2: Correlation between F2IP and antioxidants of serum in the horses with strangles and healthy horses

		GPx	SOD	CAT	TAC	Vit E	Vit C	Se	HB
Patient group	Correlation(r)	-0.972	-0.487	-0.951	-0.853	-0.803	-0.318	-0.628	0.979
	P Value	0/000	0.057	0.001	0.003	0.004	0.178	0.043	0.000
Healthy group	Correlation(r)	-0.764	-0.856	-0.903	-0.674	-0.505	-0.393	-0.216	0.484
	P Value	0.007	0.005	0.001	0.046	0.101	0.168	0.304	0.112

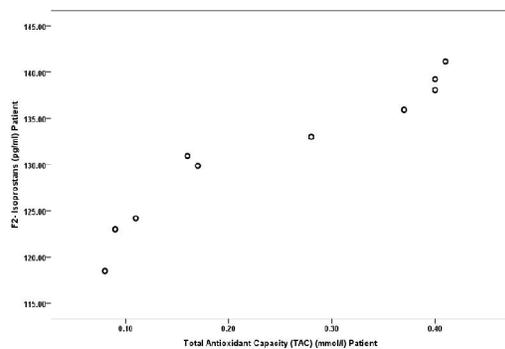
P<0.05 is significant



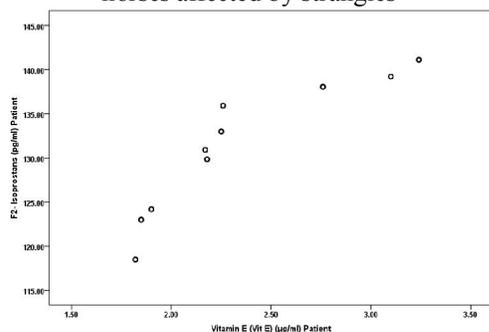
Graph 1: The correlation between F2IP and GPx in horses affected by strangles



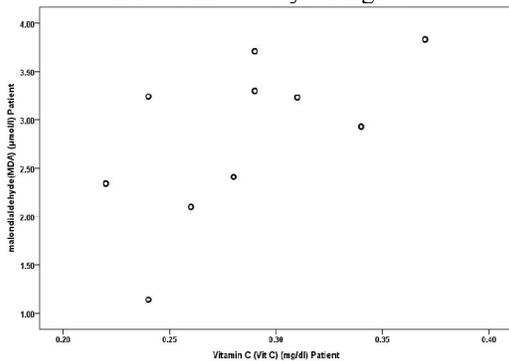
Graph 2: The correlation between F2IP and CAT in horses affected by strangles



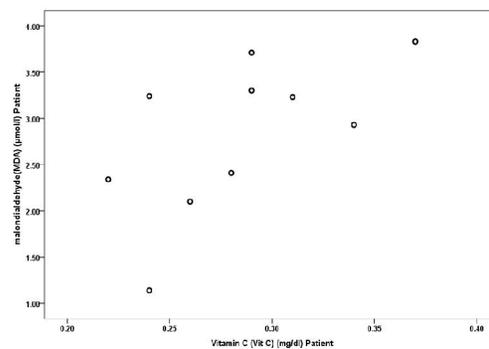
Graph 3: The correlation between F2IP and TAC in horses affected by strangles



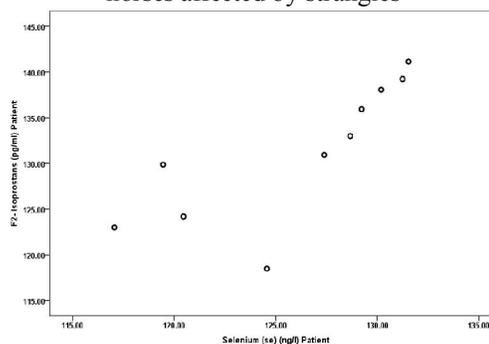
Graph 4: The correlation between F2IP and Vit E in horses affected by strangles



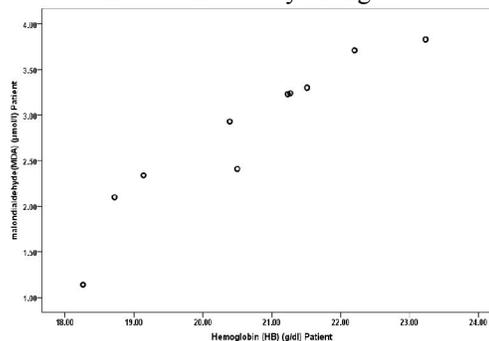
Graph 5: The correlation between F2IP and Vit E in horses affected by strangles



Graph 6: The correlation between F2IP and Vit C in horses affected by strangles



Graph 7: The correlation between F2IP and Se in horses affected by strangles



Graph 8: The correlation between F2IP and HB in horses affected by strangles

4. Discussion

This study defined that serum concentration of F2IP, as an oxidative marker, increase in horses with strangles significantly ($p < 0.05$). Serum activity of GPx, SOD and CAT enzymes and TAC, Se, Vit E and Vit C decreased in patient group, the difference of means between two groups was significant without Vit C ($P < 0.05$). In the patient group the mean of HB was high than healthy group significantly ($p < 0.05$). The correlation between F2IP with GPx, SOD, CAT, TAC, Vit E, Se and HB was significant in the patient group ($p < 0.05$). In the healthy group these correlations

were significant in SOD, CAT, TAC and HB ($p < 0.05$).

Increased levels of hemoglobin can be caused by dehydration, damaging red blood cells and releasing hemoglobin; such results have been obtained in other studies. (Bonilla et al. 2005; Szygula, 1990). Oxidant - antioxidant balance have been studied extensive clinical and laboratory in humans and in horses. (Kirschwink et al. 2008). Other studies have found that exercise causes changes in markers of oxidative (Gomez et al. 2008; Nojima, et al. 2006). Increased serum activity of glutathione peroxidase has been shown after exercise and after infectious diseases

in Standardbred after 12 weeks of exercise and in another study in mice after 8 weeks of exercise. (Demoffarts, et al. 2005; Oztasan et al. 2004). Another study reported decreased activity of the enzyme after exercise (Willians and Carlucci 2006). In a study on 96 horses which infected by Equine Viral Anemia, has been noted that infection causes oxidative stress and significantly changed glutathione peroxidase and superoxide dismutase and malondialdehyde serum activity levels but no significant change in serum catalase (Bolfá et al. 2011). Increasing of F2-Isoprostanes is because of lipid peroxidation that caused by anorexia in strangles. Superoxide dismutase is one of the enzymes which involved as endogenous antioxidant in defense against oxidants and changed free radicals to hydrogen peroxide and Catalase is another enzyme that plays a role in combating free radicals and oxidants and because of its usage in strangle, its decrease is reasonable. (Chen et al. 2009). Decreased serum activities of catalase and superoxide dismutase have been reported in horses with skeletal and neuro-skeletal diseases (Delguste et al. 2007).

In a study on Thoroughbred horses which use selenium and vitamin E supplement for 7 days was found that levels of F2Isoprostanes decreased of 194.5pg / ml to 156.8 pg / ml and serum level of selenium and vitamin E increased (Hartlova et al. 2008). Using supplement with vitamin E has determined that serum level of F2Isoprostanes and Vit E do not change significantly (Pagan et al. 2005).

In a study, the role of nutrition (fruits and vegetables) in smokers and presented its positive role in the reduction of F2Isoprostanes. (Jacab et al., 2003). On a study they have determined that F2Isoprostanes is affected by antioxidant levels and increasing serum level of antioxidant decrease F2Isoprostanes's level (Roberts and Morrow 2002). In a study on Thoroughbred horses which use selenium and vitamin E supplement for 70 days was found that levels of F2Isoprostanes decreased from 194.5pg / ml to 156.8 pg / ml and serum level of selenium and vitamin E increased but there was not any study on Glutathione Peroxidase in this research. (Hartlova et al 2008).

The final result is that there is oxidative stress in horses infected by strangles and with increasing the serum antioxidant and free radicals, antioxidant intake for dealing with them this issue should be considered in treating these patients.

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