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Effects of some Antibiotics and Heat Stress on Glutathione Peroxidase Activities in Albino Male Rats.

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ABSTRACT

The aim of study is to determine the effects of four different antibiotics (Ampicillin, Gentamycin sulphate, Cefodizime and Cefotaxime) by low and high doses on activity of glutathione peroxidase in rats when exposure to heat stress (temperature > 47 °C). Thirty healthy male of Wester Albino rats that divided into five groups. Group 1: Control group given distal water only. Group 2: Injected Ampicillin (low and high doses). Group 3: Injected Gentamycin sulphate (low and high doses). Group 4: Injected Cefodizime (low and high doses). Group 5: Injected Cefotaxime the doses are (low and high doses). When study the effect of four antibiotics on glutathione peroxidase the results shown inhibition of glutathione peroxidase by decrease in the activity of enzyme in low and high doses of ampicillin, gentamycin, cefodizime and cefotaxime treatment when compared with control group recorded significant $p<0.05$, and same significant between high and low doses of cefodizime, but no significant between another antibiotics. Correlation significant show at $p<0.05$ level by ampicillin within doses. In general, the present study indicated that with no treatment successful by these antibiotics with low and high temperature and the heat stress decrease in order to decrease oxidative stress and stay the anti-oxidative enzyme play role factor in the body.

Keywords: Glutathione peroxidase, Ampicillin, Gentamycin sulphate, Cefodizime, Heat stress.

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INTRODUCTION

Glutathione Peroxidase (GPx) is a selenium dependent antioxidant enzyme. It converts H_2O_2 to water, the increasing of production of H_2O_2 due to increased activity of superoxide dismutase (SOD) during heat stress resulted in a coordinate increase in GPx [1]. The determination of GPx activity naturally is high also in value within two days after collection of blood samples stored at (4 °C) and within (15) days under freezing samples [2]. The relation of enzymes activities with temperature, GPx is decreased after the immersion at (42 °C), but no changes at (25 °C). Indicated that heat stress causes oxidative stress in the human body and cold stress is thought to augment the activity of the antioxidative defense system [3]. Oxidative stress results from increased production of free radicals and reactive oxygen species and a decrease in antioxidant defense [4, 5]. This oxidative stress in animals during summer in tropics is heat stress. The external factors such as pollution, sunlight and smoking cause the production of free radicals like environmental extremes (cold, heat, hypoxia, physical exercise or malnutrition) [6]. Under thermal stress vary of physiological and behavioral responses in intensity and duration relation to the animal genetic make-up [7].

Stress is a commonly used term for oxidative stress [8]. It is one of the basic etiologies of disease [6]. Several studies have demonstrated the interdependency of oxidative stress, immune system and inflammation [9]. In through of oxygen stress related to immune system dysfunction seems to senescence in agreement with the oxidation/inflammation theory of aging [10]. Heat stress occurs in animals when there is an imbalance between heat production within the body and its dissipation and may be causes oxidative stress in-vivo. Reactive oxygen species (ROS) the enzymatic antioxidants like superoxide dismutase (SOD), catalase (CAT) and glutathione peroxidase (GPx) act by scavenging both intracellular and extracellular superoxide radical and preventing lipid peroxidation of plasma membrane [11].

These excessive free radicals then interact with other molecules within cells and cause oxidative damage to proteins, membranes and genes or because of many diseases [12, 13], and indicating a microbicidal role of ROS [14]. The multicomponent flavoprotein NADPH oxidase plays vital role in inflammatory processes [15]. The major pathway of pathogenesis for cell damage is via lipid peroxidation particularly in microsomes, mitochondria and endoplasmic reticulum due to oxidative stress (OS) and free radicals [16, 17]. Most of the drugs affect the enzyme systems as inhibitor [18]. Many drugs exhibit the same effects both in vivo and in vitro, while others may not show the same effects on enzymes [19].

Antibiotics on reserve capacities of the energy supply and antioxidant protection systems in newborns, erythrocyte catalase activity is high in children with perinatal asphyxia. Penicillin, aminoglycosides and cephalosporins decreased activity of the glutathione. Ampicillin inhibited ATP synthesis in erythrocytes under conditions of hypoxia and increased GPx activity in response to addition of H_2O_2. Gentamicin is least potent in this respect [20]. The aims of this study is to determine increasing the activity of GPx. in rats blood under heat stress into some antibiotics treated like ampicillin , gentamycin , cefodizime and cefotaxime under different doses.

MATERIALS AND METHODS

Thirty healthy male of Westar Albino rats that aged approximately (2.0 - 2.5 months) in the period from one July to September during summer of Iraq 2015, body weight ranged (190 - 220 g), feed on standard pellet diet and water, in the experiment period were exposure to high temperature ranged between (47- 50 °C) in cages room at the laboratory animals research of Medical College, Diyala University, Iraq. Animal experiments were conducted according to the guidelines of Animal Care and Ethics Committee of the Department of Anatomy and Biology/ College of medicine / Diyala University / Iraq.

They were divided in five main groups of six rats in each and subdivide to groups (3,3) rats with low doses and another is high doses of antibiotics , the four groups are injected I/M with PSs (0.9% Nacl sol.) in first day and injected with doses of antibiotics every 12 hour in days 2,3,4.

Group 1: Control given distal water only.
Group 2: Injected Ampicillin the doses are (low dose 20 mg / kg. B.W., high dose 100mg/ kg B.W.).
Group 3: Injected Gentamycin sulphate the doses are (low dose 2mg / kg.B.W. high dose 20 mg / kg. B.W.)
Group 4: Injected Cefodizime the doses are (low dose 5 mg / kg. B.W., high dose 50mg/ kg. B.W.).

Group 5: Injected Cefotaxime the doses are (low dose 5 mg / kg.B.W. high dose 50 mg /kg.B.W.).

After (9) hours of injection blood samples was taken from the heart by cardiac puncture under light ether anesthesia, whole blood were collected and take place in laboratory procedure and stored in the frozen at (-20 °C) for fifteen day, GPx. levels were determined by knowledge reported method of Puglia and Valentine 1967 (Randox Com.UK., Ransel Kit) by spectrophotometric enzyme tics protocols [23]. The results significantly were used statistical analysis by SPSS.

RESULTS

When study the effect of four antibiotics (Ampicillin, Gentamycin sulphat, Cefodizime and Cefotaxime) on GPx. enzyme from rats blood under high temperature (heat stress condition).(table - 1), show decreased in activity of enzyme by ampicillin and gentamycin sulphat treatment in whole three days of injection recorded (significant $p< 0.05$) when compared low and high doses in (2^{nd}, 3^{rd} and 4^{th}) days with control , but no significant value between low doses and high doses of ampicillin and gentamycin with in various among three days of treatment. The compression of low and high doses in whole days with control by cefodizime and cefotaxime, showed significant at $p< 0.05$, and no significant between low doses and high doses of cefotaxime with in various for three days of treatment, but showed the same significant $p< 0.05$ by cefodizime in low dose 2^{nd} day with high dose 3^{rd} day and between high dose 2^{nd} days with high dose 3^{rd} day.

Correlation /r (table – 2) suggest that significant value are reported at 0.05 (r = 0.010) level in ampicillin between low dose 2^{nd} day with high dose 4^{th} day and between low dose 3^{rd} day with low dose 4^{th} day (r = 0.036) and between high dose 4^{th} day with low dose 2^{nd} day (r = 0.010) , but no significant correlation between control with low and high doses in whole days from four antibiotics treated and no significant correlation between within doses of treatment in whole days by Gentamycin , Cefodizime and Cefotaxime .

DISCUSSION

When study the relation between the activity of GPx in rats blood under heat stress produced by (47 – 50 °C) with low and high doses of antibiotics treated like ampicillin, gentamycin, cefodizime and cefotaxime. Samples stored at (-20 °C) fifteen day , the average value of enzyme activity observation were decreased by this factors of store and frozen , this indeed is good agreement with [2] for the period of storage of samples two days at (42 °C) and within (15) days in frozen.

Moreover the increase of temperature more than (47 °C) at room cages may be cause heat stress in rats and oxidative stress that is well in agreement with [5] as increased of production of free radicals and reactive oxygen species lead to decrease in antioxidant defense and with [3, 11] when the body exposure to hot environment condition, the heat stress occurs in animals when there is an imbalance between heat production in the body and its dissipation. When the rats exposure to heat stress by increasing the optimal temperature more than normal imbalance of the body lead to hypoxia and an addition to treatment of antibiotics like ampicillin, gentamycin, cefodizime and cefotaxime by low and high doses causes inhibited of activity of enzyme, this agreement with [20] in case inhibition by ampicillin and gentamycin. No correlation between antibiotics within doses in three days.

The effect of gentamicin induced nephrotoxicity in albino rats, renal damage was observed, and that suggested the nephron protective effect of Bacopa monnieri which could be by enhancing activity with natural antioxidants and scavenging the free radicals [21]. When studied the effect of eight antibiotics on glutathione reductase (GR) from bovine erythrocytes, showed increased in activity of enzyme by gentamycin, streptomycin, netilmicin, teicoplanin, thiophenicol and ampicillin with increase in their concentration, while the cefotaxime and cefodizime inhibited this inhibition were competitive [22].

Recent studies suggested that some antibiotics could affect superoxide dismutase and GPx activities and reactive oxygen species production. It was reported that gentamicin decreased cardiac SOD and GPx activities and renal GPx activity in guinea pigs [24, 25], tetracycline decreased hepatic SOD activity in rats [26], cephaloridine produced ROS in kidney [27], roxithromycin [28], chloramphenicol [29] and tetracyclines [30] inhibited ROS production in phagocytic cells, bleomycin, a antineoplastic agent, produced ROS in lung and caused pulmonary fibrosis [31].

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Table (1): Comparison between Control with Low and High Doses of Antibiotics in Days

Treat / groups	Dose / day	Mean GPx				p. value < 0.05			
		Ampicillin	Gentamycin	Cefodizime	Cefotaxime	Ampicillin	Gentamycin	Cefodizime	Cefotaxime
Control 1st day	Low dose 2nd	5191.0	6323.0	6617.0	6362.0	*	*	*	*
	Low dose 3rd	5291.6	6448.3	6003.3	5849.0	*	*	*	*
	Low dose 4th	5407.0	6501.0	6107.0	5793.0	*	*	*	*
	High dose 2nd	5625.6	6716.0	6404.3	6483.0	*	*	*	*
	High dose 3rd	5365.0	6452.0	5489.6	6084.0	*	*	*	*
	High dose 4th	5282.3	6312.0	5778.6	5975.0	*	*	*	*
Low dose 2nd day	Control 1st day	5191.0	6323.0	6617.0	6362.0	*	*	*	*
	Low dose 3rd	100.66	125.3	613.6	513.0	N.S	N.S	N.S	N.S
	Low dose 4th	216.00	178.6	510.0	569.0	N.S	N.S	N.S	N.S
	High dose 2nd	434.66	393.0	212.6	121.0	N.S	N.S	N.S	N.S
	High dose 3rd	174.00	129.0	1127.3	278.0	N.S	N.S	*	N.S
	High dose 4th	91.33	10.33	838.3	387.0	N.S	N.S	N.S	N.S
low dose 3rd day	Control 1st day	5291.6	6448.	6003.3	5849.0	*	*	*	*
	Low dose 3rd	100.66	125.3	613.6	513.0	N.S	N.S	N.S	N.S
	Low dose 4th	115.33	53.3	103.6	56.00	N.S	N.S	N.S	N.S
	High dose 2nd	334.00	267.6	401.0	634.3	N.S	N.S	N.S	N.S
	High dose 3rd	73.33	3.66	513.6	235.0	N.S	N.S	N.S	N.S
	High dose 4t	9.33	135.0	224.6	126.0	N.S	N.S	N.S	N.S
Low dose 4th day	Control 1st day	540.7	6501.0	6107.0	5793.0	*	*	*	*
	Low dose 3rd	216.00	178.6	510.0	569.0	N.S	N.S	N.S	N.S
	Low dose 4th	115.33	53.3	103.6	56.00	N.S	N.S	N.S	N.S
	High dose 2nd	218.66	214.3	297.3	690.3	N.S	N.S	N.S	N.S
	High dose 3rd	42.00	49.66	617.3	291.0	N.S	N.S	N.S	N.S
	High dose 4th	124.66	189.0	328.3	182.0	N.S	N.S	N.S	N.S
High dose 2nd day	Control 1st day	5625.0	671.0	6404.0	6483.0	*	*	*	*
	Low dose 3rd	434.66	393.0	212.6	121.3	N.S	N.S	N.S	N.S
	Low dose 4th	334.00	267.6	401.0	634.3	N.S	N.S	N.S	N.S
	High dose 2nd	218.66	214.3	297.3	690.3	N.S	N.S	N.S	N.S
	High dose 3rd	260.66	264.0	914.6	399.3	N.S	N.S	*	N.S
	High dose 4th	343.33	403.3	625.6	508.3	N.S	N.S	N.S	N.S
High dose 3rd day	Control 1st day	5365.0	6452.0	5489.0	6084.0	*	*	*	*
	Low dose 3rd	174.0	129.0	1127.0	278.0	N.S	N.S	N.S	N.S
	Low dose 4th	73.33	3.66	513.6	235.0	N.S	N.S	N.S	N.S
	High dose 2nd	42.00	49.66	617.3	291.0	N.S	N.S	N.S	N.S
	High dose 3rd	260.66	264.0	914.6	399.0	N.S	N.S	N.S	N.S
	High dose 4th	82.66	139.3	289.0	109.0	N.S	N.S	N.S	N.S
High dose 4th day	Control 1st day	5282.3	6312.0	5778.0	5975.0	*	*	*	*
	Low dose 3rd	91.33	10.33	838.3	387.0	N.S	N.S	N.S	N.S
	Low dose 4th	9.33	135.6	224.3	126.0	N.S	N.S	N.S	N.S
	High dose 2nd	124.66	189.0	328.3	182.0	N.S	N.S	N.S	N.S
	High dose 3rd	343.33	403.3	625.6	508.3	N.S	N.S	N.S	N.S
	High dose 4th	82.66	139.3	289.0	109.0	N.S	N.S	NS	N.S

* means significant with p< 0.05

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Table (2): Correlation (r) between Control with Low and High Doses of Antibiotics in Days.

	Control				Low Dose 2nd day				Low Dose 3rd day				Low Dose 4th day				High Dose 2nd day				High Dose 3rd day				High Dose 4th day				P. value			
	Ampicillin	Gentamycin	Cefodizime	Cefotaxime	Ampicillin	Gentamycin	Cefodizime	Cefotaxime	Ampicillin	Gentamycin	Cefodizime	Cefotaxime	Ampicillin	Gentamycin	Cefodizime	Cefotaxime	Ampicillin	Gentamycin	Cefodizime	Cefotaxime	Ampicillin	Gentamycin	Cefodizime	Cefotaxime	Ampicillin	Gentamycin	Cefodizime	Cefotaxime	Ampicillin	Gentamycin	Cefodizime	Cefotaxime
Control	-	-	-	-	0.754	0.487	0.979	0.584	0.918	0.224	0.919	0.924	0.882	0.292	0.237	0.990	0.135	0.676	0.231	0.369	0.807	0.794	0.422	0.674	0.764	0.082	0.846	0.261	N.S	N.S	N.S	N.S
Low Dose 2nd	0.754	0.487	0.979	0.584	-	-	-	-	0.164	0.264	0.102	0.340	0.128	0.196	0.784	0.406	0.889	0.837	0.748	0.215	0.438	0.307	0.599	0.742	0.010*	0.570	0.132	0.845	0.05	N.S	N.S	N.S
Low Dose.3rd	0.918	0.224	0.919	0. 924	0.164	0.264	0.102	0.340	-	-	-	-	0.360	0.068	0.682	0.066	0.947	0.900	0.850	0.555	0.275	0.570	0.497	0.401	0.154	0.306	0.234	0.814	N.S	N.S	N.S	N.S
Low Dose 4th	0.882	0.2922	0.237	0.990	0.128	0.196	0.784	0.406	0.360	0.068	0.682	0.066	-	-	-	-	0.983	0.968	0.467	0.621	0.311	0.502	0.185	0.335	0.118	0.374	0.917	0.749	N.S	N.S	N.S	N.S
High Dose 2nd	0.135	0.676	0.231	0.369	0.889	0.837	0.748	0.215	0.947	0.900	0.850	0.555	0.983	0.968	0.467	0.621	-	-	-	-	0.672	0.530	0.653	0.956	0.899	0.594	0.616	0.631	N.S	N.S	N.S	N.S
High Dose 3rd	0.807	0.794	0.422	0.674	0.439	0.307	0.599	0.742	0.275	0.570	0.497	0.401	0.311	0.502	0.185	0.335	0.672	0.530	0.653	0.956	-	-	-	-	0.429	0.876	0.732	0.413	N.S	N.S	N.S	N.S
High Dose 4th	0.764	0.082	0.846	0.261	0.010 *	0.570	0.132	0.845	0.154	0.306	0.234	0.814	0.118	0.374	0.917	0.749	0.429	0.594	0.616	0.631	0.429	0-876	0.722	0.413	-	-	-	-	0.05	N.S	N.S	N.S

CONCLUSION

The results show decreasing in average value of four antibiotics comparative with control caused by inhibition of activity GPx. Concluded that exposure rats to heat stress caused decrease in activity of enzyme although treated by some antibiotics and no correlation between antibiotics within low and high doses comparative with control by no response to additive of antibiotics or was least potent in this respect. These indeed are well in agreement with no treat successful by these antibiotics in hot environment (high temperature) and must decrease heat stress in order to decrease oxidative stress and stay the anti-oxidative enzyme play role factor in the body.

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REFERENCES

[1] Ganaie AH, Shanker G, Bumla NA, Ghasura RS, Mir NA. Journal Veterinary Science Technology. 2013; 4: 126.
[2] Harapin I, Bedrica LJ, Gračner D, Capak D. Vet. Archive 2008; 78:387-392.
[3] Yoshinori O, Noriyuki Y, Hiroyuki F, Ichiro W, Yuko A. European Journal of Applied Physiology and Occupational Physiology. 1994; 68(1):87-91.
[4] Trevisan M, Browne R, Ram M, Muti P, Freudenheim J. Am J. Epidemiol. 2001; 1(54):48-356.
[5] Williams CA, Kronfeld DS, Hess TM, Saker KE, Waldron JN. J. Anim Sci. 2002; 82: 588-594.
[6] Dhanalakshmi S, Srikumar R, Manikandan S, Parthasarathy NJ, Devi RS. J of Health Science. 2006; 52(6): 843–847.
[7] Altan Ö, Pabuçcuoğlu A, Altan A, Konyalioğlu S, Bayraktar H. British Poultry Science. 2003; 44(4): 545 – 550.

[8] Trachootham D, Lu W, Ogasawara MA, Valle N R, Huang P. Antioxidants and Redox Signaling. 2008; 10(8): 1343–1374.
[9] Valero N, Mosquera J, Añez G, Levy A, Marcucci R, Melchor DM. PLoS ONE. 2013; 8(9) Article ID e73221.
[10] Csillag A, Boldogh I, Pazmandi K. Journal of Immunology. 2010: 184(5): 2377–2385.
[11] Sunil Kumar BV, Kumar A, Kataria M. Journal of Stress Physiology & Biochemistry. 2011;7(1): 45-54.
[12] Anu R, Amit K, Vivek S, Brijesh Y, Ruchi T, Sandip C, Kuldeep D. Pakistan Journal of Biological Sciences. 2014; Article ID 761264, 19 pages.
[13] Rahal A, Ahmad AH, Kumar A. Pakistan Journal of Biological Sciences. 2013; 16(16): 751–758.
[14] Halliwell B, Gutteridge JMC. Oxford University Press, Oxford, UK, 3rd edition. 2007.
[15] Puertollano MA, Puertollano E, de Cienfuegos GÁ, de Pablo MA. Current Topics in Medicinal Chemistry. 2011; 11(14): 1752–1766.
[16] Stehbens WE. Experimental and Molecular Pathology. 2004; 77(2):121–132.
[17] Hodgson PD, Aich P, Stookey J. Veterinary Research. 2012; 4: 21- 26.
[18] Çiftçi M, Türkoğlu V, Aldemir S. Vet. Med. – Czech. 2002; 47:283–288.
[19] Beydemir Ş, Çiftçi M, Özmen İ, Büyükokuroğlu M, Özdemir H, Küfrevioğlu Öİ. Pharmacol. Res. 2000; 42: 188–191.
[20] Sukoyan GV, Mumladze MR, Oboladze ED, Varazanashvili NA. Bull Exp Biol Med. Jun. 2005; 139(6):671- 674.
[21] Ramesh Kannan N, Sudha A, Manimaran A, Saravanan D, Natarajan E. International Journal of Pharmacy and Pharmaceutical Sciences. 2011; 3: 5.
[22] Erat M, Sakirogtu H, Ciftci M. Vet. Med. Czech. 2003: 48(11): 305 - 312.
[23] Paglia DE, Valentine WN. Journal Clinical Medicine. 1967:70: 158 – 167.
[24] Kavutcu M, Canbolat O, Öztürk S, Olcay E, Ulutepe S, Ekinci C, Gökhun IH, Durak I. Nephron Journal. 1996; 72: 269-274.
[25] Ozturk HS, Kavutçu M, Kaçmaz M, Canbolat O, Durak I. Curr. Med. Res. Opin. 1997; 14: 47-52.
[26] Vijayalekshmy KS, Menon VP, Leelamma S. Ind. Journal Biochem. Biophys.1992; 29: 371-374.
[27] Yokozawa T, Owada S. Nefron Journal. 1999; 81: 200-207.
[28] Labro MT, El Benna J, Chavaye CB. Journal Antimicrob. Chemother. 1989; 24: 561-572.
[29] Paape MJ, Miller RH, Ziv G. Journal Dairy Sci. 1990; 73: 1734-1744.
[30] Miyachi Y, Yoshioka A, Imamura S, Niwa Y. Journal Invest. Dermatol. 1986; 9(86): 449-453.
[31] Habib MP, Lackey DL, Lantz RC, Sobonya RE, Grand R, Earnest DL, Bloom JH. Res. Commun. Chem. Pathol. Pharmacol 1993; 81:199-208.