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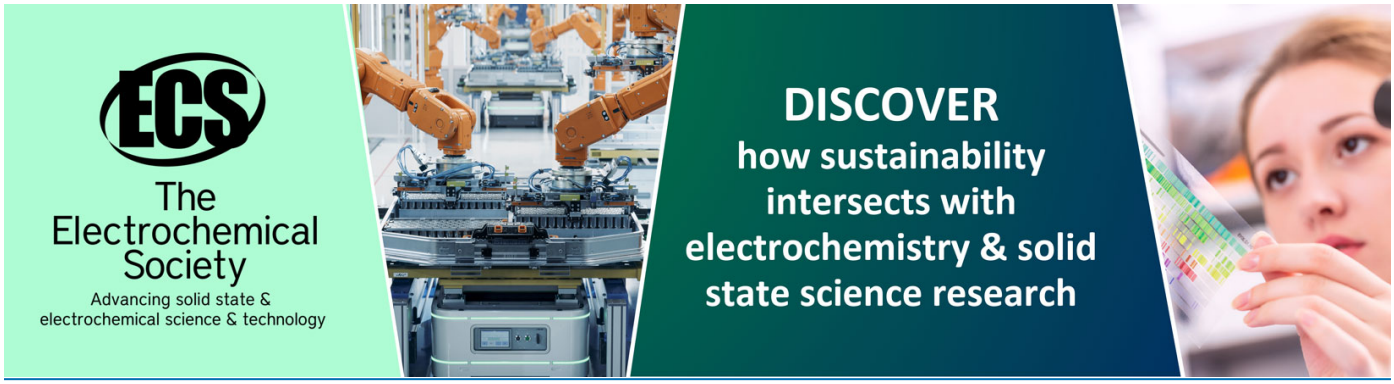

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Role of Nitroglycerine Injection in Japanese Quail *Coturnix japonica* in Some Blood Parameters

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Abstract. This study aimed to evaluate injection of Japanese quail females Countrix japonica with nitro-glycerine (NTG) in blood cells and blood plasma biochemical parameters. In this study 72 females of Japanese quail (45days age) used divided into three groups, three replicates for each one 8 females. Females injected subcutaneous once a week by 0.5 ml of NTG solution by dissolving 500 and 1000 mg of NTG per 100 ml sesame oil for Group1 (G1) and Group2 (G2) respectively while Group0 (G0) left without injection (as a control group). Blood collected from vascular one time every three weeks. Results showed a significant increasing ($P \leq 0.05$) in RBC count mean for G2 group compared with other treatments, while there is no significant differences in other blood cells parameters and no significant differences in blood plasma Aspartate aminotransferase (AST) and Alanine aminotransferase (ALT) concentrations for all groups compared with control.

1.Introduction

Nitroglycerin ($C_3H_7N_3O_9$) is a chemical compound known as glyceryl trinitrate or trinitroglycerine produced by reaction of glycerol with nitric acid (Liu, 2019). Nitroglycerine is soluble in alcohol and high nitrogen content (18.5%) (Jiangfang, 2001). It is possible that NTG is release to the environment from the military explosives and other sources of production might contain NTG causing leather effect to fresh water in vertebrates exposed to 17-18 mg/L of NTG (Lotufo, 2017). Bardai et al (2011) show that inovo injection of NTG make a poisoning effects by increasing mortality of quail embryos and decreased weight gain in the early developmental stage group.In medical community, NTG widely used as a vasodilator for angina treatment and cardiovascular pathologies (Burrows, 1989). NTG Half timework about two minutes inside body (Servent et al, 1989). Oplet et al (2016) show that NTG make activation of soluble Guanylate cyclase (sGC) and cGMP-mediated vascular vasodilator. NTG bio activation is producing high levels of reactive oxygen species (ROS) and reactive nitrogen species (RNS) such as hydroxyl (OH-), super oxide (O2-) (Hanafi et al, 2001) and Nitric oxide (NO) free radicals (Dungel et al. 2011). Nitric oxide in turn modulate molecules mediating communication between cells and signals inside it, helping vascular tone regulation and neurotransmission facilities (Crippen, et al, 2003). Therefore, NO action mediated tone of smooth muscle (Vanhoutte, 2018). NO produced from NTG is highly reaction and short-lived cytotoxic convert to Nitrite and Nitrate that more stable product (Qureshi, 2003; Severina et al, 2003). Causing a mitochondrial dysfunction via NO-independent pathway (Dungel et al. 2011). NO and ROS formatted from NTG make damaging in cell DNA and induce autophagy that protect cells from serious DNA damaging (Wang et al, 2015). Then causing Inhibition in mitochondrial respiration (Brown, 1999). Moreover, inducing ROS production from Mitochondria (Poderoso et al, 1999). Although negative effects

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of NO produced from NTG, but play a cytotoxic effects during inflammatory responses (Crippen, et al, 2003). By binding with bacteria metalloenzymes making a bacterial death by generating reactive compounds during oxidation and produce Nitrite and Nitrate (Lancaster, 1992).This study conducted to evaluate the effect of injection NTG in Japanese quails females as a biological model in blood cells parameters, AST and ALT concentration in blood plasma.

2.Material and methods

This study carried out in quail farm, Animal production department, College of agriculture, (Iraq) according to the protocol approved by the University of Anbar Ethics-Committee to study the effect of Japanese quail female's injection with NTG in blood picture and some biochemical parameters. 72 females of Japanese quail used in this study (45 days age) divided into 3 treatments, 3 replicates for each one (8 females for each replicate). Females injected subcutaneous one time weekly for 6 weeks by 0.5 ml with nitroglycerine (NTG) solution by dissolving 500 and 1000 mg of NTG per 100 ml of sesame oil for treatments (T1 and T2) respectively while (T0) left without injection as a control group. Blood samples were collected from vascular for 2 times (every three weeks) the blood trait studied were Erythrocytes count (RBC), Leucocytes count (WBC) (Natt and herrick, 1952). Packed cell volume (PCV) (Archer, 1965). Differential count for WBC (Shen and Patterson, 1983). Hetrophil to lymphocytes ratio (H/L). Aspartate aminotransferase (AST) and Alanine aminotransferase (ALT) (Reitman and Frankel, 1957).In this study complete random design (CRD) within three treatments, 3 replicates was used. Data analyzed by using GLM model procedure of SAS (Statistical analysis system) (SAS, 2001). Including NTG concentrations. Means for treatments compared by using Duncan's polynomial by using different significance levels to determine significant differences between the averages (Duncan, 1955).

3. Results and Discussion

In table (1) results, show a significant increasing ($P \leq 0.01$) in RBC for 5mg group compared with other groups in first test while, there is significant increasing ($P \leq 0.05$) 5 mg group compared with control group for mean test. There is no significant differences in WBC and PCV for all tests and means.

Table (1) Effect of Nitroglycerine injection to Japanese quail females in blood cells parameters

Test 1			
Nitroglycerine injection/ week	RBC×105	WBC×103	PCV
0 mg	126.27 B	3.83A	31.00 A
2.5 mg	149.33 B	6.16 A	33.00 A
5 mg	202.5 A	3.90 A	34.50 A
SEM	12.37	0.81	1.43
Mean	154.12	4.84	32.62
Significant level	0.01	N. S.	N. S.
Test 2			
Nitroglycerine injection/ week	RBC×105	WBC×103	PCV
0 mg	200.50 A	6.06 A	35.00 A

2.5 mg	178.33 A	6.23 A	30.33 A
5 mg	193.00 A	6.93 A	34.00
SEM	11.89	0.86	1.04
Mean	192.75	6.41	33.11
Significant level	N. S.	N. S.	N.S.
Mean			
Nitroglycerine injection/ week	RBC×105	WBC×103	PCV
0 mg	114.50. B	3.28 A	27.16 A
2.5 mg	164.33 AB	6.92 A	32.33 A
5 mg	200.50 A	5.94 A	34.00 A
SEM	16.36	0.75	2.44
Mean	165.43	5.38	31.16
Significant level	0.05	N. S.	N. S.

Letters refer to differences between rows
SEM: standard error of means
N. S.: No significant differences between columns

Results in table (2) showed no significant differences between treatments in percentage of Lymphocytes, Monocytes, Heterophil, Eosinophil, Basophil and H/L ratio for all tests and means.

Table (2) Effect of Nitroglycerine injection to Japanese quail females in WBC differentiation percentages

Test 1						
Nitroglycerine injection/ week	Lymphocytes %	Monocytes %	Hetrophil %	Eosinophil %	Basophil %	H/L
0 mg	55.27 A	1.68 A	6.48 A	28.92 A	3.75 A	0.12 A
2.5 mg	55.33 A	2.33 A	8.33 A	27.00 A	7.00 A	0.18 A
5 mg	66.80 A	2.51 A	6.03 A	20.11 A	4.53 A	0.09 A
SEM	3.58	0.51	1.36	2.31	0.76	0.38
Mean	58.17	2.13	7.06	26.00	5.16	0.13
Significant level	N. S.	N. S.	N. S.	N. S.	N. S.	N. S.
Test 2						
Nitroglycerine injection/ week	Lymphocytes %	Monocytes %	Hetrophil %	Eosinophil %	Basophil %	H/L
0 mg	61.34 A	1.64 A	9.20 A	18.56 A	4.23 A	0.15 A
2.5 mg	47.51 A	1.99 A	9.28 A	20.56 A	4.98 A	0.19 A
5 mg	53.94 A	0.98 A	13.05 A	26.82 A	5.80 A	0,25 A
SEM	3.54	0.24	1.32	9.76	0.63	0.03
Mean	54.26	1.54	10.51	21.98	5.00	0.20

Significant level	N. S.	N. S.	N. S.	N. S.	N. S.	N. S.
			Mean			
Nitroglycerine injection/ week	**Lymphocytes %**	**Monocytes %**	**Hetrophil %**	**Eosinophil %**	**Basophil %**	**H/L**
0 mg	58.30 A	1.66 A	7.84 A	23.74 A	3.99 A	0.13 A
2.5 mg	51.42 A	2,16 A	8.80 A	23.78 A	5.99 A	0.17 A
5 mg	57.60 A	1.49 A	10.30 A	25.92 A	5.11 A	0.18 A
SEM	2.28	0.28	0.85	2.22	0.43	0.01
Mean	55.55	1.80	8.81	24.30	5.02	0.16
Significant level	N. S.	N. S.	N. S.	N. S.	N. S.	N. S.

SEM: standard error of means
N. S.: No significant differences between rows
In figure (1) and figure (2) results, show no significant differences in blood plasma aspartate aminotransferase (AST) and Alanine aminotransferase (ALT) concentrations between treatments compared with control group in all tests and means.

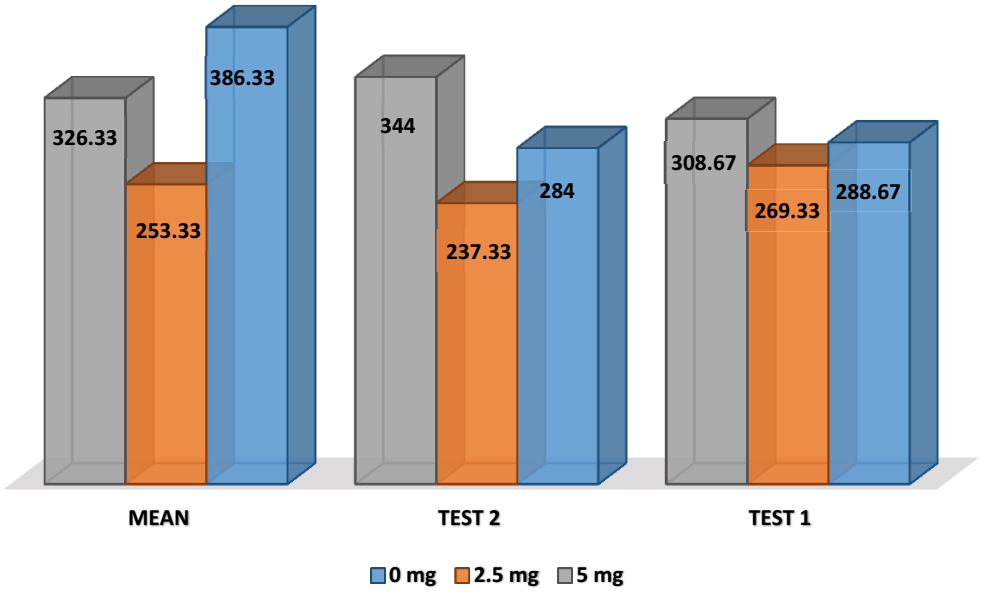

Figure (1) Effect of Nitroglycerine injection to Japanese quail females in blood aspartate aminotransferase (AST) concentration

The significant increasing in RBC may be due to the role of RBC as a powerful scavenger of endothelial cells derived NO (Khun, et al, 2017). RBC make to precipitate in NO metabolism mainly by limiting NO bioavailability. This explain the increasing in RBC as a response to increasing NO concentration produced from NTG in blood to limiting NO bioavailability in order to prevent oxidation in endothelial cells and other cells in body. NO make increasing in ROS formation (Wang et al, 2015 ; Saed et al, 2018). And excessive ROS production led to oxidative stress in cells (Förstermann et al, 2017; Farhan et al, 2020). Reactive ROS and antioxidant defenses system cause an endothelium dysfunction (Incalza et al, 2018). In addition, Hemoglobin make inactivation signals of NO by binding oxygenated Fe++-heme metal center, which followed by subsequently with high reaction constant (Qureshi, 2003).

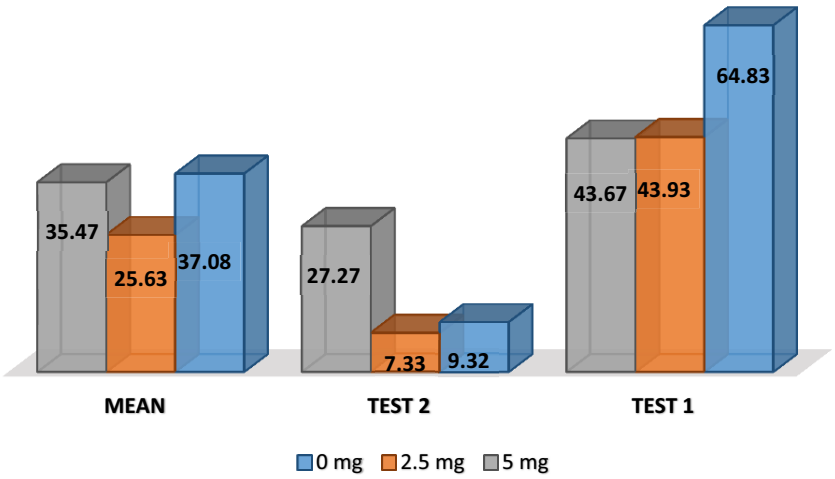

Figure (2) Effect of Nitroglycerine injection to Japanese quail females in blood Alanine aminotransferase (ALT) concentration

Oxidation stressful like NO, ROS and NOS make elevation in liver enzymes activity (Cheraghi et al, 2019). The non-significant differences in blood plasma AST and ALT concentrating mean that body did not impressed by increasing of oxidations stress. It may be as a cause of increasing in RBC count and its rule as an endothelium cells derived NO scavenger. NTG treatment did not affected generation of lymphocytes, Monocytes, Heterophil, Eosinophil and Basophil percentage. These results agree with (Shoker et al, 1997 ; Al-Bayar et al, 2020) who show that human nitric oxide did not affected Lymphocyte generation. In addition, H/L is a hematological indicator that support information about chronic stress in poultry (Siegel, 1995). In present study, results refers to no significant differences in H/L ratio between NTG treatments and control group that explain lack of stress resulted from treating with NTG or increasing in blood NO percentage.

4. Conclusion

NTG increase RBC significantly and did not effect on lymphocytes, Monocytes, Heterophil, Eosinophil, Basophil percentages, plasma AST and ALT concentration. In addition, chronic treatment with NTG did not induced chronic stress in Japanese quail.

References

[1] Jianfang, Hu. 2001. Free Radical and Radiation Biology. ProgramB-180 Med Labs The University of Iowa Iowa City, IA 1166-1181.

[2] Al-Bayar, M. A., Abdulateef, S. M., Farhan, S. M., Shawkat, S. S. & T. Th. Mohammed, Th. T. 2020. Role of Nitroglycerine injection in Japanese Quail (Coturnix japonica) testes tissues parameters. *Indian Journal of Ecology*, **47** (10): 251-255.

[3] Servent, D., Delaforge, M., Ducrocq, C., Mansuy, D., and Lenfant, M. 1989. Nitric oxide formation during microsomal hepatic denitration of glyceryl trinitrate: involvement of cytochrome P-450. *Biochemical and Biophysical Research Communications*, **163**(3), 1210-1216.

[4] Opelt, M., Eroglu, E., Waldeck-Weiermair, M., Russwurm, M., Koesling, D., Malli, R., ... & Mayer, B. 2016. Formation of nitric oxide by aldehyde dehydrogenase-2 is necessary and sufficient for vascular bioactivation of nitroglycerin. *Journal of Biological Chemistry*, **291**(46),

24076-24084.
[5] Farhan, S. M., Abdulateef, S. M. Al-Enzy, A. F. M, Mohammed, Th. T., Saeid, Z. J. M., Al-Khalani, F. M. H. & Abdulateef, F. M. 2020. Effect of heat stress on blood alkalinity of broiler chicks and its reflection in improving the productive performance. *Indian Journal of Ecology*, **47**: 107-109.
[6] Dungel, P., Haindl, S., Behling, T., Mayer, B., Redl, H., & Kozlov, A. V. 2011. Neither nitrite nor nitric oxide mediate toxic effects of nitroglycerin on mitochondria. *Journal of Biochemical and Molecular Toxicology*, **25**(5), 297-302.
[7] Crippen, T. L., Sheffield, C. L., He, H., Lowry, V. K., & Kogut, M. H. 2003. Differential nitric oxide production by chicken immune cells. *Developmental and Comparative Immunology*, **27**(6-7), 603-610.
[8] Saed, Z. J. M., Abdulateef, S. M., Mohammed, Th.T. & Al-Khalani, F. M. H. 2018. Effect of dried tomato pomace as alternative to vitamin C supplemented diets in hematological indices and oxidative stability of egg yolk of laying hens in high-ambient temperature. *Biochemical and Cellular Archives*, **18**(2): 1647-1652
[9] Vanhoutte, P. M. 2018. Nitric oxide: from good to bad. Annals of Vascular Diseases, ra-17.
[10] Qureshi, M. A. 2003. Avian macrophage and immune response: an overview. *Poultry Science*, **82**(5), 691-698.
[11] Severina, I. S., Bussygina, O. G., Pyatakova, N. V., Malenkova, I. V., and Vanin, A. F. 2003. Activation of soluble guanylate cyclase by NO donors—S-nitrosothiols, and dinitrosyl-iron complexes with thiol-containing ligands. *Nitric Oxide*, **8**(3), 155-163.
[12] Wang, X., Zhang, J., Fu, J., Wang, J., Ye, S., Liu, W., and Shao, C. 2015. Role of ROS-mediated autophagy in radiation-induced bystander effect of hepatoma cells. *International Journal of Radiation Biology*, **91**(5), 452-458.
[13] Brown, G. C. 1999. Nitric oxide and mitochondrial respiration. *Biochimica et Biophysica Acta (BBA)-Bioenergetics*, **1411**(2-3), 351-369.
[14] Poderoso, J. J., Carreras, M. C., Lisdero, C., Riobó, N., Schöpfer, F., and Boveris, A. 1996. Nitric oxide inhibits electron transfer and increases superoxide radical production in rat heart mitochondria and submitochondrial particles. *Archives of Biochemistry and Biophysics*, **328**(1), 85-92.
[15] Lancaster J. Nitric oxide in cells. Am Scientist 1992;80: 248–59.
[16] Natt, M. and C. Herrick. 1952. A new blood diluents for counting the erythrocytes and leukocytes of the chicken. *Poult. Sci.* **31**: 735-738.
[17] Archer, R. K. 1965. Haematological techniques for use on animals. Haematological techniques for use on animals.
[18] SHEN, P. F., and PATTERSON, L. T. 1983. A simplified Wright's stain technique for routine avian blood smear staining. *Poultry Science*, **62**(5), 923-924.
[19] Reitman, S. and S.Frankel . 1957. A colorimetic method for the determination of serum glutamic oxaloacetic and glutamic pyrovic transaminases. *Am. J. Clin. Path*. **28**:56-63.
[20] SAS, Institute. 2001. SAS User's Guide: Statistics Version 6.12 edn., SAS Institute, Inc., Cary, NC. USA.
[21] Duncan, D. B. 1955. Multiple range and multiple F tests. *Biometrics*, **11**(1), 1-42.
[22] Kuhn, V., Diederich, L., Keller IV, T. S., Kramer, C. M., Lückstädt, W., Panknin, C., ... & Cortese-Krott, M. M. 2017. Red blood cell function and dysfunction: redox regulation, nitric oxide metabolism, anemia. *Antioxidants & Redox Signaling*, **26**(13), 718-742.
[23] Förstermann, U., Xia, N., & Li, H. 2017. Roles of vascular oxidative stress and nitric oxide in the pathogenesis of atherosclerosis. Circulation research, 120(4), 713-735.
[24] Incalza, M. A., D'Oria, R., Natalicchio, A., Perrini, S., Laviola, L., and Giorgino, F. 2018. Oxidative stress and reactive oxygen species in endothelial dysfunction associated with cardiovascular and metabolic diseases. *Vascular Pharmacology*, **100**, 1-19.
[25] Cheraghi, M., Ahmadvand, H., Maleki, A., Babaeenezhad, E., Shakiba, S., and Hassanzadeh, F.

2019. Oxidative Stress Status and Liver Markers in Coronary Heart Disease. *Inflammation*, **13**, 14.

[26] Shoker, A. S., Yang, H., Murabit, M. A., Jamil, H., Al-Ghoul, A., and Okasha, K. 1997. Analysis of the in vitro effect of exogenous nitric oxide on human lymphocytes. *Molecular and Cellular Biochemistry*, **171**(1-2), 75-83.

[27] Siegel HS. 1995. Stress, strains and resistance. *Br Poult Sci.* **36**: 3–22.

[28] Liu, J. 2019. Mononitrate Ester. In Nitrate Esters Chemistry and Technology (pp. 189-213). Springer, Singapore.

[29] Burrows, E. P., Rosenblatt, D. H., Mitchell, W. R., & Parmer, D. L. 1989. Organic Explosives and Related Compounds. Environmental and Health Considerations (No. USABRDL-TR-8901). army biomedical research and development lab fort detrick md.

[30] Bardai GK, Hales BF, Sunhara GI. Developmental toxicity of glyceryl trinitrate in quail embryos. Birth Defects Res A Clin Mol Teratol 2011;91(Part A):230–40.

[31] Lotufo, G. R. 2017. Toxicity and Bioaccumulation of Munitions Constituents in Aquatic and Terrestrial Organisms. In Energetic Materials (pp. 445-479). Springer, Cham.

[32] Hanafy, K. A., Krumenacker, J. S., and Murad, F. 2001. NO, nitrotyrosine, and cyclic GMP in signal transduction. *Med Sci Monit*, **7**(4), 801-19.