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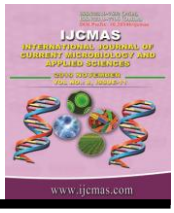

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The Toxic Effects of Paraquat (PQ) on Body Weights and Haematological Parameters in Male Albino Wistar Rats and its Amelioration with Vitamin C

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A B S T R A C T

The present experiment was carried out to investigate the protective effect of Vitamin C on body weights and haematological parameters following paraquat (PQ) intoxication in rats. Total of 48 male albino *Wistar* rats were procured and divided into 4 groups consisting of 12 in each. Group 1- Control. Group 2 - Paraquat (PQ) at the rate of 40 milligram/kg body weight/per oral/day. Group 3 - Vitamin C at the rate of250 milligram/kg body weight/per oral/day. Group 4 - paraquat (PQ) at the rate of 40 milligram/kg body weight/per oral/day + Vitamin C at the rate of 250 milligram/kg body weight/per oral/day. The experiment was carried out for a period of 21 days. Group 4 rats revealed a significant ($P<0.05$) increase in the mean values of body weights, erythrocyte indices, total erythrocyte count (TEC), total leucocyte count (TLC) and Hb concentration except packed cell volume (PCV) which was insignificant when compared with group 2 rats. These results suggested that the Vitamin C administration offered remarkable protection against PQ induced alterations in body weights and haematology.

Introduction

Herbicides or weed killers, are phytotoxic chemicals used for destroying various weeds or inhibiting their growth (Gupta, 2018). These herbicides are being rapidly used in developing countries due to shortage of hand weeding labour and to enhance the crop production (Hossain, 2015). The paraquat (PQ) is one among them which has been used

Worldwide for its high efficiency, low pollution and low residues in crops (Ren *et al.,* 2014).

The PQ (1, 1'- dimethyl- 4,4'- bipyridlium dichloride) is a non-selective nitrogen herbicide for broadleaf weed control (Guo*et al.,* 2015). Globally, it is the second highest selling herbicide with availability at the rate of 20 percent solution form (Banday*et al.,* 2013). It is highly toxic to both humans and animals (Suntres, 2002) potentially leading to Acute Respiratory Distress Syndrome (ARDS) (Huang *et al.,* 2005). The mechanisms of PQ are not fully understood, but it was assumed that the toxicity was due to generation of reactive oxygen species (ROS) through redox-cycling process, resulting in oxidative stress-related damage to cellular organelles, proteins, nucleic acids and lipids (Adam *et al.,* 1990; Bonneh-Barkey *et al.,* 2005 and Castello *et al.,* 2007). Severe PQ toxicity is characterized by multiple organ failure predominantly lungs, kidneys and liver (Tavakol *et al.,* 2015).

The cause of death is respiratory failure resulting from progressive pulmonary fibrosis, because PQ tends to accumulate in clara cells, type I and II pneumocytes through polyamine uptake system (Dinis-oliveira *et al.,* 2009).

The Vitamin C (Ascorbic Acid-AA) is a water soluble vitamin which can directly scavenge the ROS with and without enzyme catalyst, and can indirectly scavenge them by recycling $NADP^+$ to NADPH (Okolonkwo *et al.,* 2014).The fatality rates of PQ toxicity are very high due to lack of effective treatments (Hu *et al.,* 2017).

Hence, there is a need to study the effective antidotes against PQ induced toxicity. The aim of this experiment was to investigate the protective effect of Vitamin C on body weights and hematological parameters after repeated exposure of PQ in *Wistar* rats.

Materials and Methods

In the present study, a total of 48 male albino *Wistar* rats weighing 200-250 grams were procured from Sanzyme Laboratories Ltd, Hyderabad. The rats were housed in solid bottom polypropylene cages at Ruska Labs, Hyderabad and were maintained in controlled environment (20-22^{0}C) throughout the course of experiment. Sterile husk was used as standard bedding material. All the rats were provided with standard pellet diet procured from Vyas Labs, Uppal, Hyderabad and deionized water at *ad libitum* throughout the experimental period.

Rats were randomly divided into 4 groups consisting of 12 in each group. Group 1 served as control whereas group 2 served as PQ toxic control (@ 40 mg/kg b.wt/per oral/day). Group 3 and group 4 rats were administered with Vitamin C (@ 250 mg/kg b.wt/day) and PQ (@40 mg/kg b.wt/day) + Vitamin C (@ 250 mg/kg b.wt/day) respectively.

The experiment was carried out according to the guidelines and prior approval of Institutional Animals Ethics Committee (IAEC-No.02-2019).

Drugs and chemicals

Paraquat (Gramoxone® - 24% w/v solution) was procured from Seed Research and Technology Center, Professor Jayashankar Telangana State Agriculture University, Rajendranagar, Hyderabad which was manufactured by Syngenta India Ltd. Delhi. Ascorbic acid (Vitamin C) as L-Ascorbic acid was obtained from S.D. Fine-Chem Ltd., Mumbai, India.

Growth rate

Individual body weights of all the rats were recorded by using electronic balance on day

zero and subsequently on 7th, 14th and 21st day of experiment to study body weight gains.

Haematology

Prior to blood collection, the selected experimental rats were put to fast for 12 hours. On the day of sacrifice, six (6) rats from each group were used for blood collection (approximately 2-3mL) through retro-orbital plexus with the help of capillary tube into an anticoagulant coated vacutainer {(K_3-EDTA tube, 13mm x 75 mm, 4mL (Rapid Diagnostics Pvt. Ltd., Delhi)} to carry out all haematological parameters. The whole blood was used for estimation of Total Erythrocyte Count (TEC), Total Leukocyte Count (TLC), Haemoglobin (Hb) concentration, Packed Cell Volume (PCV) and erythrocyte indices {Mean Corpuscular Volume (MCV), Mean Corpuscular Haemoglobin (MCH) and Mean Corpuscular Haemoglobin Concentration (MCHC)} by using automatic whole blood analyser (Huma count, med source ozone biomedical Pvt. Ltd., Faridabad, Haryana).

Data obtained (body weights and haematological) was subjected to statistical analysis by applying one-way ANOVA and using statistical package for social sciences (SPSS) version 25.0. Differences between the means were tested by using Duncan's multiple comparison tests and significance level was set at P<0.05 (Snedecor and Cochran, 1994).

Results and Discussion

Weekly body weight gain (g)

Significantly (P<0.05) lower mean values of weekly body weights were recorded in group 2rats(255.00±4.49, 253.00±2.28 and 243.67±2.01) when compared to group 1(285.50±3.60, 318.17±5.02 and 347.00±3.46), group 3 (272.00±3.49, 317.33±4.48 and 333.83±2.60) and group 4 rats(267.00±2.73, 282.83±2.39 and 287.83±5.59) on 7th, 14th and 21st day of experiment (Table 1).

Haematology

The mean values of TEC (millions/µL) in different groups (1, 2, 3 and 4) were ranged from 8.13±0.18 to 9.88±0.10on 7thdayand 7.80±0.33 to 9.77±0.12 on 21st day of experiment. Significantly (P<0.05) decreased values were observed in group 2 rats when compared to groups (1, 3 and 4) on 7th and 21st day of experiment (Table 2).

The mean values of TLC (thousands/µL) in different groups (1, 2, 3 and 4) were ranged from 9.17±0.29 to 13.04±0.42 on 7thday and 8.34±0.21 to 13.78±0.37on 21st day of experiment. Group 2 rats showed a significant (P<0.05) decrease in the TLC values when compared with group 1, group 2 and group 3 rats on 7th and 21st day of the experiment (Table 2).

The Hb concentration (g%) mean values were significantly (P<0.05) reduced in group 2 rats(13.60±0.23 and 12.09±0.16) when compared with group 1(16.60±0.18 and 16.22±0.36), group 3 (17.06±0.45 and 17.10±0.52) and group 4 (15.85±0.15 and 15.57±0.20) (Table 2).

The PCV (%) mean values were insignificantly varied in between the groups on 7th and 21st day of experiment, but there was a numerical elevation in the mean values of group 2 rats when compared with groups 1 and 3 rats on 7th and 21st day of experiment (Table 3).

The MCV mean values were significantly (P<0.05) reduced in group 2 animals (48.76±0.78 and 49.53±0.45) when compared with groups 1 (53.24±0.25 and 52.74±0.68), group 3 (52.95±0.52 and 52.38±0.71) and

groups 4 (51.52±0.45 and 51.33±0.67) on 7th and 21st day of experiment (Table 4).

The mean values of MCH in different groups (1, 2, 3 and 4) were ranged from 14.72±0.52 to 16.83±0.27 on day 7th and 15.71±0.68 to 17.29±0.46 on day 21st of experiment. Significantly (P<0.05) lowered mean values were recorded in group 2 rats when compared to group 1, group 3 and group 4 rats on 7th and 21st day of experiment (Table 4).

The mean values of MCHC were significantly (P<0.05) decreased in group 2 rats (30.86±0.19 and 31.31±0.21) when compared to group 1 (34.22±0.21 and 33.85±0.15), group 3 (33.62±0.10 and 33.79±0.36) and group 4 rats (32.70±0.32 and 33.42±0.41) on 7th and 21st day of experiment (Table 4).

The weight loss could be due to reduced feed and water intake, on the account of toxic action of PQ on GIT and also might be due to free radical induced oxidative damage at sub cellular level in different vital organs. This observation is in accordance with the earlier studies of Dinis-Olievera *et al.,* (2008); Lalruatfela *et al.,* (2014); Haripriya *et al.,* (2017); Hu *et al.,* (2017) and Pourgholamhossein *et al.,* (2018).

These alterations in the mean values of TEC and Hb concentration could be due to haemolysis by the free radical mediated damage to erythrocyte membrane and similar opinion was expressed by Sato *et al.,* (1995). The findings in the present study are in harmony with the authors (Vuksa *et al.,* 1983 and Lalruatfela *et al.,* 2014) previous studies. In the present study, the changes in TLC mean values were similar to the observations of Nagao *et al.,* (1994) who had explained lower TLC values were due to the toxic effect of PQ on leucopoiesis. An insignificant elevated mean values of PCV might be due to haemoconcentration results from fluid loss due to mild diarrhoea and these findings are in accordance with the earlier studies of Wershana (2001).The reduction in mean values of erythrocyte indices (MCV, MCH and MCHC) in the present study might be due to toxic effect of PQ on haemopoietic system and the findings are in accordance with the earlier studies of Lalruatfela *et al.,* (2014)

In group 4, a significant increase in the mean values of weekly body weights, TEC, TLC, Hb concentration and erythrocyte indices except PCV were observed when compared to group 2 which might be due to Vitamin C antioxidant defence action against PQ induced free radical mediated oxidative stress in different tissues including blood cells.

In conclusion, the present study clearly demonstrated that the administration of Vitamin C can effectively attenuate PQ induced alterations of body weights and haematology, possibly *via* antioxidant defence mechanism.

Table.1 Weekly body weight gain (grams) in different groups

GROUP	DAY 7	DAY 14	DAY 21
G 1 (CONTROL)	285.50±3.60^{a}	318.17±5.02^{a}	347.00±3.46^{a}
G 2 (PQ)	255.00±4.49^{c}	253.00±2.28^{c}	243.67±2.01^{d}
G 3 (VITAMIN C)	272.00±3.49^{b}	317.33±4.48^{a}	333.83±2.60^{b}
G 4 (PQ+VITAMIN C)	267.00±2.73^{b}	282.83±2.39^{b}	287.83±5.59^{c}

Values are Mean±SE (n=6); One-way ANOVA
Means with different superscripts in a column differ significantly at P<0.05 (*).

Table.2 Haematological parameters (TEC, TLC and Hb concentration) in different groups

GROUP	TEC(Millions/µL)		TLC(Thousands/ µL)		Hb (g%)	
	DAY 7	DAY 21	DAY 7	DAY 21	DAY 7	DAY 21
G1	9.65±0.15^{a}	9.55±0.26^{a}	13.04±0.42^{a}	13.78±0.37^{a}	16.60±0.18^{ab}	16.22±0.36^{ab}
G 2	8.13±0.18^{c}	7.80±0.33^{b}	9.17±0.29^{c}	8.34±0.21^{c}	13.60±0.23^{c}	12.09±0.16^{c}
G 3	9.88±0.10^{a}	9.77±0.12^{a}	12.76±0.18^{a}	13.24±0.40^{a}	17.06±0.45^{a}	17.10±0.52^{a}
G 4	9.23±0.10^{b}	9.25±0.15^{a}	10.73±0.17^{b}	10.15±0.27^{b}	15.85±0.15^{b}	15.57±0.20^{b}

Values are Mean±SE (n=6); One-way ANOVA
Means with different superscripts in a column differ significantly at P<0.05 (*).

Table.3 Packed cell volume (%) in different groups

GROUP	DAY 7	DAY 21
G 1	52.33±0.20	48.23±1.57
G 2	55.85±2.62	49.92±3.71
G 3	53.43±1.25	48.27±2.53
G 4	52.68±1.60	55.93±1.96

Values are Mean±SE (n=6); One-way ANOVA
Means with different superscripts in a column differ significantly at P<0.05 (*).

Table.4 Erythrocyte indices (MCV, MCH and MCHC) in different groups

GROUP	MCV {Femtoliter (fL)}		MCH {Picogram(pg)}		MCHC {Grams perdecilitre (g/dL)}	
	DAY 7	DAY 21	DAY 7	DAY 21	DAY 7	DAY 21
G1	53.24±0.25^{a}	52.74±0.68^{a}	16.68±0.30^{a}	17.29±0.46^{a}	34.22±0.21^{a}	33.85±0.15^{a}
G 2	48.76±0.78^{c}	49.53±0.45^{b}	14.72±0.52^{b}	15.71±0.68^{b}	30.86±0.19^{c}	31.31±0.21^{b}
G 3	52.95±0.52^{ab}	52.38±0.71^{a}	16.83±0.27^{a}	17.22±0.31^{a}	33.62±0.10^{a}	33.79±0.36^{a}
G 4	51.52±0.45^{b}	51.33±0.67^{ab}	15.98±0.87^{a}	16.35±0.93^{ab}	32.70±0.32^{b}	33.42±0.41^{a}

Values are Mean±SE (n=6); One-way ANOVA
Means with different superscripts in a column differ significantly at P<0.05 (*).

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References

Adam, A., Smith, L. L. and Cohen, G. M. (1990). An assessment of the role of redox cycling in mediating the toxicity of paraquat and nitrofurantoin. *Environmental Health Perspectives*. 85: 113-117.

Banday, T. H., Bashir, S., Bhat, S., Aswin, K., Praveen, and Jagadeesh, S.G. (2013). Manifestation and Management of Paraquat Intoxiction. A deadly poison? *IOSR Journal of Dental and Medical Sciences (IOSR-JDMS)*. 12 (6): 74-76.

Bonneh-Barkay, D., Reaney, S. H., Langston, W. J. and Di Monte, D. A. (2005). Redox cycling of the herbicide

paraquat in microglial cultures. *Molecular Brain Research*. 134(1): 52-56.

Castello, P. R., Drechsel, D. A. and Patel, M. (2007). Mitochondria are a major source of paraquat-induced reactive oxygen species production in the brain. *Journal of Biological Chemistry*. 282(19): 14186-14193.

Dinis-Oliveira, R. J., Duarte, J. A., Sanchez-Navarro, A., Remiao, F., Bastos, M. L. and Carvalho, F. (2008). Paraquat poisonings: mechanisms of lungtoxicity, clinical features, and treatment. *Critical Reviews in Toxicology*. 38 (1)**:**13-71.

Dinis-Oliveira, R. J., de Pinho, P. G., Santos, L., Teixeira, H., Magalhães, T., Santos, A., Bastos, M. L., Remiao, F., Duarte, J. A. and Carvalho, F. (2009). Postmortem analyses unveil the poor efficacy of decontamination, anti-inflammatory and immunosuppressive therapies in paraquat human intoxications. *PLOS one*. 4(9): e7149.

Guo, F., Sun, Y. B., Su, L., Li, S., Liu, Z. F., Li, J., Hu, X. T. and Li, J. (2015). Losartan attenuates paraquat-induced pulmonary fibrosis in rats. *Human and Experimental Toxicology*.34 (5): 497-505.

Gupta, P.K. (2018). Toxicity of Herbicides. Veterinary Toxicology, Basic and Clinical Principles, 3rd edn., *Elsevier*: 553–567.

Haripriya, B., Lakshman, M. and Sudha, V. (2017). Influence of paraquat (PQ) induced acute toxicity on body weights and haemato-biochemical parameters in experimental rats. *International Journal of Livestock Research*.6 (7): 396-398.

Hossain, M. M. (2015). Recent perspective of herbicide: Review of demand and adoption in world agriculture. *Journal of the Bangladesh Agricultural University*. 13 (1): 13-24.

Hu, X., Shen, H., Wang, Y. and Zhao, M. (2017). Liver X receptor agonist TO901317 attenuates paraquat-induced acute lung injury through inhibition of NF-κB and JNK/p38 MAPK signal pathways. *BioMed Research International*. 2017.

Huang, C. J., Yang, M. C. and Ueng, S. H. (2005). Subacute pulmonary manifestation in a survivor of severe paraquat intoxication. *The American Journal of the Medical Sciences*. 330(5): 254-256.

Lalruatfela, P. L., Saminathan, M., Ingole, R. S., Dhama, K. and Joshi, M. V. (2014). Toxicopathology of paraquat herbicide in female wistar rats. *Asian Journal of Animal and Veterinary Advances*. 9 (9): 523-542.

Nagao, M., Zhang, W. D., Takatori, T., Itakura, Y., Yamada, Y., Iwase, H., Oono, T. and Iwadate, K. (1994). Identification and dynamics of paraquat in the bone marrow, thymus and spleen in rats using immunohistochemical techniques. *The Japanese Journal of Legal Medicine*. 48(3): 166-168.

Okolonkwo, B. N., Nwachuku, E. O., Ene, P. C. and Okeke, C. U. (2014). The preventive effect of vitamin C on the cellular and functional integrity of kidney cells in rats following repeated exposure to paraquat. *Journal of Xenobiotics*.4 (1):29-39.

Pourgholamhossein, F., Rasooli, R., Pournamdari, M., Pourgholi, L., Samareh-Fekri, M., Ghazi-Khansari, M., Iranpour, M., Poursalehi, H. R., Heidari, M. R. and Mandegary, A. (2018). Pirfenidone protects against paraquat-induced lung injury and fibrosis in mice by modulation of inflammation, oxidative stress, and gene expression. *Food and Chemical*

Toxicology. 112: 39-46.

Ren, M., Wang, Y. M., Zhao, J., Zhao, J., Zhao, Z. M., Zhang, T. F., He, J., Ren, S. P. and Peng, S. Q. (2014). Metallothioneins attenuate paraquat-induced acute lung injury in mice through the mechanisms of anti-oxidation and anti-apoptosis. *Food and Chemical Toxicology*. 73: 140-147.

Sato, Y., Kamo, S., Takahashi, T. and Suzuki, Y. (1995). Mechanism of free radical-induced hemolysis of human erythrocytes: hemolysis by water-soluble radical initiator. *Biochemistry*. 34(28): 8940-8949.

Snedecor, G. W. and Cochran, W. G.(1994). Statistical methods, 8th Edn., *Ames: Iowa State Univ. Press Iowa*.

Suntres, Z. E. (2002). Role of antioxidants in paraquat toxicity. *Toxicology*. 180 (1): 65-77.

Tavakol, H. S., Farzad, K., Fariba, M., Abdolkarim, C., Hassan, G., Seyed-Mostafa, H. Z. and Akram, R. (2015). Hepatoprotective effect of *Matricaria chamomilla.* L in paraquat induced rat liver injury. *Drug Research*. 65 (02): 61-64.

Vuksa, M., Neskovic, N., Vitorovic, S. and Karan, V. (1983). Subacute toxicity of paraquat in rats—biochemical effects. *Ecotoxicology and Environmental Safety*. 7 (5): 475-483.

Wershana, K. Z. (2001). The influence of vitamin C or selenium on paraquat induced toxicity in Guinea Pigs. *Pakistan Journal Biological Sciences*.4: 81-88.

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