

Ameliorative effects of *Aloe vera* gel on male reproductive performance in *Drosophila melanogaster* after paraquat exposure

Usha Rani^{1&2}, Manvender Singh² and Krishan Kumar Selwal^{1*}

¹Department of Biotechnology, Deenbandhu Chotu Ram University of Science & Technology, Murthal, India

²Department of Biotechnology, University Institute of Engineering & Technology, Maharshi Dayanand University, Rohtak, India

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ABSTRACT

Paraquat (PQ) generally considered safe and widely used in agriculture as herbicide to protect crops against unwanted organisms. The oxidative stress generated by PQ may adversely affect reproductive performance in model organisms and humans. In the present study, we made an investigation on paraquat exposure and male reproductive performance in *Drosophila melanogaster*. Newly emerged virgin individuals of *Drosophila melanogaster* were separated gender-wise and were grown on control diet. Male individuals were divided into five different groups namely (i) Control, (ii) PQ intoxicated, (iii) Pre-treated, (iv) Post-treated and (v) Co-treated. For Paraquat intoxication, three day old virgin male individuals were exposed to Paraquat for 24 hours in 18mM concentration. Control flies were not intoxicated with Paraquat. PQ intoxicated flies were fed on control diet before and after paraquat intoxication. For Pre-treated groups and Post treated groups, flies were fed on *Aloe vera* supplemented diet before and after paraquat treatment, respectively whereas flies were fed on *Aloe vera* supplemented diet both before and after paraquat treatment for Co-treated groups. After PQ treatment, individuals from PQ-treated group, Post-treated group; and PQ-AV Co-treated groups were put in control and *Aloe vera* gel supplemented diet respectively for next 2 days to recover from stress. 6-day old male individuals from all the five groups namely, control, PQ-treated, Pre-treated, Post-treated and PQ-AV Co-treated were allowed to copulate with same aged virgin female individuals for mating assay. For each group, at-least 10 pairs were used in triplicates. After mating, male individuals were scarified to evaluate superoxide dismutase (SOD) activity and female individuals were analyzed for fecundity for next seven consecutive days. The results of the present study suggest that Paraquat exposure to male individuals prior to mating significantly and negatively affects the reproductive and fitness parameters in *Drosophila melanogaster*. It may infer from the results that *Aloe vera* gel supplementation to the paraquat exposed male *Drosophila* individuals improves the male reproductive performance by reducing paraquat induced oxidative stress.

Key words : Pesticide, Paraquat, Fecundity, Oxidative stress, *Drosophila melanogaster*.

Introduction

Pesticides are used extensively in agriculture throughout the world to protect crops against un-

wanted microorganisms (Mehdi and Qamar, 2011, 2013). Among so many others, paraquat is generally considered safe and is widely used pesticide. Its oxidative stress generation capacity which targets

the nervous system of the insects makes it a good pesticide. However, a dermal contact or spray exposure may result in limited and localized injury (Lock and Wilks, 2010). Some countries have banned the use of Paraquat as a casual or deliberate ingestion of it has an extremely high mortality rate (Senarathna *et al.*, 2009). Paraquat is selectively absorbed by the alveolar cells of the lung resulting in lung fibrosis (Agarwal *et al.*, 2009). It has been reported to cause Parkinson's disease in farm workers (Tanner *et al.* 2011), death by asphyxiation (Schiefer *et al.*, 1997), induce Parkinson's-like neurological disease in rats (Ossowska *et al.*, 2006), severe oxidative stress by interacting in REDOX system (Bus and Gibson, 1984), adversely affects the spermatogenesis (Fathi *et al.*, 2015). It has also been reported to reduce sperm quality, including count, motility, and morphology (Ashoka and De Silva, 2013).

Aloe vera is also known as a miracle plant due to its healing and curative properties (Davis *et al.*, 1989, Wu *et al.*, 2006). In spite of the large number of studies, only a few studies are available which demonstrates the reproduction enhancing potential of *Aloe vera* in different model organisms. Therefore, in the present study, copulation duration of male individuals and fecundity of female individuals were analyzed to evaluate the PQ induced oxidative and reproductive stress and ameliorative effect of *Aloe vera* gel supplementation in *Drosophila melanogaster*.

Materials and Methods

Wild type Oregon-K flies were used in the present study. Paraquat was purchased from Sigma Aldrich. 18mM concentration of Paraquat was used to induce the stress in three-day old male individuals. After stress of Paraquat, flies were put in vials with control diet for 2-days to recover from the stress. On 6th day, the Paraquat exposed male individuals were allowed to mate with same aged virgin female individuals grown on control diet. Parallel experiments were run with control male and female individuals without Paraquat exposure for comparison and analysis of Paraquat effect. Subsequently copulation duration and SOD activity of male individuals and fecundity of female individuals were analyzed.

Statistical analysis

The results were analyzed using Statistica software. The data was expressed as mean value with stan-

dard error. Differences were considered significant at $p < 0.05$.

Results and Discussion

Copulation duration in minutes for five different groups of *Drosophila* male individuals has been shown in Figure 1. Results showed that male flies from control groups copulate longer whereas individuals from paraquat intoxicated group showed lesser copulation duration. *Aloe vera* supplementation to *Drosophila* flies showed improved copulation duration in all three conditions i.e. prior to PQ intoxication, Post PQ intoxication and Co-treatment. The results showed that the fecundity also followed similar pattern (Figure 2 & 3). It may be due to that the male individuals from control group were able to fertilize the optimum number of ovules and resulted in optimum fecundity of mated females. Similarly the male flies supplemented with *Aloe vera* gel showed increased female fecundity after mating whereas the Paraquat treated male individuals

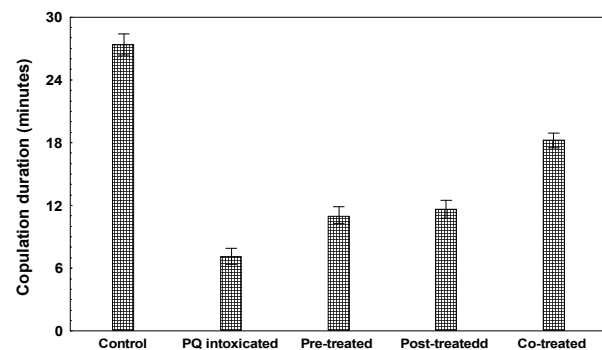


Fig. 1. Bar graph showing mean values for copulation duration in five different groups of *Drosophila melanogaster* male individuals.

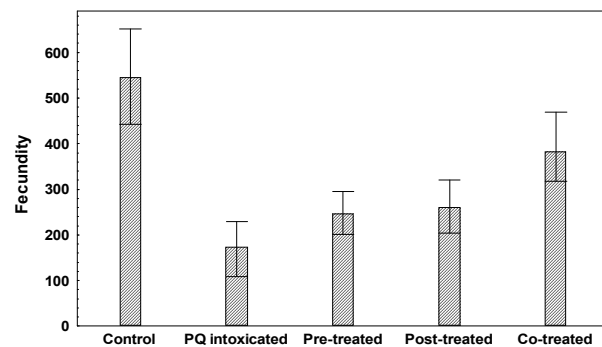


Fig. 2. Bar graph showing total fecundity of female individuals mated with five different groups of *Drosophila melanogaster* male individuals.

showed oxidative stress in the mating experiment which was evident by the significant reduction in female fecundity after mating. Paraquat induces oxidative stress in a wide range of living organisms including insects, rats, mice, mammals. Under the Paraquat induced oxidative stress, male individuals of *Drosophila melanogaster* were unable to reproduce efficiently (Figure 4). As a result the fecundity of female flies reduced significantly.

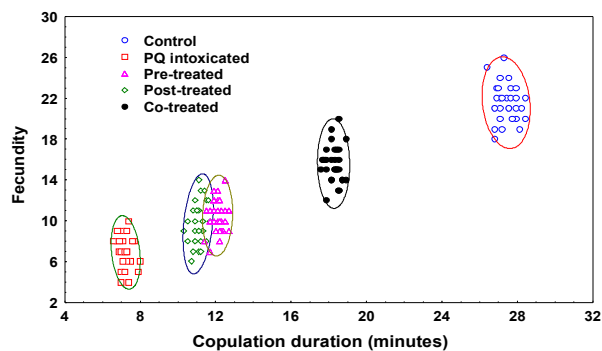


Fig. 3. Scatter-plot showing positive correlation of copulation duration with female fecundity in five different groups of *Drosophila melanogaster*.

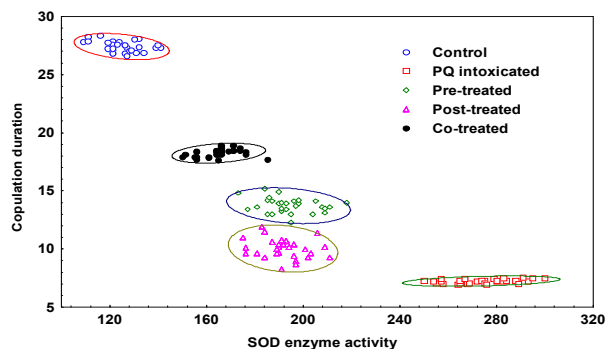


Fig. 4. Scatter-plot showing positive correlation of copulation duration with SOD enzyme activity in five different groups of *Drosophila melanogaster*.

Conclusion

The results of the present study revealed significant effect of oxidative stress induced by paraquat on reproductive performance of male *Drosophila melanogaster*. The results of the present study support the hypothesis of concerning the existing relationships between oxidative stress and reproduction performances.

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