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Paracetamol mediated changes modifies the photosynthetic efficiency of *Vigna radiata*

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ABSTRACT

In our previous study, we observed that paracetamol (panadol) influences the germination rate and growth biomarker of the *Vigna radiate* plants. To dissect out the paracetamol mediated changes in this study was undertaken. For this study, 10 days old seedling of *Vigna radiata* exposed to range of pandol concentration (.005, .01, .1, or 1.0 mg/L) through foliage of plants and harvested at 31st day. Obtained data revealed the dual response of panadol concentration: 0.1 mg/L of panadol significantly increased chlorophyll and leaf gas exchange traits (net photosynthetic rate, stomatal conductance, internal carbon dioxide concentration, water use efficiency and maximum quantum yield of PS II), whereas, 1.0 mg/L of panadol significantly reduced the above parameters. It is therefore concluded that low level of panadol enhanced photosynthetic efficiency of *Vigna radiata* plants; however, the response is concentration dependent and could be used as growth regulators for *Vigna radiata* plants.

Key words: Maximum quantum yield of PSII, Panadol, Photosynthesis.

INTRODUCTION

Plants have the ability to accumulate toxic metals and organic pollutants in their tissues and act as a phytomarker with distinguished change in the growth, morphology and physiology of the plants. In the recent past, excess use of drugs or pharmaceuticals included among the substances hazardous to living organisms directly or indirectly by altering its metabolism and physiological process (Kümmerer, 2004; Verkleij, 1994). It is primarily about environmental contamination and wastewater sludge. In addition to acidification and eutrophication of soils and water may also indirectly contribute to the reverse effects on plant and animal species. Moreover, pharmaceutically active substances are detected in surface water, groundwater and soil, wherein their concentration ranges from ng/l in µg/l (Hirsch *et al.*, 1999), to small flows of 1 mg/l. These toxic mobile elements enter the environment continuously (Hilton and Thomas, 2003) and their impact depends on exposure and time (Heberer, 2002). Studies also revealed that high polarity and low volatility drugs bioaccumulate and persistent in the environment for longer time (Breton and Boxall, 2003).

One of the various pharmaceutically active substances, paracetamol, is among the medicines which represents one of the most widespread pharmaceuticals in waste waters which has negative effects on human, animals, microbes, and plants. In Madinah City-Saudi Arabia alone, more than five tons of paracetamol had been dispensed to public hospital and medical clinics for the year of 2009 (Shraim *et al.*, 2017). Chemically, paracetamol is a benzene ring substituted by a hydroxyl group and an amide group in the (1, 4)-para position. With its two activated groups, the benzene ring is highly reactive towards electrophilic aromatic substitution. In humans, paracetamol is detoxified by liver, where sulfation and glucoronidation result in the formation of inactive, nontoxic and glucuronid conjugates. With their multiple functions and the variety of possible substrates, glycosyltransferases apply a predominant function of buffering the impacts concerning either biotic then abiotic challenges on the plant (Bowles *et al.*, 2005). Paracetamol is available as a generic medication with trade names including Tylenol and Panadol among others.

Mung bean has been used by researchers as an indicator for toxicity of environmental pollution (Oliveira, 2012). This study was designed to analyse the effect of increasing concentrations of panadol (paracetamol) on the photosynthetic efficiency of mung bean(*Vigna radiata)* grown under environmentally controlled conditions through sand culture and dissect out concentration dependent study that will unravel the influence of panadol on the photosynthetic efficiency and its related attributes of mung bean.

MATERIALS AND METHODS

Authentic and certified seeds of *Vigna radiata* were procured from local market of Riyadh, Saudi Arabia. Prior to sowing, healthy and uniform sized seeds were surface sterilized with 1% sodium hypochlorite solution for 10

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minutes followed by repeated washing with double distilled water (DDW). Panadol (PD) was used as the source of paracetamol. It was procured from Sigma Aldrich India Ltd. A stock solution of PD (10 mg/L) was prepared by dissolving the required quantity of paracetamol in 10 mL of DDW in a 100 mL volumetric flask and final volume was made up to the mark by using deionized water. The required concentration (0.0005, 0.01, 0.1 or 1.0 mg/L) of PD were prepared by the dilution of stock solution and the concentrations of PD was based on the screening of various concentrations (Data unpublished). Tween 20 added to the solution prior to the application.

The experiment was conducted with 25 cups of 350 ml size under completely randomized block design. Each treatment was replicated five times and each replicate (cup) contains three plants. In a plant growth chamber, the surface sterilized seeds were sown in acid washed sand, irrigated with deionized water only till germination. After germination, the seedlings were supplemented with nutrient solution (Hewitt, 1966) on alternate days till the termination of experiment. At 10 d stage, the seedlings were treated with deionized water (control), 0.0005, 0.01, 0.1 or 1.0 mg/L of PD for 10 days through its foliage. Each seedling was sprinkled thrice. The nozzle of the sprayer was adjusted to give each seedling about 3 mL of double distilled water (DDW) or PD solution. All replicates allowed to grow under controlled environmental conditions; 28/22°C (day/ night); 65/75 % RH (day/night) and 14 hours photoperiod and were irrigated with deionized water and nutrient solution on alternate days. Plants were harvested at 30 days stage to assess the chlorophyll content, net photosynthetic rate, maximum quantum yield of PSII and related attributes.

The Soil-Plant Analyses Development (SPAD) values of chlorophyll content in the leaf was measured, under natural conditions by using the SPAD chlorophyll meter (SPAD-502; Konica, Minolta sensing, Inc., Japan). Photosynthetic traits were determined on the third fully expanded leaves between 11:00 and 12:00 hours by using an infrared gas analyzer (IRGA) portable photosynthetic system (LI-COR 6400, LI-COR, Lincoln, NE, USA). To measure net photosynthetic rate (P_N) and its related attributes [stomatal conductance (*gs*), internal CO_2 concentration (*ci*), water use efficiency (WUE)], the air temperature, relative humidity, CO_2 concentration and Photosynthetic Photon Flux Density (PPFD) were maintained at 25ºC, 85%, 600 µmol mol^{-1} and 800 µ mol mol^{-2} s^{-1}, respectively.

Fv/Fm tests whether or not plant stress affects photosystem II in a dark adapted state, in this experiment, maximum quantum yield of Photosystem II (Fv/Fm) was measured by using Leaf Chamber Fluorometer (LI-COR 6400-40, Portable photosynthesis system, LI-COR, Lincoln, NE, USA). All the measurements were carried out at a PPFD of 1500 µmol m^{-2} s^{-1}, with a constant airflow rate 500 µmol s^{-1}. The sampled leaves were dark adapted for 30 min prior to measurement of Fv/Fm.

Data was statistically analyzed using SPSS, 17.0 for windows (SPSS, Chicago, IL, USA). Analysis of variance (ANOVA) was performed on the data to determine the least significant difference (LSD) between treatment means with the level of significance at $P<0.05$.

RESULTS AND DISCUSSION

SPAD chlorophyll content: PD (0.1mg/L) generated a maximum increase in chlorophyll content (SPAD values) which was 27.99 % more than that of control. Out of three other concentrations (0.01, 0.005 or 1.0 mg/L) of PD treatments, 0.005 and 1.0 showed similar responses. The order of effectiveness of PD on the SPAD chlorophyll content was 0.1 mg/L > 0.01 mg/L > 0.005 ≥ 1.0 mg/L.

Net Photosynthetic rate and stomatal conductance: Treatment of plants with 0.1 mg/L of PD significantly increased net photosynthetic rate and stomatal conductance in comparison to other concentrations (0.01, 0.005, and 1.0 mg/L) over control plants. In contrast, 0.005 and 1.0 mg/L showed similar response and are non-significant among themselves. The positive response of PD on net

Table 1: Effect of Panadol (PD; 0.0001, 0.005, 0.001, 0.005, 0.01, 0.1, or 1.0 mg/L) induced changes on SPAD chlorophyll content, net photosynthetic rate and stomatal conductance of *Vigna radiata* at 30 days after sowing.

Treatment	SPAD chlorophyll	Net photosynthetic rate (µ mol CO_2 m^{-2} s^{-1})	Stomatal conductance [mol (H_2O) m^{-2} s^{-1}]
Control	35.40 ± 0.30	11.97 ± 0.11	0.030 ± 0.001
PD (0.005)	39.64 ± 0.40	13.64 ± 0.12	0.033 ± 0.002
PD (0.01)	42.48 ± 0.41	14.39 ± 0.14	0.035 ± 0.002
PD (0.1)	45.31 ± 0.44	15.68 ± 0.11	0.037 ± 0.001
PD (1.00)	39.64 ± 0.39	13.76 ± 0.09	0.033 ± 0.001
LSD @ 5%	1.71	0.42	0.012

ANOVA Table

SPAD chlorophyll			Net photosynthetic rate			Stomatal conductance		
Source of variation	**DF**	**F-calculated**	**Source of variation**	**DF**	**F-calculated**	**Source of variation**	**DF**	**F-calculated**
Replication	2		Replication	2		Replication	2	
Treatment	4	159.19	Treatment	4	139.94	Treatment	4	116.88
Error	8		Error	8		Error	8	
Total	14		Total	14		Total	14	

Table 2: Effect of Panadol (PD; 0.0001, 0.005, 0.001, 0.005, 0.01, 0.1, or 1.0 mg/L) induced changes on internal CO_2 concentration, water use efficiency and maximum quantum yield of *Vigna radiata* at 30 days after sowing.

Treatment	Internal CO_2 concentration(mg/L)	Water use efficiency(m mol mol^{-1})	Maximum quantum yield(Fv/Fm)
Control	246 ± 1.25	4.20 ± 0.04	0.71 ± 0.005
PD (0.005)	275 ± 1.95	4.66 ± 0.03	0.78 ± 0.004
PD (0.01)	295 ± 1.85	5.37 ± 0.02	0.86 ± 0.005
PD (0.1)	312 ±2.01	5.62 ± 0.02	0.90 ± 0.006
PD (1.00)	270 ± 1.61	4.78 ± 0.01	0.79 ± 0.005
LSD @ 5%	9.84	0.16	0.027

ANOVA Table

Internal CO_2 concentration			Water use efficiency			Maximum quantum yield		
Source of variation	DF	F-calculated	Source of variation	DF	F-calculated	Source of variation	DF	F-calculated
Replication	2		Replication	2		Replication	2	
Treatment	4	144.79	Treatment	4	192.25	Treatment	4	113.08
Error	8		Error	8		Error	8	
Total	14		Total	14		Total	14	

photosynthetic rate and stomatal conductance was 0.1 mg/L > 0.01 mg/L > 0.005 ≥ 1.0 mg/L.

Internal CO_2 concentrations and water use efficiency: The internal CO_2 concentration and water use efficiency affected by the foliar application of PD in the concentration dependent manner. Out of various concentrations (0.1, 0.01, 0.005, or 1.0 mg/L) of PD tested, 0.1 mg/L is proved the best and significantly increased the internal CO_2 concentration and water use efficiency by 26.82 % and 33.80 % in comparison to the other concentrations and control as well.

Maximum quantum yield of PS II (Fv/Fm): A considerable variability in Fv/Fm of *Vigna radiata* plants was observed under various levels of PD (Table 2). Fv/Fm increased significantly under 0.1 mg/L of PD compared to the control. The maximum increase was 26.76 % in the presence of 0.1 mg/L of PD over the other concentrations and non-treated control plants.

In the present study, chlorophyll and net photosynthetic rate decreased at the higher concentration whereas 0.1 mg/L PD significantly increased the chlorophyll content and net photosynthetic rate. Azevedo *et al.* (2005) reported that the reduction in chlorophyll content and net photosynthetic rate may occur because of result of enzymatic degradation of these pigments or by inhibiting their biosynthesis.

Due to structural similarity between paracetamol and salicylic acid, it is believed that paracetamol also act in the similar manner when exogenously exposed to plants. Various workers reported a significant increase in net photosynthesis *Brassica juncea* L. Czern & Coss,)maize or soybean plants sprayed with an optimum concentration of SA (Fariduddin *et al.*, 2003; Khan *et al.*, 2003), while higher concentrations of SA had an inhibitory effect. Similar response is shown in the present study. In addition to this, exogenous application of paracetamol enhanced the internal CO_2 concentration, water use efficiency, stomatal conductance and transpiration rate in *Brassica juncea* (Fariduddin *et al.*, 2003). Further, Khan *et al.* (2003 reported an increase in transpiration rate and stomatal conductance in response to foliar application of SA and other salicylates in corn and soybean. In a study on soybean, foliar application of salicylic acid enhanced the water use efficiency, transpiration rate and internal CO_2 concentration (Kumar *et al.*, 2000).

In the recent years, the technique of chlorophyll fluorescence has become ubiquitous in plant ecophysiologicalstudies. No investigation into the photosynthetic performance of plants seems complete without fluorescence data. In this study, maximum quantum yield of PSII (Fv/Fm) are concentration dependent and highest concentration showed decrease in the Fv/Fm, however, 0.1 mg/L of PD significantly increase the maximum quantum yield of PS II. Structurally similar compound, salicylic acid also maintaining a higher Rubisco activation state and greater PSII efficiency and also accelerated the increase of net photosynthetic rate mainly by recovery of PSII function (Wang *et al.*, 2010).

CONCLUSION

It may be concluded that the exposure of *Vigna radiata* plants to varied concentrations of Panadol (paracetamol) resulted in increase of chlorophyll content and photosynthetic efficiency. However, the highest concentration (1.0 mg/L) showed damaging response and reduced the photosynthetic efficiency of *Vigna radiata*. It could be believed that promising response of Panadol for increasing photosynthetic efficiency is concentration dependent.

REFERENCES

Azevedo, H., Glória Pinto, C.G., Fernandes, J., Loureiro, S. and Santos, C. (2005). Cadmium effects on sunflower growth and photosynthesis. *J. Plant Nutr.* **28:** 2211-2220.

Bowles, D., Isayenkova, J., Lim, E.-K. and Poppenberger, B. (2005). Glycosyltransferases: managers of small molecules. *Curr. Opin. Plant Biol.* **8:** 254-263.

Breton, R. and Boxall, A. (2003). Pharmaceuticals and personal care products in the environment: regulatory drivers and research needs. *QSAR & Combinatorial Science*. **22**: 399-409.

Fariduddin, Q., Hayat, S. and Ahmad, A. (2003). Salicylic acid influences net photosynthetic rate, carboxylation efficiency, nitrate reductase activity, and seed yield in *Brassica juncea*. *Photosynthetica*. **41:** 281-284.

Heberer, T. (2002). Occurrence, fate, and removal of pharmaceutical residues in the aquatic environment: a review of recent research data. *Toxicol. Lett*. **131**: 5-17.

Hewitt, E.J. (1966). Sand and Water Culture Methods Used in the Study of Plant Nutrition: Commenwealth Agricultural Bureaux, U.K.

Hilton, M.J. and Thomas, K.V. (2003). Determination of selected human pharmaceutical compounds in effluent and surface water samples by high-performance liquid chromatography–electrospray tandem mass spectrometry. *J. Chromatogr*. **1015:** 129-141.

Hirsch, R., Ternes, T., Haberer, K. and Kratz, K.-L. (1999). Occurrence of antibiotics in the aquatic environment. *Sci. Total Environ*. **225**: 109-118.

Khan, W., Prithiviraj, B. and Smith, D.L. (2003). Photosynthetic responses of corn and soybean to foliar application of salicylates. *J. Plant Physiol*. **160**: 485-492.

Kümmerer, K. (2004) Pharmaceuticals in the Environment: Sources, Fate, Effects and Risks. Springer, Berlin.

Kumar, P., Lakshmi, N. and Mani, V. (2000). Interactive effects of salicylic acid and phytohormones on photosynthesis and grain yield of soybean (*Glycine max* L. Merrill). Physiol. *Mol. Biol. Plants.* **6:**179-186.

Oliveira, H. (2012). Chromium as an environmental pollutant: insights on induced plant toxicity. J Bot. 2012.

Shraim, A., Diab, A., Alsuhaimi, A., Niazy, E., Metwally, M., Amad, M., Sioud, S. and Dawoud, A. (2017). Analysis of some pharmaceuticals in municipal wastewater of Almadinah Almunawarah. *Arab. J. Chem*. **10**: S719-S729.

Verkleij, J. (1994). Effects of heavy metals, organic substances, and pesticides on higher plants. Ecotoxicology of Soil Organisms. SETAC Press139-161.

Wang, B.-L., Shi, L., Li, Y.-X. and Zhang, W.H. (2010). Boron toxicity is alleviated by hydrogen sulfide in cucumber (*Cucumis sativus* L.) seedlings. *Planta*. **231:**1301-1309.