

EFFECT OF GLYPHOSATE ON CHLOROPHYLL AND CAROTENOIDS IN WEED SPECIES

(Parthenium hysterophorus L. and Cyperus rotundus L.)

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ABSTRACT

Photosynthetic effects of the Glyphosate (X-(Phosphonomethyl) glycine) on *Parthenium hysterophorus L* and *Cyperus rotundus L*. were studied. Glyphosate was diluted with distilled water (25ppm) for foliar application. The seedlings were maintained under natural photoperiod condition (about 2000 //E M'2 Sec" 400-700nm (PAR) at 12.00 noon with a light regime of approximately 11 hours at 34 to 40°C followed by 13 hours dark at 16-20°C. The experiment was carried out to analyze the effects of glyphosate by measuring chlorophyll 'a', chlorophyll 'b', total chlorophyll and carotenoid contents in weed species. The experimental plants were sprayed 20 days after emergence and control plants were maintained by a simultaneous spray of deionized water. Leaves of *Parthenium hysterophorus L* and *Cyperus rotundus L* weed species were collected at 5days interval following the foliar application of herbicide up to 15th day. Results showed that Chlorophyll (Total chlorophyll, chlorophyll a and b) and carotenoid content was adversely affected as the duration and concentration of glyphosate increased

Key words: Glyphosate, Weed species *Parthenium hysterophorus L.*, *Cyperus rotundus L.*, Total chlorophyll, Carotenoids.

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INTRODUCTION

Glyphosate [N-(Phosphonomethyl) glycine] is a relatively non-selective herbicide developed by Monsanto and is considered to be a very significant discovery (Stephen *et al.*, 2012). Glyphosate [N-(phosphonomethyl) glycine] has become the most widely used herbicide in the world; in large part because of its use in transgenic, glyphosateresistant (GR) crops (Duke, 2012; Duke and Powles, 2008, 2009). It is particularly valuable for control of most annual and perennial weed species. It is applied to the foliage of vegetation to control. It is readily absorbed by leaves and translocated through the symplast (Sprankle *et al.*, 1975). Since its introduction,

glyphosate has been considered a toxicologically and environmentally safe pesticide, due to its low mammalian toxicity, relatively short environmental half-life, and extremely low activity in soil due to its binding to soil minerals (reviewed by Duke *et al.*, 2012). Besides several competitive factors, weeds are also known to inhibit crop growth due to their allelopathic activity in the agro-ecosystems. Weeds often produce enormous biomass during their life cycle which upon decay can be phototoxic to other plants in the vicinity (Weston and Duke 2003; Batish *et al.* 2007). Glyphosate has several desirable properties that have contributed to its widespread use (Duke *et al.*, 2008). Singh *et al.* (2005) concluded that *Parthenium hysterphorus* L. residues exert a negative influence on the early growth of Brassica crops by releasing water-soluble phenolics. Likewise, Batish *et al.* (2009) attributed the negative effect of *Ageratum conyzoides* residues on rice germination and early growth to the release of water soluble phenolic acids into the soil environment. During its several decades of use over vast areas, no significant adverse secondary effects of the herbicide have been established, other than the intense selection pressure that has resulted in the evolution of GR weeds. In fact, its use in GR crops has been associated with several environmental benefits (Cerdeira and Duke, 2006 and Garner and Nelson, 2008). The topic of evolution of GR weeds has been dealt with in detail in many research papers and reviews (Powles and Yu, 2010 and Shaner *et al.*, 2012).

Once glyphosate interacts with soil, whether applied directly to the soil surface, exuded from a plant root, or released from decomposing plant tissue, it is subject to various processes that control its environmental behavior and fate, including retention (sorption-desorption), transport, and degradation. Of these processes, sorption is arguably the most important as it controls the availability for degradation, plant uptake, and offsite transport. Sorption of glyphosate to soil has been extensively reviewed (Duke, 1988; Vereecken, 2005; Borggaard and Gimsing, 2008).

In the present investigation an attempt was made to study the comparative account of biological response of herbicide glyphosate in two weed species. Among herbicides used to control weeds glyphosate is one of the foliar applied herbicide.

MATERIAL AND METHODS

Pot culture experiment was conducted with freshly young seedlings of *Parthenium hysterophorus* L (Congress Grass) and *Cyperus rotundus* L (nut grass). Weed species are common and dominant in the vegetable crop fields in the surrounding area of Tirupati. Tirupati is located between 79°25' 1-ongitude and 3°58' Latitude and is situated in Chittoor District, Andhra Pradesh, India. The weed seeds were collected from the ground nut crop fields and seeds were soaked in the seed pan for 2 days to obtain maximum germination percentage. After 20 days, the plantlets were sown in the experimental pots containing red soil supplemented with farmyard manure in the ratio of 1:3. The seedlings were maintained under natural photoperiod condition (about 2000 //E M² Sec" 400-700nm (PAR) at 12.00 noon with a light regime of approximately 11 hours at 34 to 40°C followed by 13 hours dark at 16-20°C.

Herbicide Treatment

The herbicide used in the present study was (X-(Phosphonomethyl) glycine) marketed under trade name glyphosate and was obtained from Monsanto Company. Glyphosate diluted with distilled water (25ppm) was selected for foliar application. In the present study only 20-22

days old seedlings of *Parthenium hysterophorus* L and *Cyperus rotundus* L were treated with glyphosate using "Aspee" sprayer (Aspee, Bombay) to the drip point at 8.00 «LID without using any wetting agent. The control plants were maintained by a simultaneous spray of deionized water. Pot culture experiments were conducted in the Department of Environmental Science, Sri Venkateswara University, Tirupati, India.

Sampling

Leaves of *Parthenium hysterophorus* L and *Cyperus rotundus* L weed species were collected at 5 days interval following the foliar application of herbicide upto 15th day. The leaves were thoroughly washed first with tap water followed by deionized water and were dried with filter paper. Fresh leaf material was used for the determination of chlorophyll and carotenoids. The chlorophyll content was estimated according to the method of Arnon (1949), Total carotenoids were determined as per the method of Jensen and Jensen (1971).

Data were expressed as Mean \pm Standard deviation of mean (SD). Results were statistically analyzed by student's test (Pillai and Shinha 1968)

RESULTS AND DISCUSSION

The present work was undertaken to study the effect of glyphosate on chlorophyll fractions like chlorophyll 'a', chlorophyll 'b' and total chlorophyll were estimated in leaves of *Parthenium hysterophorus* L and *Cyperus rotundus* L. The results of this study showed that, the control plants chlorophyll 'a' content was more in *Parthenium hysterophorus* L and *Cyperus rotundus* L, but the glyphosate treatment caused decrease in all the plants up to 15th day over the control respectively in both the weed species. Thus, the chlorophyll 'a' content is decreased significantly in both the treated weed species (Tab.1, Fig.1). The treated plants showed that the gradual decrease in chlorophyll 'b' content in all the stages of experimental days. The maximum decrease was noticed in both the weed plants on 15th day, The decreased levels chlorophyll-b was observed in *Parthenium hysterophorus* L and *Cyperus rotundus* L (0.220; 0.186; 0.146; 0.125 and 0.218; 0.173;0.137; 0.118) on 5th-10th and 15th day respectively over the control (Tab.2, Fig.2). The results revealed that the total chlorophyll content was continuously decreased up to 15th day in *Parthenium hysterophorus* L and *Cyperus rotundus* L over the control. The reduction in chlorophyll a, chlorophyll b and total chlorophyll contents in 15 days treated leaves is nearly 50% over control in both the cases. However, the weed species showed consistent decrease at all the experimental days. The result was similarly compared with several reporters (Luiz Henrique Saes Zobiolo et al, 2012, Zobiolo et al., 2010e). The content of total carotenoid was also reduced with effect of glyphosate caused gradual decreased carotenoid content in both the weed species up to 15th day. In both cases the maximum declined was noticed on 15th day only, The decreasing level of carotenoid content in *Parthenium hysterophorus* L and *Cyperus rotundus* L, (-0.746,-0.470,-0.315,-0.218; and -0.814,-0.513,-0.422,-0.132) (Tab.4, Fig.4) when compare to control. It was also resulted by the reports of Peter J. Ralph, 2000).

Table: 1. Effect of glyphosate on chlorophyll ‘a’ content on weed species (mg/g.fwt)

Name of the weed species	Days after treatment			
	Control	5 th day	10 th day	15 th day
<i>Parthenium hysterophorus L.</i>	0.360 ± 0.032	0.241 ± 0.028	0.205 ± 0.034	0.170 ± 0.021
<i>Cyperus rotundus L.</i>	0.345 ±0.020	0.247 ±0.028	0.198 ± 0.031	0.160 ±0.040

= ± SD Values are mean of 5 replications

Table: 2. Effect of glyphosate on chlorophyll ‘b’ content on weed species (mg/g.fwt)

Name of the weed species	Days after treatment			
	Control	5 th day	10 th day	15 th day
<i>Parthenium hysterophorus L.</i>	0.220 ± 0.029	0.186 ± 0.021	0.146 ± 0.023	0.125 ± 0.092
<i>Cyperus rotundus L.</i>	0.218 ±0.074	0.173 ±0.016	0.137 ± 0.013	0.118 ±0.060

= ± SD Values are mean of 5 replications

Table: 3. Effect of glyphosate on total chlorophyll content on weed species (mg/g.fwt)

Name of the weed species	Days after treatment			
	Control	5 th day	10 th day	15 th day
<i>Parthenium hysterophorus L.</i>	0.580 ± 0.027	0.399 ± 0.012	0.351 ± 0.019	0.295 ± 0.048
<i>Cyperus rotundus L.</i>	0.392 ±0.023	0.337 ±0.019	0.278 ± 0.071	0.256 ±0.031

= ± SD Values are mean of 5 replications

Table: 4 Effect of glyphosate on carotenoid content in weed species (mg/g.fwt)

Name of the weed species	Days after treatment			
	Control	5 th day	10 th day	15 th day
<i>Parthenium hysterophorus</i> L.	0.746 ± 0.094	0.470 ± 0.147	0.315 ± 0.084	0.128 ± 0.0719
<i>Cyperus rotundus</i> L.	0.814 ±0.024	0.513. ±0.019	0.422 ± 0.130	0.132 ±0.042

= ± SD Values are mean of 5 replications

Fig. 1 Effect of glyphosate on chlorophyll 'a' content on weed species (mg/g.fwt)

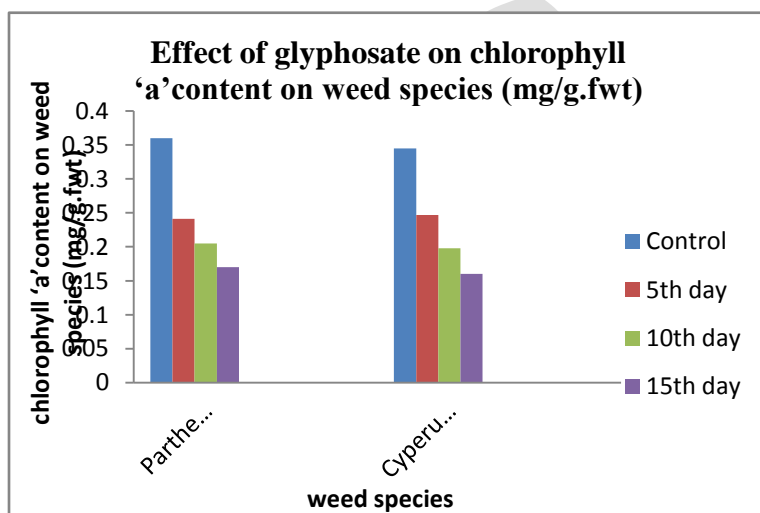


Fig. 2 Effect of glyphosate on chlorophyll 'b' content on weed species (mg/g.fwt)

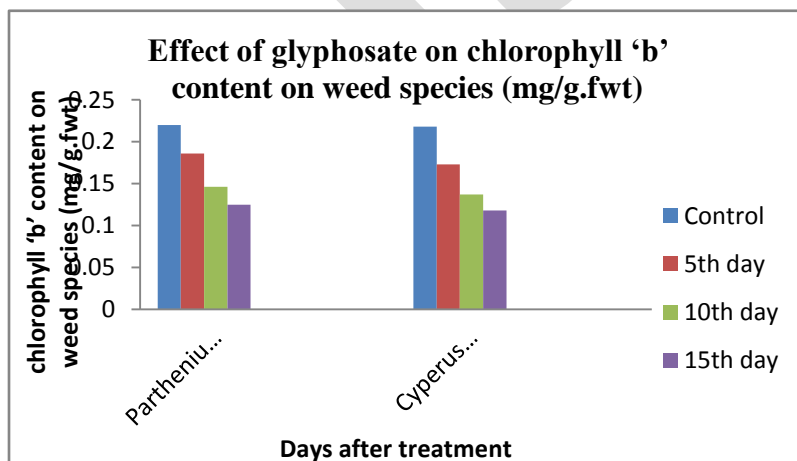


Fig. 3 Effect of glyphosate on total chlorophyll content on weed species (mg/g.fwt)

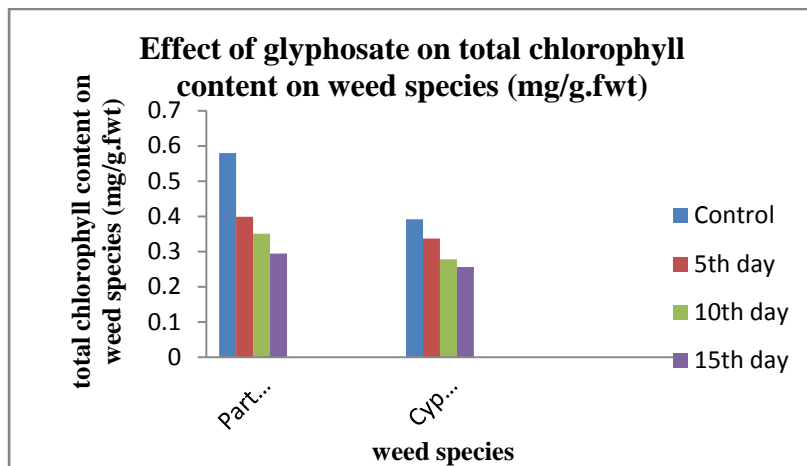
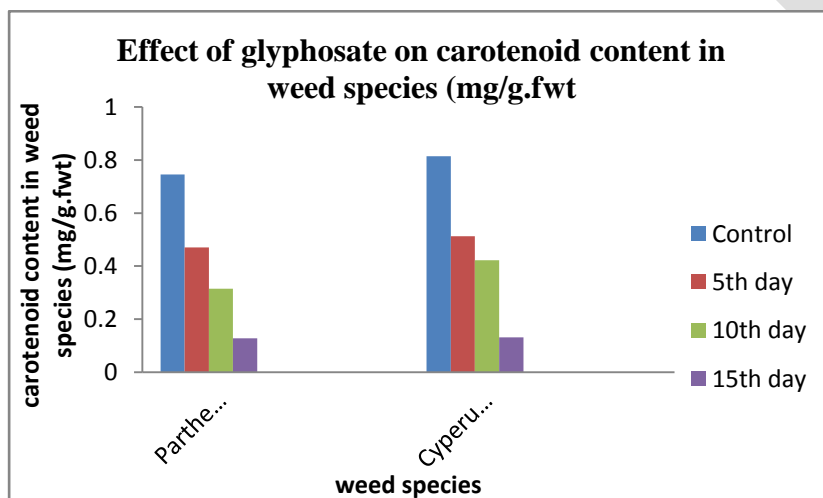


Fig.4 Effect of glyphosate on carotenoid content in weed species (mg/g.fwt)



CONCLUSION

The present investigation concluded that obviously from our results that the glyphosate has a strong effect on *Parthenium hysterophorus* L and *Cyperus rotundus* L. with the increase of glyphosate concentration applied to weeds, even at low dose (25 ppm) effects are directly related to changes in the total chlorophyll and carotenoid content. Similarly chlorophyll 'a' and chlorophyll 'b' contents were also decreased significantly in both the weeds upto 15th day. Carotenoid content was declined in both the weed species might have contributed to the photo oxidation of chlorophylls. The degradation of chlorophyll and carotenoid content in the present study testifies glyphosate action on photosynthetic activity.

REFERENCES

1. Stephen O. Duke, John Lydon, William C. Koskinen, Thomas B. Moorman, Rufus L. Chaney, and Raymond Hammerschmidt. (2012). Glyphosate Effects on Plant Mineral Nutrition, Crop Rhizosphere Microbiota, and Plant Disease in Glyphosate-Resistant Crops. *J. Agric. Food Chem.* 60, 10375–10397.
2. Sprankle, P, W.F. Megdtt, and D. Penner. (1975). Absorptions action, and of glyphosate, *WeedSci.* 23; 235-240.
3. Duke, S. O., Baerson, S. R., Rimando, A. M. (2012). Herbicides: Glyphosate. In Encyclopedia of Agrochemicals ([http:// onlinelibrary.wiley.com/book/10.1002/047126363047126363X](http://onlinelibrary.wiley.com/book/10.1002/047126363047126363X)September 12, 2012).
4. Duke, S.O., Powles, S.B., 2008. Glyphosate: A once in a century herbicide. *Pest Manag. Sci.* 64, 319–325.
5. Duke, S.O., Powles, S.B., 2009. Glyphosate-resistant crops and weeds: Now and in the future. *AgBioForum* 12, 346–357.
6. Cerdeira, A. L.; Duke, S. O. (2006). The current status and environmental impacts of glyphosate-resistant crops: A review. *J. Environ. Qual.* 35, 1633–1658.
7. Garner, J. G.; Nelson, G. C.(2008). Herbicides, glyphosate resistance and acute mammalian toxicity: simulating and environmental effect of glyphosate-resistant weeds in the USA. *Pest Manag. Sci.* 64, 470–478.
8. Powles, S. B.; Yu, Q. (2010). Evolution in action: plants resistant to herbicides. *Annu. Rev. Plant Biol.* 2010, 61, 317–347.
9. Shaner, D. L.; Lindenmeyer, R. B (2012). Ostie, M. H. What have the mechanisms of resistance to glyphosate taught us? *Pest Manag.* 2012, 68, 3–9.
10. Duke, S. O. (1988). Glyphosate. In *Herbicides: Chemistry, Degradation, and Mode of Action*, Vol. 3; Kearney, P. C., Kaufman, D. D., Eds.; Dekker: New York, pp 1–70.
11. Vereecken, H.(2005). Mobility and leaching of glyphosate: a review. *Pest Manag. Sci.*, 61, 1139–1151.
12. Borggaard, O. K.; Gimsing, A. L.(2008). Fate of glyphosate in soil and the possibility of leaching to ground and surface waters. a review. *Pest Manag. Sci.* 64, 441–456.
13. Arnon DI. 1949. Copper enzymes in isolated chloroplasts, polyphenoxidase in beta vulgaris. *Plant physiology* 24: 1-15.
14. Jensen. S.L. and Jensen. A. (1971). Quantitative determination of carotenoids in photosynthetic tissue. In: *Methods in enzymology.* (Sampietro A, H.d.) Acad. Press. Xcw York and London.
15. Pillai, SK and Sinha, HC, 1968, Statistical methods for biological works. Ram Prasad and sons, Agra, India.
16. Weston La, Duke So. 2003. Weed and crop allelopathy. *Crit Rev Plant Sci* 22: 367–389.
17. Singh Hp, Batish Dr, Pandher Jk, Kohli Rk. 2005. Phytotoxic effects of Parthenium hysterophorus residues on three brassica species. *Weed Biol Manage* 5: 105–109.
18. Batish Dr, Kaur S, Singh Hp, Kohli Rs. 2009. Nature of interference potential of leaf debris of *Ageratum conyzoides*. *Plant Growth Regul* 57: 137–144.
19. Luiz Henrique Saes Zobiolo, Robert John Kremer, Rubem Silvério de Oliveira Jr and Jamil Constantin. Glyphosate effects on photosynthesis, nutrient accumulation, and nodulation in glyphosate-resistant soybean. *J. Plant Nutr. Soil Sci.* 2012, 175, 319–330.

20. Zobiolo, L. H. S., Oliveira Jr., R. S., Huber, D. M., Constantin, J., de Castro, C., Oliveira, F. A., Oliveira Jr., A. (2010c): Glyphosate reduces shoot concentration of mineral nutrients in glyphosateresistant soybeans. *Plant Soil* 328, 57–69.
21. Zobiolo, L. H. S., Kremer, R. J., Oliveira Jr., R. S., Constantin, J (2010e): Glyphosate affects photosynthesis in first and second generation glyphosate-resistant soybean. *Plant Soil* 336, 251–265.
22. Peter J. Ralph. Herbicide toxicity of *Halophila ovalis* assessed by chlorophyll a fluorescence. *Aquatic Botany* (66). (2000), 141–152.

