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Effect of Herbicide (Glyphosate) upon the fecundity and moulting of a terrestrial isopod (Philoscia javanensis) under lab condition

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Abstract

Soil isopods plays critical role to increase soil fertility. Indiscriminate use of Herbicides on agricultural purposes resulted deleterious effects on the biology and species diversity of soil arthropods. The effect of sublethal concentration of a herbicide glyphosate (N-(Phosphonomethyl)glycine), were treated to find out the fecundity and maturation of an edaphic isopod (Philoscia javanensis). The LC 50 and LC 100 values of Philoscia javanensis were calculated. The safe and sublethal concentrations of Glyphosate were very low indicating high vulnerability of these isopods to agrochemicals. The safe level concentration of Glyphosate was 0.6341ppm. The moulting intervals of P.javanensis showed variation between herbicide treated and normal P.javanensis. Herbicide treated P. javanensis showed drastic reduction in fecundity and also their moulting intervals were prolonged.

Key words: Glyphosate, Herbicide, Isopod, *Philoscia javanensis*, Moulting interval and Fecundity.

1. Introduction

Soils are integral part of ecosystems and constitute large number of soil micro arthropods. Soil micro arthropods enhance soil aggregation, porosity and thus increasing infiltration and reducing runoff. Agricultural practices negatively affects on different groups of soil fauna (Ponge *et al*., 2013) (Paredes and Lebeis 2016). Many pesticides are not easily degradable, they persist in soil, leach to groundwater and runoff can carry pesticides into aquatic environment and also entered into the food chain thereby affecting all living beings. In all parts of the world, farmers are addicted to using agrochemicals indiscriminately (Conway, 1984) Overuse of agrochemicals will hamper soil, environment as well as human health. Glyphosate (N-phosphonomethyl-glycine) base product is the leading non-selective amino phosphonate type herbicide for the annual and perennial weeds control (Piola *et al.,* 2013).

2. Materials and methods

Collection and rearing of *Philoscia javanensis*:

Live *Philoscia javanensis* were randomly collected from study area and cultured in laboratory in special culture (plastic) chambers. Plaster of paris and activated charcoal in the ratio 5:2 mixed with distilled water is used as base in the container. Baker's yeast pellets were offered as food for hatching animals along with decaying leaves of rubber and jack tree. Animals were acclimatized to the lab condition (Green, 1969)

Preparation of Stock Solution of Glyphosate (N-(Phosphonomethyl)glycine):

1000 ppm stock solution of Glyphosate was prepared by dissolving required quantity of chemicals in one liter of distilled water (APHA, 2012). From this stock solution various concentration like 0.5, 1, 2, 3, 5 and 6 ppm Glyphosate were prepared for bioassay.

Bio assay:

Experiments conducted in laboratory using cultured animals. Ten replicates of *P. javanensis* and a control were tested for each concentration of this agro chemicals. The mortality was recorded at 12, 24, 48, 72 & 96 hours intervals. Bioassay lethal concentrations LC 100 and LC 50 were calculated using probit analysis of Finney (Finney, 1980); Safe concentrations were determined by the method suggested by Hart *et al*., (1945). Total fecundity was calculated by counting the number of eggs in total oviposition and compared the normal group fecundity with glyphosate treated groups. Moulting interval of normal and agrochemical treated groups were also recorded. A pair of subadults of ten replicates were exposed to sub lethal concentration for 15 days and the fecundity was observed, compared the fecundity with normal group

Safe level was calculated using the formula

$$SF = \frac{\text{LC50 96 hours x 0.3}}{S^2}$$

Were, 0.3 is a constant and SF = Safe Level

$$S = \frac{\text{L C-50 48 hour}}{\text{L C-50 72 hour}}$$

3. Results and Discussion

Mortality:

Table 1: Percentage mortality of *Philoscia javanensis* treated with herbicide, Glyphosate

	Percentage mortality				
Con: in ppm	12 hr	24 hr	48hr	72 hr	96 hr
0.5	0	0	2	2	2
1	0	3.8	5.4	10.33	12.46
2	2.86	15.86	19.99	30.33	44.76
3	15	30.46	38.48	40.99	53.76
4	30.44	40.2	48.14	60.86	72.18
5	40.86	50.45	62.31	75	80.46
6	54.38	67.37	78	90.36	100

Phyloscia javanensis treated with glyphosate were found that at 0.5 ppm, the mortality started. The mortality was recorded at 12, 24, 48, 72 and 96 hours intervals. 2, 5, 19, 38, 48, 62 and 78 percentage mortality were recorded for glyphosate concentration of 0.5,1,2,3,4,5 and 6 ppm at 48 hours intervals. 2, 10, 30, 40, 60, 75 and 90 percentage mortality was found at 72 hours intervals. 2, 12, 44, 53, 72, 80 and 100 percentage mortality was recorded for 0.5,1,2,3,4,5 and 6 ppm concentration of glyphosate at 96 hours intervals (Table 1). The result showed that the concentration of herbicide, glyphosate increases the mortality rate of *Philoscia javanensis* also increases.

Table 2: Lethal Concentration 50 (LC 50) and Lethal Concentration 100 (LC 100) values of glyphosate for *Philoscia javanensis*

Glyphosate	12 hr	24 hr	48hr	72 hr	96 hr
LC 50	5.789	4.7382	4.0433	3.4264	2.9432
LC 100	10.399	8.8532	7.6033	6.5364	5.7732

Lethal concentration 50 of Herbicide, glyphosate treated Philoscia javanensis were 5.879 ppm at 12 hours, 4.7382 ppm at 24 hours, 4.0433 ppm at 48 hours, 3.4264 ppm at 72 hours and 2.9432 ppm at 96 hours. LC 100 values of Glyphosate treated *P. javanensis* were calculated to be 10.399, 8.8532, 7.6033, 6.5364 and 5.7732 ppm at 12, 24, 48, 72 and 96 hours intervals (Table 2). The safe level concentration of Herbicide, Glyphosate was 0.6341 ppm and its sublethal concentration found to be 0.7358 ppm. (Table 3).

Table 3: Safe level and sublethal concentrations of Glyphosate for *Philoscia javanensis*

Herbicide	S	SF (ppm)	Sublethal (ppm)
Glyphosate	1.18	0.6341	0.7358

Fecundity:

Table 4: Fecundity of normal *Philoscia javanensis*

Replicates	Oviposition 1	Oviposition 2	Oviposition 3	Oviposition 4	Oviposition 5	Mean
1	26	28	25	18	15	22.4
2	24	22	24	20	17	21.4
3	22	24	20	15	12	18.6
4	24	20	18	16	11	17.8
5	20	23	18	17	13	18.2
6	22	23	20	18	12	19
7	25	27	24	20	16	22.4
8	23	26	24	18	15	21.2
9	24	25	21	18	13	20.2
10	25	27	23	19	12	21.2

The maximum fecundity of normal *P. javanensis* was seen in oviposition 2 in each groups. The maximum fecundity was observed in the group 1 & 7, 22 eggs per broods and minimum was recorded in group 4, 17 eggs per broods (Table 4).

Table 5: Fecundity of *Philoscia javanensis* after treatment with sublethal concentration of Glyphosate

Replicates	Oviposition 1	Oviposition 2	Oviposition 3	Oviposition 4	Oviposition 5	Mean
1	9	8	7	6	9	7.8
2	6	7	6	5	9	6.6
3	10	8	7	5	8	7.6
4	10	11	8	9	6	8.8
5	9	6	8	6	5	6.8
6	12	10	9	8	7	9.2
7	12	12	10	7	6	9.4

8	11	9	9	9	8	9.2
9	11	10	7	7	7	8.4
10	12	9	9	7	6	8.6

The average number of eggs laid by *P.javanensis* after the treatment of sublethal concentration of Glyphosate was observed between a minimum of 7.6 eggs/broods and maximum of 9.4 eggs /broods. The number of eggs produced by females after the treatment of Glyphosate was less, it's revealed that the herbicide affect drastically on isopods (Table 5).

Moulting interval:

Table 6: Normal moulting interval of *Philoscia javanensis*

Stage	Sample 1 (Days)	Sample 2 (Days)	Sample 3 (Days)	Sample 4 (Days)	Sample 5 (Days)	Sample 6 (Days)	Sample 7 (Days)	Sample 8 (Days)	Sample 9 (Days)	Sample 10 (Days)	Group mean (Days)
Rest stage	5.5	5	4.5	4	4	5	5.5	4.5	4	5	4.7
1st moult	14.25	13.25	12	13	14	14.5	13	12	12.65	14	13.265
2nd moult	18	17	17	18.5	17	18	17.5	18	17.23	18	17.623
3rd moult	21.53	20	22	22.56	22.4	21.52	20.26	20	21.45	20	21.175
4th moult	29	30.25	31.36	31	29.5	30.45	30	29	31	31.26	30.277
5th moult	38.12	38.16	37.56	38	37	37.96	38	38.13	37	38.42	37.835

Normal *Philoscia javanensis* showed 5 moults, the first moult started after 5 days of hatching, second moult after 13 days of hatching, third moult after 21 days of hatching, fourth moult after 30 days of hatching and fifth moult after 37 days of hatching (Table 6).

Table 7: Moulting interval of *Philoscia javanensis* after treatment with Glyphosate

Stage	Sample 1 (Days)	Sample 2 (Days)	Sample 3 (Days)	Sample 4 (Days)	Sample 5 (Days)	Sample 6 (Days)	Sample 7 (Days)	Sample 8 (Days)	Sample 9 (Days)	Sample 10 (Days)	Group mean (Days)

Rest stage	7	7.8	6.26	6.5	7	7.94	6.46	7	6	6.28	6.824
1st moult	17.23	16.25	17	18	18.5	17.43	18	16.32	17	17.46	17.319
2nd moult	28.23	27	27.54	29	28.5	27.5	28	29.46	27.87	28	28.114
3rd moult	34.13	32.56	33.16	32	33.9	33	32.15	34	32.58	34.5	33.194
4th moult	41.56	42.45	40.26	43.22	40.6	42.36	41.15	39	41.5	40.15	41.223
5th moult	53.23	49.23	50.48	50	51.5	52.13	49.36	50.11	53	51.36	51.035

After the treatment with the sub lethal concentration of glyphosate treated *P. javanensis* moulting intervals becomes prolonged. First moult only being after 17 days, second moult on 28 days, third moult on 33 days, fourth moult on 41 days and fifth moult on 51 days of hatching (Table 7).

4. Conclusion

From the study revealed that extensive application of herbicides affect on the soil microarthropod *Philoscia javanensis,* these also indirectly affecting the soil fertility. Soil isopods body covered with thin cuticle which can absorb the agricultural pesticides easily through their body surface. Herbicide treated *P. javanensis* showed drastic reduction in fecundity and also their moulting intervals were prolonged. Indiscriminate agrochemical applications kills both pests and natural enemies concurrently and results in the resurgence of non-target insects, and the misuse and overuse of insecticides can harm farmers, consumers, and the environment too.

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