

*Full Length Research Paper*

# Effects of acetochlor (herbicide) on the survival and avoidance behaviour of spiders

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**This study was designed to evaluate the potential effects of acetochlor (herbicide) on the survival and avoidance behaviour of lycosid spiders example *Lycosa terrestris*. During the topical toxicity experiment, *P. birmanica* was found to be more susceptible to acetochlor than *L. terrestris*. Although, there was 10% mortality at field rate concentration by topical exposure, we did not observe any mortality during residual toxicity experiment for both spider species even at double field rate concentration. There was no difference in the time spent by both species on the herbicide or water treated part of filter paper. It was concluded that use of acetochlor at the recommended rate in the agricultural field is safe for tested spider species, which are important biological control agents in the study area.**

**Key words:** Herbicide, residual toxicity, acetochlor.

## INTRODUCTION

Spiders are the most important, abundant and beneficial predators in agro-ecosystem (Zhang, 1992). Being polyphagous predators, they are able to prey on a number of insect pests and play an important role in regulation of insect populations in agricultural crops (Riechert and Bishop, 1990). To support the beneficial effect of spiders, management practices that are safer or have least effect on their survival should be adopted by farmers. Herbicides are being applied more frequently for the management of unwanted plants in the agricultural crops (Ammann, 2005). Although, herbicides are supposed to have least effects on spiders but evidence showed that spider numbers are affected even by relatively low rate of herbicide application (Baines et al., 1998; Bell et al., 2002).

Integrated pest management (IPM) is aimed to use only selective herbicides in agricultural crops. Before introducing a new herbicide in agro-ecosystems, it is necessary to evaluate its side effects on non target organisms (Amalin et al., 2000). Spiders are important

candidate in the bio-control programs throughout the world. Use of herbicides in agro-ecosystems not only affect the spider density but influence their behaviour (Haughten et al., 2001; Tietjen, 2006). Exposure of herbicides may also affect different aspects of the biology of spiders, such as their reproduction and development (Schneider et al., 2009). Thus, before attempting to use a herbicide in the fields, there is a need for the clearer understanding of potential effects of herbicides on the spiders and other non-target invertebrates (Bell et al., 2002). Although, the effects of insecticides on the survival and behaviour of spiders has been studied, there is scarcity of the data on impact of herbicides on spiders (Asteraki et al., 1992).

*Lycosa terrestris* and *Pardosa birmanica* are two most abundant lycosid spiders in rice ecosystems of central Punjab, Pakistan (Tahir and Butt, 2008). Both are very active hunters. *L. terrestris* is often found on the foliage to chase prey, while *P. birmanica* restrict itself on the ground. These spiders were found to be very effective against rice pests (Tahir and Butt, 2009). Farmers in Pakistan use large quantities of herbicides in their agricultural fields including rice fields. In this study, we investigated the effect of residues of acetochlor

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(commonly used herbicide in the study area) on the survival of these two important biological control agents. This study will help to evaluate the effects of acetochlor (if any) on the selected spider species and to decide whether these herbicides could be safe candidate for integrated pest management program in the study area or not. Some researchers have reported that inhabitants of agro-ecosystem avoid the wide spread and frequently used herbicide even if they are not exposed directly (Lautenschlager and Sullivan, 2002; Tatum, 2004). So, avoidance behavior (whether spider species avoid any surface treated with herbicide or not) of the selected spiders was also studied in the laboratory.

## MATERIALS AND METHODS

### Collection of spiders

Spider specimens were collected by visual search from unsprayed rice fields situated in District Sheikupura, Punjab, Pakistan from August to November, 2008. Adult specimens of *L. terrestris* and *P. birmanica* were selected for the study. Collected spiders were placed individually in 28 ml plastic cups, covered with mesh cloth and transported to the laboratory for experiment. In the laboratory, each spider was transferred to an opaque plastic container (8.75 cm diameter, 3.75 cm length) containing pieces of rice plant leaves for shelter and moisture wick. Each container was covered with mesh cloth and maintained at room temperature ( $34 \pm 5^\circ\text{C}$ ), humidity (60 to 78%) and a light : dark cycle of 12:12 h. The spiders were fed with larvae of wild-type *Drosophilla* cultured in the laboratory.

### Topical bioassays

Effects of acetochlor (herbicide commonly used on rice, cotton, sugarcane, peanuts and soybean in the study area) on spiders was evaluated by topical and residual bioassay tests. For each experiment, three replicate tests were performed. For topical application, three concentrations of acetochlor viz., double field rate, field rate and half field rate were used. Field rate is 500 ml / 100 L of water per acre. For each concentration, 20 specimens (10 males and 10 females) of each species were used. Before application of acetochlor, spiders were anesthetized by brief exposure to ethanol. The herbicide was applied (0.5  $\mu\text{l}$ ) topically on the dorsum of each spider using a micropipette. Response of organism was recorded as unaffected (showing normal behaviour), affected (showing irregular pattern of movement), paralyzed (completely immobilized but showed movements when touched) and dead (totally non-responsive). Response of organisms was recorded at each dose after 2, 4, 8, 16 and 24 h of exposure.

### Residual toxicity

For residual testing, filter papers (6 x 12 inches) were dipped in 150 ml solution of herbicide and dried at room temperature. Filter papers were kept in sunlight to stimulate degradation under natural conditions for 12 h. For each concentration, 20 specimens (10 males and 10 females) of each species were used. Spiders were exposed to the treated filter papers for one hour and then transferred to the clean glass jars individually. Control spiders were exposed to the filter papers dipped in simple water. None of the specimens was used in more than one trail and no food was offered

to the spiders during the experiment. Mortality was assessed both in the treated and control cohort at 2, 4, 8, 16, 24 and 48 h. Criterion for recording the response of spiders after exposure to the herbicide was same as described for topical bioassay experiment.

### Avoidance behaviour

To record the avoidance behaviour, only recommended rate of herbicide was used and Whatman filter papers (12 x 9 inches) were divided into two halves (by drawing a line in the centre of the filter paper). One half of the filter paper was dipped in herbicide solution, while the other half was dipped in the distilled water. Filter papers were dried in sunlight for one hour. Spiders were release on the filter papers and time spend by each spider on herbicide or water treated part was recorded. For each spider, reading was recorded for 30 min. T-test was used to compare the mortality of males and females. Paired t-test was used to compare the time spent by spiders on the herbicide or water treated part. Analysis of variance (ANOVA) was used to compare the time spent by both species on the herbicide or water treated part of filter paper. Statistical software SPSS (version 10) was used for the statistical analyses.

## RESULTS

### Topical bioassays

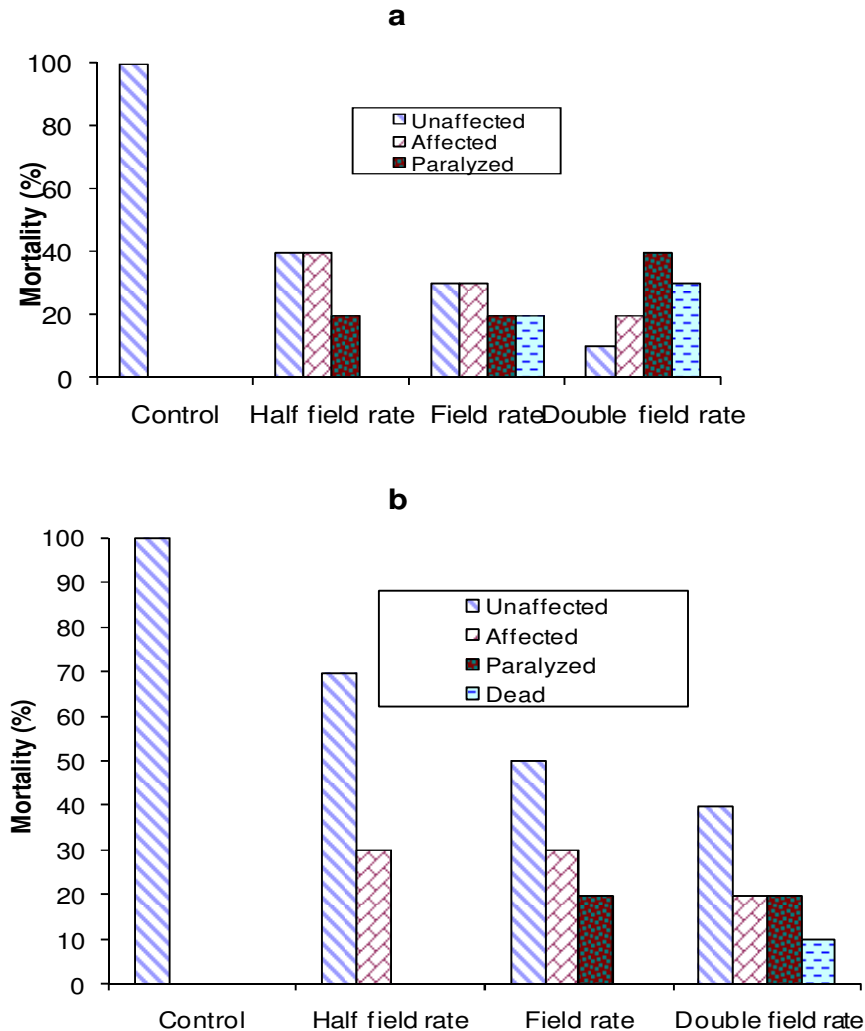
*P. birmanica* was found to be more susceptible to acetochlor than *L. terrestris*. For *P. birmanica*, 20% mortality was recorded at field rate and 30% at double field rate (Figure 1a). However, we did not recorded any mortality at half field rate or field rate in *L. terrestris* but observed 10% mortality at double field rate (Figure 1b). There was no difference in the mortality between males and females ( $t = 1.40$ ;  $P = 0.23$  for *P. birmanica* and  $t = 1.40$ ;  $P = 0.23$  for *L. terrestris*).

### Residual toxicity

Residuals toxicity bioassay test showed that acetochlor is safe for both species at three tested concentration as no dead or paralyzed specimens was recorded in any concentration even at 48 h of exposure. Some specimens were found to be affected (8% *P. birmanica* and 6% *L. terrestris*) at early h (after 2 to 8 h after the exposure) with double field rate dose but they become active again after 24 h. More than 90% of the specimens of both species which were found to be affected after exposure to the double field rate were males. None of the female specimens was found to be affected.

### Avoidance behaviour

There was no difference in the time spent by *P. birmanica* and *L. terrestris* on the part of the filter paper treated with herbicide or water ( $t = 0.45$ ;  $P = 0.65$ ; Figure 2a for *P. birmanica* and  $t = 0.10$ ;  $P = 0.92$ ; Figure 2b for *L. terrestris*). Results were also non significant when two



**Figure 1.** Percent mortality of *P. birmanica* (a) and *L. terrestris* (b) at different concentrations of acetochlor during topical exposure experiment.

species were compared for the time spent on the part of the filter paper treated with herbicide or water (df = 1, 58; F = 0.53; P = 0.92 for herbicide treated part and df = 1, 58; F = 0.58; P = 0.44 for water treated part).

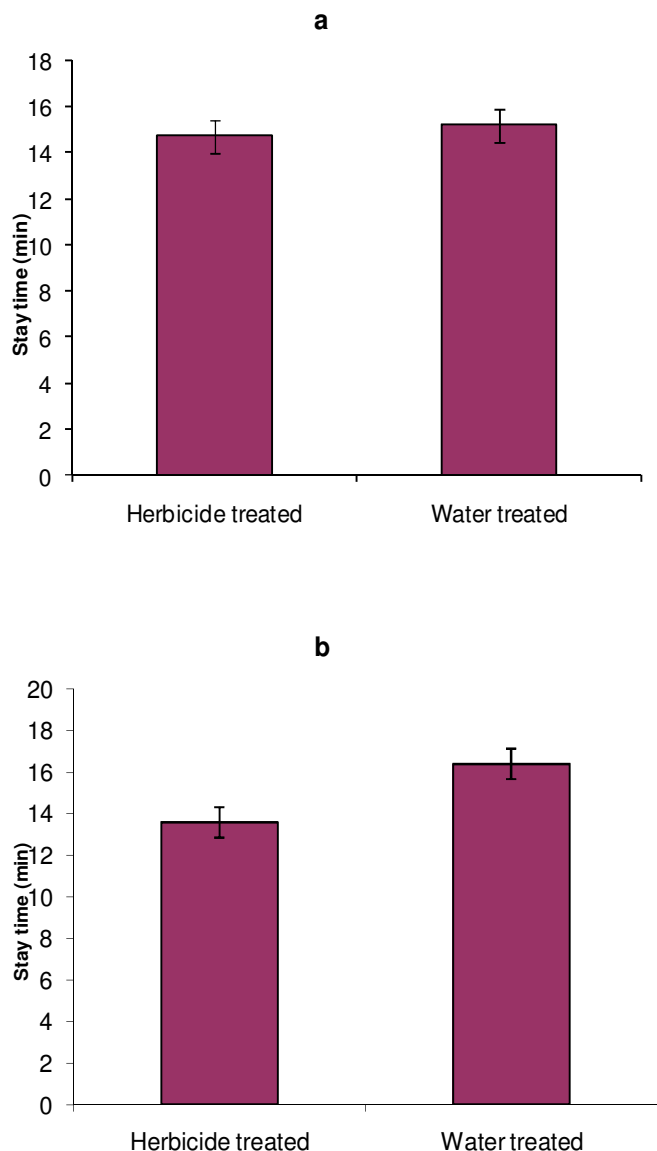
## DISCUSSION

Lycosids are important predatory arthropods that are sensitive to the commonly applied chemical sprays. However, several researchers have reported that effects of chemical sprays are species specific (Kawahara et al., 1971; Shaw et al., 2004; Pekar and Benes, 2008). During residual toxicity studies, none of the specimens of both spider species died or paralyzed even after 48 h of exposure of herbicide. Some specimens that were affected just after the exposure to the herbicide residues became active again after 24 h. These results suggested that acetochlor residues had negligible effects on *P.*

*birmanica* and *L. terrestris*. Michalkova and Pekar (2009) and Yardim and Edwards (1998) also reported negligible effects of herbicide (glyphosate) on *Pardosa agrestis*. Although, we also observed negligible effects of herbicide on the both experimental spider species, however, in agreement with Michalkova and Pekar (2009), we also cannot exclude the possibility that mortality would occur few days later, so there is need to record the data after 48 h.

It has been reported that spiders avoid some chemical (herbicides or pesticides) but not others (Pekar and Haddad, 2005). However, there is scarcity of data on the avoidance behaviour of spiders to acetochlor. Pekar and Haddad (2005) reported that members of genus *Pardosa* avoided surfaces treated with fresh residues of Decis or Nurelle but did not avoid surface contaminated with command (herbicide).

Although, we observed 10% mortality at field rate concentration by topical exposure, however, residual



**Figure 2.** Time spent by *P. birmanica* (a) and *L. terrestris* (b) on herbicide or water treated part of filter paper.

toxicity experiment showed that mortality was zero for both spider species even at double field rate concentration. The herbicide did not cause any repellence or mortality even at double field rate in the residual toxicity experiment. Based on these results, it can be concluded that biological control potential of the two spider species is not directly affected even after the herbicide (acetochlor) application in the field. Therefore, use of acetochlor at the recommended field rate in the agricultural field is safe for the tested spider species. As the effects of pesticides and herbicides are species specific, further studies should be conducted in order to test the effects of acetochlor on the other predatory

arthropods of agricultural crops.

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