# Research Article Effect of Low-lying Land Environment on Abundance of Cotton Spider Mites and Total Phenolic Content of Leaves and their Relationship

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**Abstract:** The possible advantage of cotton production established in low-lying land area over intensive culture pond was systematically investigated by comparing the abundance of cotton spider mites on these plants to that in conventional production areas at the Jiangbei farm, Hubei Province, China, over the period 26 May and 11 September 2011. Cotton fields (Ezamian No. 24F1) recently grown at low-lying land area supported significantly lower populations of cotton spider mites than conventional long established cotton fields. There were no significant differences in mite populations between cotton fields established in low-lying land area in the current year or 1 or 2 years earlier. The pest control advantage provided by pond areas was present whether or not acaricides were used. The total phenolic content of cotton leaves differed occasionally between treatments but did not seem to have affected the abundance of mites. The number of eggs, larva-nymph-adults, egg-larva-nymph-adults, the percentage of host plants colonized by cotton spider mites and the plant damage index were independent of the total phenolic content in leaves. The results are discussed in relation to integrated pest management and the mineral balance hypothesis.

Keywords: Cotton spider mite, Gossypium hirsutum, low-lying land, total phenolics content

## **INTRODUCTION**

Jingzhou city is located in the Middle Reaches of the Yangtze River, one of the most important bases of agricultural products of China and a typical wetland agricultural area adjacent to Yangtze River and Han River. The groundwater level is usually 100-150 cm in winter-spring and 50-100 cm in summer. In 2011, in Jingzhou city there were some 14100 km<sup>2</sup>, of land used to grow rice, cotton, fish, rape and vegetables, etc., with 108000 hm<sup>2</sup> in cotton and 90700 hm<sup>2</sup> in intensive culture ponds (fish, finless eel, crab, etc.). Tests were carried out in 2011 at the Jiangbei farm (Longitude 112.27°-112°43', North latitude 30.08°-30°22'), Jingzhou city, Hubei Province, China. There were 6600 hm<sup>2</sup> of agricultural planting with some 227 hm<sup>2</sup> of intensive culture ponds.

It is easy and common practice for land owners to use their land interchangeably as intensive culture ponds, cotton fields or rice paddy fields. The use of the land is often determined by changes in the market price of fish, cotton or rice paddy as well as the incidence of fish disease. It is a legal practice in Jingzhou city. Some 1400 and 22 hm<sup>2</sup> cotton of low-lying land have been transformed from intensive culture ponds to cotton production in Jingzhou city and at the Jiangbei farm test



Fig. 1: Cotton production field established in low-lying land area transformed from intensive culture ponds to since the current year (2011) at the Jiangbei farm (longitude 112.27°-112°43', North latitude 30.08°-30°22"), Jingzhou city, Hubei Province, China

site, respectively. In this type of cotton field, Cotton fields grown at low-lying land (the bottom of intensive culture ponds) are 1.5-2 m lower than the soil surface of traditional cotton fields, each low-lying land measured 1300-7000 m<sup>2</sup>, there are 1 ditches (100 cm width×100 cm depth) or 2 ditches (50 cm width×50 cm depth) that can be used to drain storm water away using small electric pumps (Fig. 1).

Cotton spider mites are the most destructive herbivorous arthropods of cotton, *Gossypium hirsutum* L. in the world. The group is composed of 5 dominant species in China, namely *Tetranychus cinnabarinus* 

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Boisduval, T. truncates Ehara, T. urticae Koch, T. turkestani Ugarov et Nikoskil, T. dunhuangensis Wang (Acari: Tetranychidae) (Krantz and Walter, 2009; Xu et al., 2011). T. cinnabarinus, the dominant species in the Middle Reaches of the Yangtze River of China, has a 15-18d life-cycle. Cotton spider mites of all stages overwinter in crop fields on winter crop green manure, grass, soil or litter. In late February-early March the following year, cotton spider mites start feeding in emerging host plants. Mites move to cotton seedlings as they become available. Cotton spider mites prefer the underside of leaves. The mites pierce the leaves to suck the sap causing a range of foliar symptoms, such as red leaves, commonly referred to as "the dragon's fire". Cotton spider mites can cause important yield losses and even total yield loss during outbreaks.

Recent studies reported controlling cotton spider mites using a range of farming practices such as cottonpaddy rice rotation (Ke *et al.*, 1995), horse bean-cotton inter-planting (Xu *et al.*, 2011), wheat-cotton interplanting (Cheng *et al.*, 1997) and jujube-cotton interplanting (Xu, 2001). However, the effect of low-lying land environment (pond-cotton rotation) and total phenolic content of cotton leaves on the abundance of cotton spider mites remains largely unknown.

Along with the time planted with cotton in Jingzhou city increases so does the damage caused by cotton spider mites. This study looked at the population dynamics of cotton spider mites and the relationship between the total phenolic content of leaves and the number of cotton mites, the percentage of damaged host plants and plant damage indices in 4 types of cultivated cotton fields with and without acaricide sprays.

The objective of this study was also to provide a theoretical basis for integrated pest management of cotton spider mite in the Middle and Lower Reaches of the Yangtze River of China.

## MATERIALS AND METHODS

**Treatment:** The study was conducted between 4 June and 24 September 2011. There were four treatments consisting of low-lying fields transformed from pond to cotton production 0 (2011), 1 (2010) or 2 years ago (2009) and a control field under yearly rape-cotton rotation since 2001 (CK). Except in the current year when low-lying fields was transformed into cotton production, a yearly rape-cotton rotation was used all treatments. Intensive culture ponds were built and cultured fish since 2006. Cotton (Ezamian No. 24F1) was used in all fields.

**Monitoring:** The experimental design consisted in a randomized complete block with four factors (2011, 2010, 2009, CK) replicated 3 times, each block (4 treatments) measured approximately 7000 m<sup>2</sup> and was divided into acaricides spray and no-spray area. The number of cotton spider mites in each treatment plot

was estimated using 5-point sampling monitoring pattern (fixed locations, but not fixed cotton plants). Acaricides were sprayed 8 times in the spray sections.

Monitoring was carried out 18 times throughout the season. Before 16 July 2011, mites were counted on 20 cotton plants at each sample point for a total of 100 cotton plants per sampling date. After 16 July 2011, mites were counted on 5 cotton plants at each sample point for a total of 25 cotton plants per sampling date. The number of cotton spider mites was counted on 3 randomly chosen leaves (upper, middle and lower) per plant and the percentage of plants with mites calculated. Acaricides were applied throughout the season in spray areas but only until 15 July 2011 in no spray areas. Monitoring of the no spray areas was carried out 8 times between 23 July and 3 September 2011 using the same method as in the spray areas.

In the acaricide sprayed fields, the total phenolic content of each of 10 cotton leaves per treatment, randomly collected (third complete leaf from the top of the plant) on 27 June, 17 August and 11 September 2011, was determined and the percentage cotton plants infested with cotton spider mites recorded at the same time. In the fields without acaricide sprays after July 15, on 24 July 2011, thirty cotton leaves were randomly collected in the same manner as above on 27 June 2011. The number of eggs, larva, nymphs and adults of cotton spider mites and the damage index of each leaf were recorded before they were dried, then dried, ground and stored at -60°C prior to the determination of total phenolic content. Total phenolic content of each cotton leaf was determined for each leaf (three replicates for each sample) using Folin-Ciocalteu's assay with gallic acid as standard (Folin and Denis, 1915).

The damage level of erythematous area in cotton leaves was estimated using a 1 to 5 scale (Zhang *et al.*, 1993).

**Statistical analysis:** All data was analyzed using DPS (Tang and Feng, 2002). Average percentage of cotton plants infested with cotton spider mites and total phenolic content of leaves were transformed by arcsin of the square root before applying a one-way ANOVA followed by Tukey test for multiple comparisons. A regression analysis was carried out to determine if the number of eggs larva-nymph-adults, egg-larva-nymph-adults, or the percentage of cotton plants infested with cotton spider mites was correlated with the total phenolic content in leaves. The correlation between plant damage level and total phenolic content in leaves was measured using multiple sequence correlation.

#### RESULTS

**Population dynamics of cotton spider mites in acaricide treated plots:** Over the period 4 June-11 August 2011 (Fig. 2 and 3A), the average percentage of cotton plants infested with cotton spider mites was



Fig. 2: Population dynamics of cotton spider mites in 4 cultivated cotton fields sprayed throughout the season with acaricides (4 June-24 August 2011)



Fig. 3: Percentage of cotton plants infested with cotton spider mites in 4 types of cultivated cotton fields on 4 June-11 August (A), 17-28 August (B) and 3-24 September (C) 2011. The cotton fields received acaricide sprays throughout the season

significantly higher in CK ( $6.37\pm2.56\%$ ) than in cotton plots 0 ( $0.20\pm0.13\%$ ), 1 ( $0.10\pm0.07\%$ ) or 2 ( $0.03\pm0.03\%$ ) years away from low-lying field transformation ( $F_{3,40} = 12.04$ , p = 0.0001). Also, there were no significant differences in the number of mites on cotton between treatments with 0, 1 or 2 years from low-lying field transformation. The average percentage of infested cotton plants was very low in all treatments in mid-season, 17-28 August 2011 (Fig. 2 and 3B), only CK (22.22±8.65%) and the 1 year after transformation treatment (1.33±0.77%) differed significantly ( $F_{3,8} =$ 5.49, p = 0.024), Towards the end of the season, on 3-24 September 2011 (Fig. 2 and 3C), the average percentage of cotton plants infested with cotton spider mites was as high in the current year transformation treatment (37.67±7.31%) than CK (43.67±8.46%) and both were significantly higher than the 1 (1.00±0.29%) or 2 (1.33±1.15%) years since transformation treatments ( $F_{3,12} = 21.45$ , p = 0.001).

Population dynamics of cotton spider mites in without acaricide treated plots: In the period 23 July-11 August 2011 (Fig. 4A and 5), the average percentage of cotton plants infested with cotton spider mites was low in all treatments and there were no significant differences among them ( $F_{3, 12} = 1.31$ , p = 0.316). In the period 17-28 August 2011 (Fig. 4B and 5), the average percentage of cotton plants infested increased over time in the conventional treatment  $(37.33\pm4.68\%)$  where it was significantly higher than in the 0  $(11.11\pm0.44\%)$ , 1 (6.67±3.08%) or 2 (4.89±0.89%) years since transformation treatments ( $F_{3,8} = 20.94$ , p = 0.0004). There were not significant differences among the transformation treatments. Over the period 23 July-28 August 2011 (Fig. 5), the average percentage of cotton plants infested with cotton spider mites increased with time and peaked on 3 September 2011 in all treatments: CK (74.7%), 2011 (89.3%), 2010 (16.0%), 2009 (18.7%).

Total phenolic content of cotton leaves from different cultivation treatments (plots sprayed with acaricides): There were differences ( $F_{3,36} = 66.51$ , p =



Fig. 4: Percentage of cotton plants infested with cotton spider mites in 4 types of cultivated cotton fields on 23 July-11 August (A) and 17-28 August (B) 2011. No acaricides were applied to the fields during that period



Fig. 5: Population dynamics of cotton spider mites in 4 cultivated cotton fields with no acaricide sprays after July 23, 2011

0.0001) in total phenolic content of cotton leaves (n = 10) among the different treatments from the acaricide sprayed treatment plots (Fig. 6). On 27 June 2011, total phenolic content of cotton leaves was highest in the 2 years since transformation treatment (3.634±0.220%) followed by the current year transformation treatment (2.77±0.18%), the 1 year since transformation (1.92±0.11%) and finally CK (1.01±0.08%). On 17 August 2011, total phenolic content in cotton leaves of the 2 year treatment (M.S.±S.E.) (4.38±0.17%) was significantly higher than the current year (2.75±0.17%), 1 year (3.20±0.25%) or CK (3.03±0.29%) treatments ( $F_{3.36} = 9.14$ , p = 0.0001). There were no significant



Fig. 6: Total phenolic content of cotton leaves (replicated 10 times) of different treatments sprayed with acaricides on 23 July-3 August 2011

differences among current year, 1 year or CK treatments. On 11 September 2011, there were significant differences in total phenolic content ( $F_{3, 36} = 4.82$ , p = 0.006) between the current year (3.61±0.21%) and 1 year (2.66±0.08%) treatments but no significant difference between other treatments: 2 year (2.90±0.29%), CK 3.38±0.17%).

The total phenolic content also varied significantly between sampling dates (Fig. 6). In the current year treatment, total phenolic content in cotton leaves was significantly higher on 11 September than on 17 August or 27 June ( $F_{2,27} = 6.74$ , p = 0.004). In the 1 year treatment, the in total phenolic content of cotton leaves was significantly higher on 17 August and on 11 September than 27 June ( $F_{2.27} = 16.55$ , p = 0.0001). There were no significant differences between on 11 September and on 17 August. In the 2 year treatment, the only significant difference in total phenolic content of cotton leaves was between 17 August and 11 September ( $F_{2,27} = 10.36$ , p = 0.0005). There were no significant differences between on 27 June and on 17 August, or between on 27 June and on 11 September. In the conventional treatment CK, the total phenolic content of cotton leaves was similar on 11 September and 17 August, both significantly higher than on 27 June  $(F_{2.27} = 57.97, p = 0.0001)$ . There were no significant differences between on 11 September and on 17 August.

Relationship between different indicators of population abundance and total phenolic content of leaves caricides: There was no significant linear correlation between the number of eggs (y = 0.41x + 9.91, r = 0.016,  $r_{28,0.05} = 0.349$ , p = 0.934), larva-nymph-adults (y = 3.11x + 26.52, r = 0.046,  $r_{28,0.05} = 0.349$ , p = 0.814), egg-larva-nymph-adults (y = 3.52x + 36.43, r = 0.032,  $r_{28,0.05} = 0.349$ , p = 0.869) or percentage of cotton plants infested by cotton spider mites (y = -11.18x + 45.05, r = -0.513,  $r_{28,0.05} = 0.349$ , p = 0.085) respectively and total phenolic content of

leaves. There was also no significant multiple sequence correlation between plant damage indices and total phenolic content (r = 0.359, df = 1, 28, p = 0.071).

## **DISCUSSION AND CONCLUSION**

Our results showed clearly that cotton fields established in low-lying land area will help reduce the abundance of cotton spider mites compared to that in conventional cotton fields. The pest control advantage was shown to be independent of the use or not of acaricides and to last for at least 2 years after transformation of the pond areas into cotton producing areas.

There were some differences between treatments in total phenolic content of cotton leaves but no relationship could be established with the abundance of mites. The number of eggs, larva-nymph-adults, egglarva-nymph-adults, percentage of plants with cotton spider mites and the plant damage level were independent of the total phenolics content in leaves. Zhang et al. (1993) reported a similar absence of correlation between red spider mite, T. urticae, plant damage level and total phenolic content in cotton leaves. However, some authors have found that an increase in total phenolic content in cotton can result in increase repellency to red spider mites and reduced number of eggs (Wu et al., 1996). Also, an aqueous solution of hydroxyphenylalanine and quinic acid was found to increase the content in phenolic compounds of strawberry leaves and high concentrations of catecholamines significantly delayed the development time of two spotted mites, T. urticae (Luczynski et al., 1990).

One can only speculate on the factor(s) responsible for the lower abundance of spider mites in cotton fields grown low-lying land area. The most likely factors would however be humidity and perhaps soil nutrients. Cotton spider mites prefer high temperature and low relative humidity (Cheng *et al.*, 1997). Wu *et al.* (1988) results showed that the increase of numbers of population of red spider mites, *T. cinnabarinus*, was inhibited in high air humidity. Cotton fields grown at low-lying land are 1.5-2 m lower than the soil surface of traditional Cotton fields (CK) (Fig. 1). The air humidity at low-lying land area is likely to be higher than that in conventional cotton fields creating an environment that may not be conducive to the growth and development of cotton spider mites.

Zang (1992) found that sediments from intensive culture ponds contain a large quantity of fish manure, feed, fertilizer and grass not used by fish, with an average of 0.80% N, 0.26% P, 1.70% K and 5.62% content organic matter content. The pond sediment can slowly release the fertilizer over 3-year or more (Tang, 1959). Organic crop production is credited, in a similar manner, with increased plant resistance to insect pests.

For example, Alyokhin *et al.* (2005) have also reported a general reduction in the abundance of the Colorado potato beetle on potato plants that had received manure amendments. Both cases may have a relationship to the mineral balance hypothesis (Phelan *et al.*, 1996) suggesting that the organic matter and microbial activity of organically managed soils maintain nutrient balance in plants. In turn, healthy plant growth would provide a level of resistance against insect pests.

It has also been suggested that the nitrogen concentrations in foliar tissues or phloem of plants growing in these balanced soils only reaches levels that are insufficient or unfavorable to outbreaks of sucking insects aphids (Van Bruggen, 1995). However, the link between insect pest levels and crop N levels remains difficult to confirm (Letourneau *et al.*, 1996). Actually, Dang *et al.* (2008) who studied the impact of different soil nitrogen concentrations on the population dynamics of cotton spider mites found that, the number of cotton spider mites on cotton leaves was higher when the content of nitrogen in cotton field soils was low.

Results have shown that low-lying land area transformed into cotton fields provide an additional level of plant resistance against cotton spider mites for at least two years after transformation. Additional research will be required to determine if the use of organic amendments would be helpful in prolonging the period of control.

#### ACKNOWLEDGMENT

The authors would like to acknowledge the comments of Dr. Gilles Boitaeu on an earlier version of the manuscript. This study was supported by grants from Public-benefit Industry Special Foundation No. 201103020 for Ministry of Agriculture of the People's Republic of China.

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