

## THE IMPACT OF DIFFERENT NITROGEN LEVELS ON *APHIS GOSSYPHII* AND THEIR PREDATORY BEETLE (*COCCINELLA SEPTEMPUNCTATA*) POPULATIONS, AND THE GROWTH OF PLANT LEAVES

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### ABSTRACT

The effect of different nitrogen (N) levels on cotton plant vegetative growth, and the occurrence of aphids (*Aphis gossypii*) and their predatory beetle (*Coccinella septempunctata*) was studied in an outdoor pot trial on the Experimental Farm of Huazhong Agricultural University. The cultivar “Huamian3109” was sown in pots filled with 33 kg of soil. Five different nitrogen levels — 120, 150, 180, 210 and 240 kg N/ha — were applied at the first flowering stage of the crop. The crop was observed in the morning when the insect pest and predators were not active. *Aphis gossypii* were recorded by observing 6 leaves per plant and 10 plants in each treatment. *C. septempunctata* was observed at 10 sites per treatment and each site consisted of 3 plants. The number of leaves on each plant was also counted. The maximum number of *C. septempunctata* observed was  $5.50 \pm 0.53/\text{site}$  and the peak population of *A. gossypii* was  $29.10 \pm 1.86/\text{plant}$ ; these were both recorded at the 5<sup>th</sup> observation time in the maximum N level treatment group (240 kg/ha), when the plants had on average  $28.90 \pm 0.74$  leaves. Maximum N levels encourage more vegetative growth, which attracts *A. gossypii*; later, natural enemies appear on the crop and help to control pests. Maximum levels of N fertilizer help an *A. gossypii* population to develop, but also indirectly cause an increase in the predator population. In this way, a sufficient doze of N fertilizer at a specific time allows more plant growth with limited pest population growth.

**Key words:** Fertilizer effect, pest, predator, plant growth.

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### INTRODUCTION

Plant dry matter is commonly composed of approximately 40% carbon and 2% nitrogen (Beevers and Hageman, 1980). Nitrogen fertilizer is essential throughout the growth season for the development of the structural and non-structural (enzymes, amino acids, protein, chlorophyll, etc.) components of a plant (Tisdale *et al.*, 1993). Nitrogen, potassium, and phosphorus are basic nutrients for growth and their rational application enhances the crop yield (Uhart and Andrade, 1995). Nitrogen (N) helps to improve the root systems, making the plant more vigorous; however, if the N level is too high it will cause excessive vegetative growth and delay maturity in the plant (Johnson *et al.*, 2005).

While the sufficient use of nitrogen is necessary to meet the crop’s needs, a balance is needed to encourage vegetative growth and enhance yield (Johnson *et al.*, 2005). Irrational use of nitrogen not only causes wastage of resources but also gives the plant more greenery, which leads to pest problems that can reduce the yield, causing economic losses (Setamou *et al.*, 1993; 1995). A balanced diet of carbohydrates and nitrogen improves insect longevity and fecundity (Joern and Behmer, 1997). High aphid populations will occur when high levels of nitrogen are applied to the crop, whereas nitrogen and phosphorous in combination can suppress an aphid attack (Khattak *et al.*, 1996).

Sucking insect pests such as the aphids feed on sucrose from phloem and much of the sucrose is secreted as a dew of concentrated sugars called honeydew, which contains glucose, sucrose, fructose, and trehalulose (Hendrix and Wei, 1994). The infestation rate of *A. gossypii* increased as the nitrogen level increased, however, the infestation of *A. gossypii* caused the plant to wilt and suppressed its growth (Setyaningrum *et al.*, 2018.) Aphid population increased as nitrogen level increased, high dose of nitrogen application as 450.00 mg N/plant gives the higher incidence of aphids (478.13 aphids/plant) compared to 225 mg N/plant (Megaladevi *et al.*, 2018). Furthermore, when honeydew falls on leaves, it encourages the growth of sooty mold fungi, which interrupts the photosynthetic process (Yee *et al.*, 1996).

While we know that fertilizers affect plant physiology, and insect pest occurrence and abundance, there are still some gaps in our understanding of the impact of fertilizers on crops and pest populations. Insects such as aphids feed only on sucrose from phloem in plants (Tarczynski *et al.*, 1992). The present study aimed to investigate the effect of different levels of nitrogen on plant leaf growth, and populations of *Aphis gossypii* and *Coccinella septempunctata*. The results from our study will help to control pest populations without sacrificing the plant's nutrition.

## MATERIAL AND METHODS

The study was conducted as an outdoor pot trial on the Experimental Farm of Huazhong Agricultural University. The average altitude of the plot was 23.3 meters (76 feet), latitude 30.5931° N and longitude 114.3054° E, 15 m MSL, at 30 ± 5 °C and 60% ± 15 RH. Seeds of cotton cultivar "Huamian 3109" (*Gossypium hirsutum* L.) were sown in round plastic pots filled with 33kg of soil on 14 June 2014, 3-5 seeds per pot, 20 pots for each treatment. Germination was completed within 5 days of sowing. Five different fertilizer doses were applied: 120, 150, 180, 210 and 240 kg N/ha, at the first flowering stage of crop. Pots were irrigated whenever was necessary. The rest crop managements such as hoeing and weeding were carried out based on the local practice.

### Data recording

Observations of the numbers of insect pests, predators, and leaves per plant were commenced 35 days after sowing (DAS) and carried out at weekly intervals. The crop was observed for 8 weeks and during each recording time, 10 randomly selected plants from each treatment were observed. Six leaves from each plant were observed for *A. gossypii* (2 from the top, the middle, and the bottom, respectively). The number of green leaves per plant was recorded. *C. septempunctata* was observed in 10 sites per treatment and each site consisted of 3 plants.

### Data analysis

The mean numbers of *A. gossypii*, *C. septempunctata*, and leaves per plant at different N levels were compared using ANOVA and Tukey's post-hoc tests. All statistical analyses were conducted using SPSS version 20, at a significance level of  $P < 0.05$ . Figures were drawn using Sigma plot version 10.0.

## RESULTS

Natural enemies in the populations of predators showed significant differences with different N levels at the 2<sup>nd</sup>, 3<sup>rd</sup>, 4<sup>th</sup>, 5<sup>th</sup>, and 8<sup>th</sup> recording time, while no significant difference was observed at the rest three times. Both *A. gossypii* populations and number of leaves in different treatment pots showed significant differences at the 2<sup>nd</sup>, 3<sup>rd</sup>, 4<sup>th</sup>, 5<sup>th</sup>, 6<sup>th</sup>, and 7<sup>th</sup> observations, with no significant difference at the rest two.

*C. septempunctata* population size at different N levels was increasing with prey and N levels increased. The maximum numbers of *C. septempunctata* ( $5.50 \pm 0.53/\text{site}$ ) was recorded at the 5<sup>th</sup> recording time with the maximum N dose (240 kg/ha) treatment, followed by 210, 180, 150, and 120 kg/ha with mean numbers  $4.10 \pm 0.70$ ,  $3.90 \pm 0.81$ ,  $3.60 \pm 0.25$ , and  $2.70 \pm 0.34$ , respectively (Fig. 1).

As for the treatment of 120 kg Nitrogen/ha, the lowest N levels, the minimum populations of *A. gossypii* were observed. At this N level, the maximum *A. gossypii* numbers of  $9.80 \pm 0.94/\text{plant}$  were found on the 5<sup>th</sup> observation a few days after fertilizer application (Fig. 2) when the plants had an average of  $25.8 \pm 0.88$  leaves (Fig. 3).

As for 150 kg Nitrogen/ha, *A. gossypii* infestation was observed during all recording times. The minimum adult population size of  $1.90 \pm 0.39/\text{plant}$  was observed in first recording period (Fig. 2) when the plants had  $9.40 \pm 0.67$  leaves (Fig. 3). The maximum numbers of *A. gossypii* ( $10.40 \pm 0.58 / \text{plant}$ ) were recorded at the 5<sup>th</sup> recording time (Fig. 3).

As for 180 kg Nitrogen/ha, the medium N levels, plants were infested with *A. gossypii* at all recording times. The highest infestation was recorded during the 5<sup>th</sup> observation, with populations of  $13.90 \pm 1.07$  individuals/plant

(Fig. 2), and  $26.20 \pm 0.95$  leaves/plant (Fig. 3). The minimum number of *A. gossypii* ( $2.00 \pm 0.37$  /plant) was recorded in the first week after N application when the plants had the minimum number of leaves.

As for 210 kg Nitrogen/ha, as the N level increased, the number of leaves also increased, as did the size of the *A. gossypii* population. The maximum numbers of *A. gossypii* were recorded at the 5<sup>th</sup> observation time, with an average of  $18.10 \pm 1.08$  individuals/plant (Fig. 2), and  $26.30 \pm 0.75$  leaves/plant (Fig. 3). The minimum insect populations in this treatment group were  $2.1 \pm 0.26$  individuals/plant and  $10.30 \pm 0.85$  leaves/plant were observed at the 1<sup>st</sup> recording time.

And as for 240 kg Nitrogen/ha, the maximum N level produced the most greenery plants, the maximum number of leaves per plants, and the maximum number of *A. gossypii* adults per plant. The peak population of *A. gossypii* was  $29.10 \pm 1.86$  adults/plant, which was observed at the 5<sup>th</sup> recording time (Fig. 2) when the plants had an average of  $28.90 \pm 0.74$  leaves (Fig. 3). The minimum pest numbers of  $2.80 \pm 0.32$  individuals/plant were observed at the 1<sup>st</sup> recording time, with  $9.90 \pm 0.72$  leaves per plant.

Correlation values of *A. gossypii*, natural enemies and number of leaves on plants are presented in Table 1. The result shows that there are positive and negative relationships between correlation values of *A. gossypii*, natural enemies and number of leaves on plants. When the *C. septempunctata* population increased then *A. gossypii* population decreases and when the *A. gossypii* population were increased the then the leaves growth were slow.

Table 1. Correlation of *A. gossypii*, *C. septempunctata* and number of leaves on plants at different recording times

Obs	Treatment 1			Treatment 2			Treatment 3			Treatment 4			Treatment 5		
	Aph	Lbb	Lea	Aph	Lbb	Lea	Aph	Lbb	Lea	Aph	Lbb	Lea	Aph	Lbb	Lea
1	1.7	1.1	9.5	1.9	1.2	9.4	2	1.2	10.2	2.1	1.5	10.3	2.8	1.4	9.9
2	1.4	1.3	11.2	2.0	1.4	11.4	2.1	1.8	13.9	2.7	2.3	12.5	4.6	1.8	15.6
3	2.2	1.5	13.7	3.5	1.8	14.5	3.7	2.2	16.4	4.1	2.8	14.3	4.5	3.3	17.7
4	1.6	2.1	16.2	1.8	2.3	17.3	3.5	2.7	20.2	4.2	2.8	17.9	5.7	3.8	19.9
5	9.8	2.7	25.8	10.4	3.6	25.4	13.9	3.9	26.2	18.1	4.1	26.3	29.1	5.5	28.9
6	2.0	1.5	30.2	6.0	1.7	29.7	6.2	1.8	32.6	7.3	2.4	33.3	11.8	2.4	34.3
7	2.1	1.2	33.8	4.4	1.6	34.1	4.5	1.4	34.8	5.3	1.6	35.7	8.5	1.8	36.9
8	1.7	1.3	54.9	3.0	1.3	54.6	3.4	1.7	56.4	4.4	2.7	56.4	5.1	2.9	57.8

Obs= observation, Aph= Aphid, Lbb= Lady bird beetle, Lea= leaves

Data are the mean of 10 replicated plants, correlation is significant at  $P < 0.05$  value

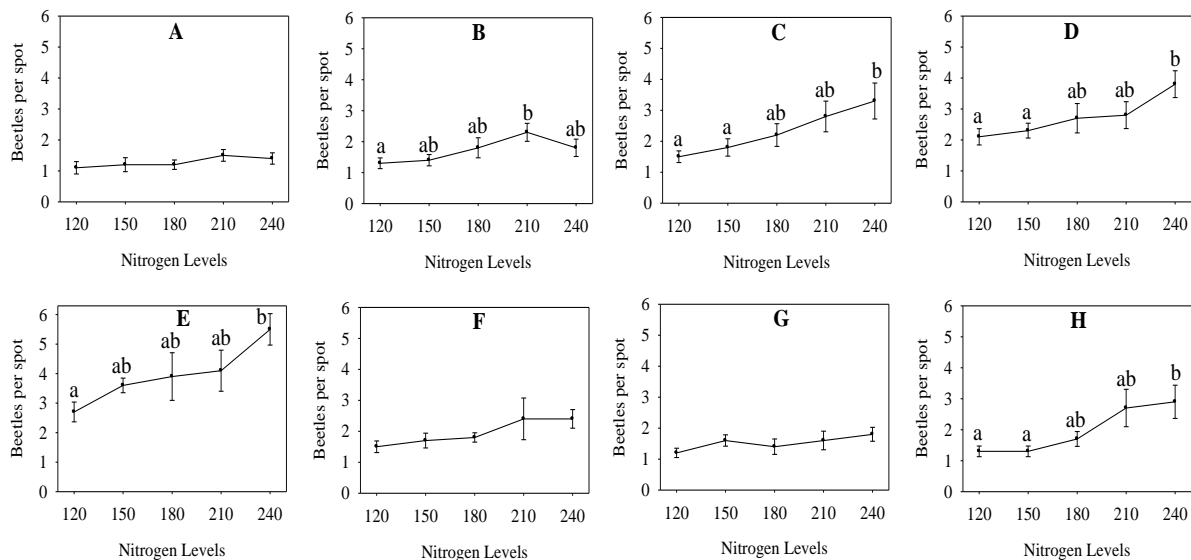


Fig. 1. Effects of nitrogen levels on *C. septempunctata* population. Values are means of 10 replicated plants. A: 1<sup>st</sup>, B: 2<sup>nd</sup>, C: 3<sup>rd</sup>, D: 4<sup>th</sup>, E: 5<sup>th</sup>, F: 6<sup>th</sup>, G: 7<sup>th</sup> and H: 8<sup>th</sup> observations.

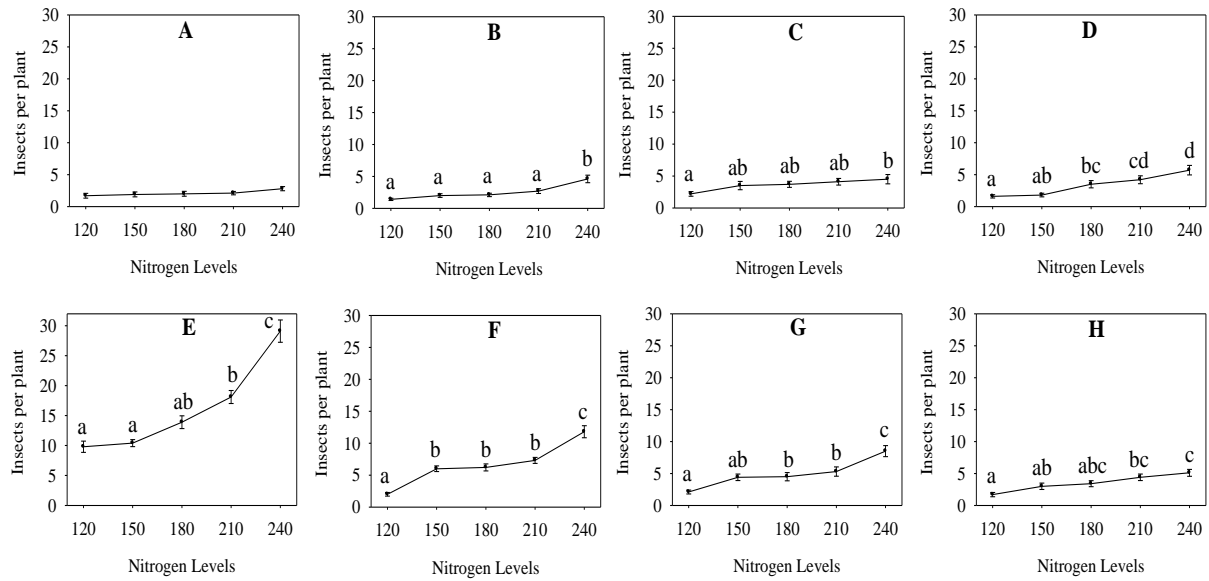


Fig. 2. Effects of nitrogen levels on *A. gossypii* population. Values are means of 10 replicated plants. A: 1<sup>st</sup>, B: 2<sup>nd</sup>, C: 3<sup>rd</sup>, D: 4<sup>th</sup>, E: 5<sup>th</sup>, F: 6<sup>th</sup>, G: 7<sup>th</sup> and H: 8<sup>th</sup> observations.

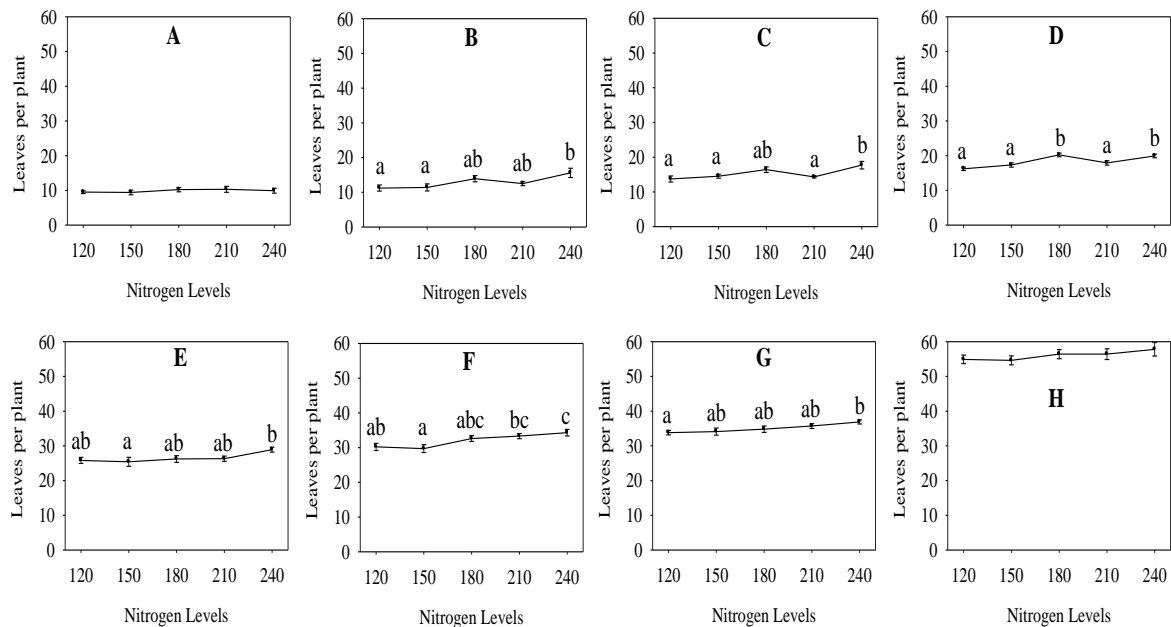


Fig. 3. Effects of nitrogen levels on leaf number per plant. Values are means of 10 replicated plants. A: 1<sup>st</sup>, B: 2<sup>nd</sup>, C: 3<sup>rd</sup>, D: 4<sup>th</sup>, E: 5<sup>th</sup>, F: 6<sup>th</sup>, G: 7<sup>th</sup> and H: 8<sup>th</sup> observations.

## DISCUSSION

The results from our study showed that if the N level is increased, the plant has more leaves and more greenery. Nitrogen helps develop a plant's physical structure and gives a green color to the plant, which attracts insect pests. Our results concur with those of previous report (Sohail *et al.*, 2007) that maximum nitrogenous fertilizer application gave more greenery to plants and delayed the maturity of a crop. Braham *et al.* (2023) received the positive response of Nitrogen fertilizer for aphids, highest Nitrogen dose increase the aphid populations by 3 times compared to the control. Nitrogen application helped aphid growth, high nitrogen application (8.1g/kg) significantly

increased the population of aphids (Guo *et al.*, 2024). In addition, more greenery attracts more insect pests, especially phytophagous sucking insects.

Nitrogen is important for crop growth, but high nitrogen fertilizer urea applied to a crop can cause high levels of insect pests. It is better to apply fertilizers in split application when it is necessary for plant growth (Ramzan *et al.*, 1992). When the nitrogen in the soil is increasing, nitrogen in the leaves will also be increasing (Marks and Clarke, 1995). There is a specific level of N at which a plant can develop physical structure and a limited number of insects can attack. We found that if the crop is supplied with 180kg N/ha, an optimum amount of plant growth is observed and the minimum number of *A. gossypii* is found on the plants.

Predator populations were significantly correlated with *A. gossypii* populations: low numbers of *C. septempunctata* was observed with low numbers of pests. There was no positive nor negative effect of N on *C. septempunctata* community structure. Natural enemies of insect pests occur when the pest populations appear on the crop. There is a direct correlation between pest and predator populations and levels of nitrogen applied to an area, since nitrogen may have an indirect effect on pests through predators (Siemann, 1998). Our result showed the relationship between leaf and *A. gossypii*, natural enemies and *A. gossypii* were negative, so if the one factor increased then another factor decreased. However our results showed positive correlation between natural enemies and leaf numbers.

## CONCLUSIONS

Maximum N levels encourage more vegetative growth, which attracts *A. gossypii*; later, natural enemies appear on the crop and help to control pests. Maximum levels of N fertilizer help an *A. gossypii* population to develop, but also indirectly cause an increase in the predator population. In this way, a sufficient dose of N fertilizer at a specific time allows more plant growth with limited pest population growth.

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