

CHANGES IN PHOTOSYNTHETIC PIGMENT OF WHEAT (*TRITICUM AESTIVUM* L. VAR. BARI GOM-25) AS AFFECTED BY NAA AND N-FERTILIZER

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Key words: Photosynthetic pigment, wheat, NAA, N-fertilizer

Abstract

A field experiment was conducted to investigate the changes in photosynthetic pigment of BARI Gom-25 as affected by foliar spray of NAA and N-fertilizers. The experiment consisted of seven different concentrations of NAA (NAA₀ = 0, NAA₁ = 10, NAA₂ = 20, NAA₃ = 30, NAA₄ = 40, NAA₅ = 50 and NAA₆ = 60 ppm) and five N-levels (F₀=0, F₁=25, F₂=50, F₃=75 and F₄=100% of the recommended dose) and their 35 combination treatments. Results revealed that foliar application of 40 ppm NAA (NAA₄) produced higher chlorophyll a content at all stages (tillering, flowering and grain filling) and were significantly higher than control at tillering and flowering stages. Chlorophyll b and carotenoid contents did not show any general trend to NAA at any stage except at tillering stage. Application of 50% of the recommended N-fertilizer (F₂) produced significantly maximum photosynthetic pigment contents at tillering stage whereas, at flowering stage the same results were obtained from 100% of the recommended N-fertilizers (F₄) but with few exceptions. At grain filling stage, photosynthetic pigment did not show any general trend to N-fertilizer application. Findings on combined effects indicated that 20 and 30 ppm NAA in combination with 50% N-fertilizer (NAA₂F₂ and NAA₃F₂) had produced significantly maximum chlorophyll a content at tillering stage. However, at flowering stage the highest chlorophyll a content was noted from NAA₂F₄ treatment (20 ppm NAA in combination with 100% N-fertilizer). Findings also revealed that combined application of NAA and N-fertilizer had not resulted any general trend on chlorophyll b and carotenoid contents in most of the cases where the maximum value in photosynthetic pigment was found depending on the concentrations of NAA, fertilizer dose and plant growth stages.

Introduction

Fertilization has a strong effect on productivity, nutritional quality management and the regulation of harvest formation processes⁽¹⁾. Nitrogen (N) fertilizers plays key role in agriculture and stimulating a lot of essential processes in plants. It is an important constituent of chlorophyll molecules which enables the plant to transfer energy from

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sunlight by photosynthesis. But excessive use of fertilizer is proven to cause a number of environmental and ecological problems within and outside of farmlands *viz.* air pollution, soil acidification and degradation, water eutrophication, crop yield reduction etc.⁽²⁻³⁾. In addition, the amount of nitrogen applied to plants must be carefully managed to ensure that N will be available throughout the growing season and the vegetative and reproductive development will be not restricted⁽⁴⁾. However, reports regarding the effects of N fertilizer on growth, yield, grain quality and nitrogen use efficiency are available from all over the world⁽⁵⁻⁶⁾ but are lacking in relation to pigment contents⁽⁷⁾.

On the other hand, the use of plant growth regulators especially Naphthalene acetic acid (NAA) has played dramatic role in reducing the demand of N fertilizers and producing improved results in wheat⁽⁸⁻⁹⁾. Previous investigations have also shown that application of optimum concentrations of NAA produced better stimulations in various cereal crops including wheat⁽¹⁰⁻¹¹⁾. Thus, an attempt was taken to investigate the changes in photosynthetic pigment of wheat var. BARI Gom-25 as affected by NAA in combination with various N-levels.

Materials and Methods

A field experiment was conducted at the botanical garden of the Department of Botany, University of Dhaka. The experiment was laid out in randomized block design with four replications. The total area of the experimental field was 98.56 m² (17.60 m × 5.60 m) and was prepared conventionally. Nitrogen, phosphorus and potassium content of experimental soil were determined by Micro-Kjeldahl's method⁽¹²⁾, ascorbic acid blue colour method⁽¹³⁾ and flame photometer⁽¹⁴⁾, respectively. The initial properties of soil revealed low amount of nitrogen, very high amount of phosphorus and very low amount of potassium. Chemical fertilizers and cow-dung (4 ton/ha) were applied at doses recommended by Fertilizer Recommendation Guide 2012⁽¹⁵⁾. Two-thirds of urea and full of the other fertilizers were applied as basal dose during final land preparation. The remaining urea was applied immediately after the first irrigation (18 days). A modern high yielding variety of wheat var. BARI Gom-25 was selected for this experiment. Seeds were collected from BARI, Joydebpur, Gazipur and were sterilized with 0.5% calcium hypochlorite before sowing. Seeds were sown on November 15, in lines 20 cm apart maintaining plant to plant distance of 10 cm. Cultural practices *viz.*, thinning, irrigation, weeding etc. were done following Hand Book of Agricultural Technology⁽¹⁶⁾.

The trial consisted of seven different treatments of NAA *viz.* NAA₀ = foliar spray of distilled water only and NAA₁, NAA₂, NAA₃, NAA₄, NAA₅ and NAA₆ represent foliar spray of 10, 20, 30, 40, 50 and 60 ppm NAA, respectively. There were also five levels of N-fertilizers treatments *viz.* F₀ = without application of urea and F₁, F₂, F₃, F₄ = 25, 50, 75 and 100% of the recommended dose of urea. Considering seven foliar NAA and five levels of N-levels, this experiment comprised of 35 combination treatments. Foliar spray

was done at the age of 30 days. Photosynthetic pigment viz. Chlorophyll a (Chl. a), Chlorophyll b (Chl. b) and total carotenoid contents of leaves were determined at tillering, flowering and grain filling stages. Fresh leaves were collected randomly from eight plants (2 from each replication) for each treatment at all stages. The analysis of pigments was determined spectrophotometrically. The amount of Chl. a and b were determined by using specific absorption co-efficient of McKinney⁽¹⁷⁾ and the formulae of Maclachalan and Zalik⁽¹⁸⁾. The amount of carotenoid were determined by the equation of von Wettstein⁽¹⁹⁾. Data were analyzed statistically and treatment means were compared by LSD test at 5% level of significance⁽²⁰⁾.

Results and Discussion

Application of NAA and N-fertilizer as well as their combined effect had significant influences upon photosynthetic pigment contents of leaves at different stages but with few exceptions.

Photosynthetic pigment contents of leaves were positively influenced by NAA application at tillering stage except chl. b content due to NAA₁, NAA₂ and NAA₃ treatments (Table 1). At this stage, all treatments resulted significantly higher Chl. a content except due to NAA₁ treatment and the maximum value (0.15 mg/g) was recorded from NAA₄ treatment. The range of increases were 12.50 to 175% whereas, the only decrease due to NAA₁ was 50%. However, application of NAA treatments had no significant effect on Chl. b content. Significantly higher amount of carotenoid content of leaves were recorded from all NAA treatments where, the highest increase due to NAA₆ treatment was 66.42% followed by NAA₄ treatment (63.75%). At flowering stage, significantly higher Chl. a content of leaves was obtained from NAA₄ and NAA₆ treatments where the maximum increase due to NAA₄ was 25% over the control. At this stage, Chl. b and carotenoid content obtained from NAA₅ treatment although higher than control but statistically identical to each other. Application of NAA₄ treatment although resulted higher Chl. a and carotenoid content at grain filling stage but statistically not different from control. At this stage significantly maximum carotenoid content was obtained from NAA₄ treatment (6.61 mg/g).

Similarly, Singh and Gill (1985)⁽²¹⁾ reported the stimulating effects of NAA on chlorophyll contents of leaves in wheat and barley. Depending on the concentrations of NAA both increase and decrease in photosynthetic pigment were obtained in maize⁽²²⁾. Results of this experiment are also in accord with the findings of Jahan and Adam in two varieties of rice⁽²³⁾.

Results presented in Table 1 showed that application of various doses of N-fertilizers had significant responses on pigment contents of leaves at different stages. At the tillering stage, all the photosynthetic pigment contents were recorded maximum following F₂ treatment whereas, minimum due to F₁. Treatment F₂ resulting maximum

Chl. a and carotenoid were significantly different from the rest of the treatments. In case of Chl. b content although the maximum and minimum were also recorded from F₂ and F₁ treatments, respectively, where both are significantly different from each other but statistically similar to control.

Table 1. Effects of NAA and N-fertilizer on photosynthetic pigment (mg/g) of leaves of BARI Gom-25 at three different stages.

Treatments	Tillering stage			Flowering stage			Grain filling stage		
	Chl. a	Chl. b	Carotenoids	Chl. a	Chl. b	Carotenoids	Chl. a	Chl. b	Carotenoids
NAA									
NAA ₀	0.08 c	0.09 ab	4.11 f	0.20 b	0.07 ab	6.70 a	0.19 ab	0.07 a	6.05 b
NAA ₁	0.03 d	0.06 b	4.18 e	0.19 b	0.06 bc	4.76 c	0.18 bc	0.06 a	5.81 b
NAA ₂	0.13 ab	0.07 b	5.88 c	0.19 b	0.05 c	5.52 b	0.16 d	0.07 a	5.22 c
NAA ₃	0.11 b	0.07 b	5.04 d	0.20 b	0.06 bc	6.36 a	0.19 ab	0.07 a	5.96 b
NAA ₄	0.15 a	0.09 ab	6.73 ab	0.25 a	0.06 bc	6.39 a	0.20 a	0.07 a	6.09 b
NAA ₅	0.13 ab	0.11 ab	6.51 b	0.20 b	0.08 a	6.78 a	0.19 ab	0.07 a	6.61 a
NAA ₆	0.13 ab	0.13 a	6.84 a	0.24 a	0.06 bc	6.64 a	0.17 cd	0.04 a	5.37 c
N-Fertilizer									
F ₀	0.08 c	0.10 ab	4.55 d	0.21 c	0.07 a	6.87 b	0.15 c	0.08 ab	5.67 b
F ₁	0.04 d	0.05 b	4.47 d	0.02 d	0.04 b	3.37 c	0.13 d	0.09 a	6.13 a
F ₂	0.22 a	0.12 a	7.79 a	0.25 b	0.07 a	6.72 b	0.20 b	0.05 b	5.79 b
F ₃	0.12 b	0.10 ab	5.92 b	0.24 b	0.07 a	6.41 b	0.22 a	0.06 ab	5.96 ab
F ₄	0.09 c	0.08 ab	5.32 c	0.33 a	0.07 a	7.44 a	0.20 b	0.05 b	5.65 b
CV (%)	10.21	9.57	14.05	18.03	17.96	12.50	19.63	14.44	9.32
LSD(0.05)	0.02	0.05	0.32	0.01	0.01	0.48	0.01	0.03	0.33

Mean in a vertical column followed by same letter or without letter do not differ significantly at 5 % level.

At flowering stage, significantly higher Chl. a (57.14%) and carotenoid (8.30%) were obtained from F₄ treatment whereas, Chl. b content was affected with similar statistical values. Results revealed that F₁ treatment produced significantly lowest photosynthetic pigment at this stage.

At grain filling stage, significantly higher amount of Chl. a was recorded from F₂, F₃ and F₄ treatments and increased by 33.33, 46.67 and 33.33%, respectively. Different doses of N-fertilizer resulted higher carotenoid content of leaves where, F₁ treatment produced 8.11% higher value over the control. However, the maximum Chl. b was obtained from F₁ and the minimum from F₂ and both these are significantly different from each other but statistically at par with the control.

Present findings indicated that photosynthetic pigment are very sensitive to nitrogen levels and 50% of the recommended dose (F₂) produced maximum photosynthetic pigments at tillering stage whereas at flowering stage the maximum was noted from

100% of the recommended dose (F_4) treatment. But no general trend was found in photosynthetic pigment contents at grain filling stage. Although reports regarding the effect of N-levels on photosynthetic pigments are scanty. But, findings of Tranavičienė *et al.*⁽⁷⁾ revealed that total photosynthetic pigment contents increased with plant age and were higher at higher fertilization rates.

Pigment contents of leaves were both significantly and non-significantly influenced by NAA at varying N-levels (Table 2). At tillering stage, Chl. a content of leaves was obtained maximum (0.38 mg/g) following both NAA_2F_2 and NAA_3F_2 treatments and were significantly different from rest of the treatments. The least amount of Chl. a (0.01 mg/g) was recorded from the leaves of NAA_0F_1 and NAA_3F_4 treatments and were significantly different from control. Combined treatments had retarding effect on Chl. b content of leaves although NAA_6F_2 and NAA_6F_3 treatments produced higher values but statistical identical to control. At this stage, carotenoid content of leaves were significantly higher following all treatments where the maximum increase due to NAA_2F_2 treatment was 383.08% followed by NAA_4F_3 treatment (380%).

Table 2. Combined effects of NAA and N-fertilizer on photosynthetic pigment (mg/g) of leaves of BARI Gom-25 at three different stages.

Treatments (NAA × F)	Tillering stage			Flowering stage			Grain filling stage		
	Chl. a	Chl. b	Carotenoid	Chl. a	Chl. b	Carotenoid	Chl. a	Chl. b	Carotenoid
NAA_0F_0	0.09 e	0.16 a-d	1.95 p	0.22 ij	0.08 bc	7.95 c-e	0.21 ef	0.12 a	7.30 bc
NAA_1F_0	0.08 ef	0.11 d-g	4.40 jk	0.21 j	0.06 de	6.43 lm	0.08 op	0.11 a	4.83 n
NAA_2F_0	0.18 d	0.12 d-f	6.80 e	0.06 o	0.02 hi	2.87 u	0.07 p	0.06 cd	3.30 q
NAA_3F_0	0.08 ef	0.12 d-f	5.50 g	0.13 lm	0.07 cd	6.49 l	0.21 ef	0.11 a	7.57 b
NAA_4F_0	0.05 gh	0.11 d-g	5.95 f	0.35 b	0.07 cd	7.85 c-f	0.13 m	0.06 cd	4.98 l-n
NAA_5F_0	0.02 ij	0.03 i	2.62 o	0.14 l	0.12 a	8.01 b-d	0.18 hi	0.06 cd	6.85 d-f
NAA_6F_0	0.02 ij	0.05 hi	4.60 ij	0.34 b	0.08 bc	8.48 ab	0.13 m	0.04 cd	4.86 n
NAA_0F_1	0.01 j	0.03 i	2.76 no	0.04 p	0.12 a	4.92 pq	0.14 lm	0.07 bc	5.86 gh
NAA_1F_1	0.02 ij	0.04 i	4.79 hi	0.02 q	0.05 ef	1.25 w	0.21 ef	0.06 cd	6.80 ef
NAA_2F_1	0.05 gh	0.03 i	4.57 ij	0.01 q	0.01 i	4.78 q	0.09 no	0.10 ab	4.85 n
NAA_3F_1	0.06 fg	0.03 i	4.68 ij	0.02 q	0.03 gh	4.24 r	0.10 n	0.11 a	6.62 f
NAA_4F_1	0.03 h-j	0.07 f-i	4.74 i	0.01 q	0.01 i	2.36 v	0.13 m	0.11 a	5.37 i-k
NAA_5F_1	0.03 h-j	0.07 f-i	5.07 h	0.02 q	0.02 hi	3.40 s	0.13 m	0.13 a	8.09 a
NAA_6F_1	0.09 e	0.07 f-i	4.71 ij	0.02 q	0.02 hi	2.64 uv	0.15 kl	0.04 cd	5.29 j-l
NAA_0F_2	0.06 fg	0.07 f-i	5.56 g	0.29 ef	0.07 cd	7.41 f-i	0.14 lm	0.04 cd	4.42 o
NAA_1F_2	0.02 ij	0.07 f-i	4.18 kl	0.10 n	0.04 fg	3.36 s	0.17 ij	0.04 cd	5.21 j-m

Treatments (NAA × F)	Tillering stage			Flowering stage			Grain filling stage		
	Chl. a	Chl. b	Carotenoid	Chl. a	Chl. b	Carotenoid	Chl. a	Chl. b	Carotenoid
NAA ₂ F ₂	0.38 a	0.10 e-h	9.42 a	0.19 k	0.06 de	5.51 no	0.20 fg	0.06 cd	4.88 mn
NAA ₃ F ₂	0.38 a	0.11 d-g	9.35 a	0.30 de	0.08 bc	7.63 d-h	0.21 ef	0.04 cd	5.11 k-n
NAA ₄ F ₂	0.31 b	0.11 d-g	8.83 b	0.28 fg	0.08 bc	7.06 i-k	0.24 bc	0.06 cd	7.13 c-e
NAA ₅ F ₂	0.17 d	0.16 a-d	8.42 c	0.30 de	0.09 b	8.30 a-c	0.25 b	0.06 cd	7.16 cd
NAA ₆ F ₂	0.25 c	0.19 ab	8.79 b	0.28 fg	0.09 b	7.76 d-g	0.22 de	0.05 cd	6.61 f
NAA ₀ F ₃	0.05 gh	0.07 f-i	3.59 m	0.12 m	0.04 fg	5.31 op	0.24 bc	0.06 cd	6.12 g
NAA ₁ F ₃	0.04 g-i	0.06 g-i	3.47 m	0.31 cd	0.06 de	4.88 pq	0.21 ef	0.05 cd	5.47 ij
NAA ₂ F ₃	0.04 g-i	0.06 g-i	4.00 l	0.23 i	0.06 de	5.95 mn	0.24 bc	0.06 cd	6.17 g
NAA ₃ F ₃	0.04 g-i	0.07 f-i	2.99 n	0.26 h	0.07 cd	6.82 j-l	0.24 bc	0.05 cd	6.15 g
NAA ₄ F ₃	0.31 b	0.14 b-e	9.36 a	0.31 cd	0.07 cd	7.47 e-i	0.27 a	0.07 bc	6.78 f
NAA ₅ F ₃	0.27 c	0.11 d-g	8.74 bc	0.22 ij	0.09 b	7.17 h-j	0.16 jk	0.04 cd	4.88 mn
NAA ₆ F ₃	0.08 ef	0.20 a	9.29 a	0.27 gh	0.07 cd	7.31 g-i	0.21 ef	0.06 cd	6.12 g
NAA ₀ F ₄	0.18 d	0.11 d-e	6.66 e	0.30 de	0.03 gh	7.91 c-e	0.21 ef	0.05 cd	6.54 f
NAA ₁ F ₄	0.02 ij	0.03 i	4.07 l	0.32 c	0.09 b	7.89 c-f	0.25 b	0.05 cd	6.76 f
NAA ₂ F ₄	0.02 ij	0.03 i	4.62 ij	0.45 a	0.11 a	8.52 a	0.19 gh	0.06 cd	5.67 hi
NAA ₃ F ₄	0.01 j	0.02 i	2.66 o	0.30 de	0.07 cd	6.60 kl	0.17 ij	0.05 cd	4.34 o
NAA ₄ F ₄	0.03 h-j	0.02 i	4.76 hi	0.32 c	0.07 cd	7.18 h-j	0.22 de	0.05 cd	6.18 g
NAA ₅ F ₄	0.17 d	0.18 a-c	7.69 d	0.31 cd	0.08 bc	7.02 i-k	0.23 cd	0.07 bc	6.09 g
NAA ₆ F ₄	0.19 d	0.13 c-e	6.81 e	0.30 de	0.06 de	7.00 i-k	0.14	0.03 d	3.95 p
CV (%)	10.21	9.57	14.05	18.03	17.96	12.50	19.63	14.44	9.32
LSD (0.05)	0.02	0.05	0.32	0.01	0.01	0.48	0.01	0.03	0.33

Mean in a vertical column followed by same letter or without letter do not differ significantly at 5 % level.

At flowering stage, the maximum increase in Chl. a content due to NAA₂F₄ treatment was 104.55% over the control followed by 59.09% (NAA₄F₀) and 55.55% (NAA₆F₀), respectively (Table 2). The maximum amount of Chl. b content of leaves was observed from two treatments, NAA₅F₀ and NAA₀F₁. Increases in Chl. b content due to NAA₅F₀, NAA₀F₁ and NAA₂F₄ treatments were 50.0, 50.0 and 37.50% over the control respectively. Significantly maximum carotenoid content of leaves was observed from NAA₂F₄ treatment which was 7.17% higher over the control followed by 6.67% (NAA₆F₀), 4.40% (NAA₅F₂) and 0.75% (NAA₅F₀) treatments respectively.

At grain filling stage, only a few combined treatments (NAA₄F₂, NAA₅F₂, NAA₀F₃, NAA₂F₃, NAA₃F₃, NAA₄F₃, NAA₁F₄, NAA₅F₄) had significant beneficial effect on Chl. a

content of leaves. But in case of Chl. b content of leaves it was affected almost negatively having an exception due to NAA₅F₁ treatment. Significantly maximum (8.09 mg/g) carotenoid content of leaves at grain filling stage was due to NAA₅F₁ treatment where it was 10.82% higher over the control. However, the highest decrease in carotenoid content was 54.79% due to NAA₂F₀ treatment. Islam 2015⁽²⁴⁾ obtained significantly higher Chl. a and Chl. b content of leaves of BARI Gom-26 following NAA in combination with various N-levels at flowering and grain filling stages but with few exceptions. However, combination of NAA and N-fertilizer had not resulted any significant difference in carotenoid content of leaves. Application of NAA at varying N-level produced negative influences in pigment content of leaves at tassel initiation stage and both stimulatory and retarding effects at grain filling stage of maize⁽²⁵⁾.

Conclusion

In plants, utilization of solar energy mainly depends on the relative amount and efficiency of the photosynthetic pigments. Chlorophyll content of leaf is an important factor for increasing or decreasing photosynthesis of any crop plant. It was evident that application of NAA, N-fertilizer and their combined treatments had both stimulatory and retarding effects on pigment content of leaves at different stages of BARI Gom-25. Findings also revealed that combined application of NAA and N-fertilizer had not produced any general trend on leaf pigments and the maximum value in photosynthetic pigment was obtained depending on the concentrations of NAA, fertilizer dose and plant growth stages.

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