

## MORPHO-PHYSIOLOGICAL AND YIELD RESPONSES OF SOYBEAN TO FOLIAR SPRAYS OF CHITOSAN AND IBA

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### ABSTRACT

An experiment was conducted during the *kharif* season of 2016-2017 at farm of Botany section, College of Agriculture, Nagpur, to study the effect of foliar sprays of chitosan and IBA on morpho-physiological parameters and yield of soybean cv. JS-335. The experiment was laid out in randomized block design with eleven treatments and three replications. The different concentrations of chitosan and indole-3-butyric acid (25, 50, 75, 100 and 125 ppm) was applied at 30 DAS. Foliar sprays of 25 ppm chitosan and 100 ppm IBA significantly enhanced plant height, number of branches, leaf area, dry matter, RGR, NAR, yield ha<sup>-1</sup> and harvest index when compared with control and rest of the treatments under study. Considering the Benefit:Cost ratio 25 ppm chitosan was found more economical having B:C ratio of 2.75 as compared to 2.19 in control.

(Key words: Soybean, chitosan, IBA, foliar sprays, morpho-physiological parameters, yield)

### INTRODUCTION

Soybean (*Glycine max* L.) is a diploid species having chromosome number 2n=40. It belongs to family "Leguminosae" and subfamily "Papilionoidae". It is annual leguminous herbaceous plant. Soybean is also known as "Gold of Soil" due to its various qualities such as ease in cultivation, less requirement of fertilizers and labour resulting in high cost: benefit ratio. Soybean being a legume crop is gifted naturally to fix atmospheric nitrogen in the root nodules with the help of *Rhizobium* (Anonymous, 1985).

This crop in fact has made revolution in the agricultural economy with its immense potential, quality as food, feed, numerous industrial product commodity. It is soil erosion resistant crop and suited for most of the soil.

Among oilseeds soybean ranks fifth in the world. The important soybean growing countries in world are America, Brazil, Argentina, China and India. The largest soybean producing state in India are Madhya Pradesh, Maharashtra and Rajasthan. Area under soybean during 2015-2016 in India is 109.71 million ha with the production of 114.91 million tonnes and productivity of 1047 kg ha<sup>-1</sup>.

Chitosan is polysaccharide and composed of 2-deoxy-2-(acetylamino) glucose unit (N-acetyl glucosamine). Chitosan is a linear polysaccharide composed of randomly distributed  $\beta$ -(1-4)- linked D-glucosamine (deacetylated unit) and N-acetyl-glucosamine (acetylated unit) combined by 1,4 glycosidic linkages, forming a long chain linear polymer and due to this NH<sub>2</sub> group chitosan is more versatile in properties. Chitosan produced as processing waste from

shellfish krill, oyster squid and fungi. Primarily, chitosan is used for plant defence (Walkar, 2004) and yield increase. In agriculture, chitosan is typically used as a natural seed treatment and plant growth enhancer. It is one of the most abundant biodegradable (Gooday, 1990) materials in the world.

IBA is a plant growth regulator, used to promote and accelerate root formation of plant clippings and to reduce transplant shock of non-food ornamental nursery stock. IBA is also used on fruit and vegetable crops, field crops and ornamental turf to promote growth and development of flowers and fruit and to increase crop yields. IBA has been classified as a biochemical pesticide because it is similar in structure and function to the naturally occurring plant growth hormone indole-3-acetic acid.

Considering the above facts present investigation was undertaken to study the responses of chitosan and IBA on morphophysiological parameters and yield of soybean.

### MATERIALS AND METHODS

Experiment was laid out in randomized block design with eleven treatments and three replications. Plot size of individual treatment was gross 2.10 m x 2.20 m and net 1.50 m x 2.00 m. Seeds were sown at the rate of 75 kg ha<sup>-1</sup> by dibbling method at spacing of 30 cm x 10 cm on 11<sup>th</sup> July, 2016. Treatments comprised of control (T<sub>1</sub>), 25 ppm chitosan (T<sub>2</sub>), 50 ppm chitosan (T<sub>3</sub>), 75 ppm chitosan (T<sub>4</sub>), 100 ppm chitosan (T<sub>5</sub>), 125 ppm chitosan (T<sub>6</sub>), 25 ppm IBA (T<sub>7</sub>), 50

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ppm IBA ( $T_8$ ), 75 ppm IBA ( $T_9$ ), 100 ppm IBA ( $T_{10}$ ) and 125 ppm IBA ( $T_{11}$ ). The foliar application of chitosan and IBA was given at 30 DAS on soybean. Observations on plant height, number of branches, leaf area plant<sup>-1</sup>, total dry weight of plant, NAR, RGR were recorded at 30, 45, 60 and 75 DAS and seed yield ha<sup>-1</sup> were also recorded after harvesting. Per cent increase and B:C ratio were also calculated. The crop was kept free from disease and pest during the growth period. Harvesting was undertaken after the crop attained maturity. Data were analysed by statistical method suggested by Panse and Sukhatme (1954).

## RESULTS AND DISCUSSION

### Plant height

Plant height is an important measure of plant growth. It is one of the visible measurements and is a function of internodes and leaf emergence. Since, leaves are born on stem leaf area development and biomass production shows a close relationship with plant height.

Data regarding plant height were recorded at 30, 45, 60, and 75 DAS. The data are presented in table 1. At 30 DAS the data regarding plant height was found non significant, because foliar sprays of chitosan and IBA were given from this stage onwards.

At 45 DAS significantly highest plant height was recorded in treatment  $T_2$  (25 ppm chitosan) among all treatments when compared with treatment  $T_1$  (control) and remaining treatments under study. Also, treatments  $T_{10}$  (100 ppm IBA),  $T_3$  (50 ppm chitosan),  $T_9$  (75 ppm IBA),  $T_4$  (75 ppm chitosan) and  $T_{11}$  (125 ppm IBA) were found significantly superior over treatment  $T_1$  (control). Remaining treatments were found at par with control ( $T_1$ ).

At 60 DAS significantly maximum plant height was recorded in treatment  $T_2$  (25 ppm chitosan) followed by the treatments  $T_{10}$  (100 ppm IBA) and  $T_3$  (50 ppm chitosan) when compared with treatment  $T_1$  (control) and remaining treatments under study. Rest of the treatments were found at par with control ( $T_1$ ).

At 75 DAS significantly maximum plant height was recorded in treatment  $T_2$  (25 ppm chitosan) followed by treatments  $T_{10}$  (100 ppm IBA) and  $T_3$  (50 ppm chitosan) when compared with treatment  $T_1$  (control) and remaining treatments under study. Rest of the treatments were found at par with control ( $T_1$ ).

The per cent increase in plant height with respect to foliar application of 25 ppm chitosan ( $T_2$ ) were 19.25 at 45 DAS, 11.80 at 60 DAS and 11.88 at 75 DAS when compared with control ( $T_1$ ).

Chitosan increased the growth character viz., plant height might be due to increased number of internodes or length of internodes because of increased cell number (Hong Yan and Shu Yu, 2001). Chibu *et al.* (2002) in rice reported that application of chitosan at early stages increased plant height.

IBA stimulate dry mass production through enhancement of cell division and chlorophyll accumulation and in turn reflected on the increase in vegetative growth (Amin *et al.*, 2007). This might be the reason for increase in plant height in the present investigation.

Amin *et al.* (2007) tested different concentrations of indole-3-butyric acid (25, 50, and 100 mg l<sup>-1</sup>) and salicylic acid (50, 100 and 200 mg l<sup>-1</sup>) and reported that foliar application of indole-3-butyric acid and salicylic acid at 100 mg l<sup>-1</sup> significantly increased plant height.

Mondal *et al.* (2013,a) tried different concentrations of chitosan (0, 50, 75, 100 and 125 ppm) sprayed three times at 35, 50 and 65 DAS. They suggested that foliar application of chitosan at 100 ppm significantly increased plant height of maize.

### Number of branches plant<sup>-1</sup>

Number of branches plant<sup>-1</sup> is one of the main parameter which contributes to high yield. Since, leaves are born on stems, leaf area development and biomass production shows a close relationship with plant height and number of branches.

The data regarding number of branches plant<sup>-1</sup> as influenced by foliar sprays of chitosan and IBA are presented in table 1. Data regarding number of branches were recorded at 45, 60 and 75 DAS found statistically significant.

At 45 DAS significantly maximum number of branches were recorded in treatment  $T_2$  (25 ppm chitosan) followed by treatments  $T_{10}$  (100 ppm IBA),  $T_3$  (50 ppm chitosan) and  $T_9$  (75 ppm IBA) in a descending manner when compared with treatment  $T_1$  (control) and remaining treatments under study. Also, treatments  $T_4$  (75 ppm chitosan),  $T_{11}$  (125 ppm IBA) and  $T_5$  (100 ppm chitosan) were found significantly superior over treatment  $T_1$  (control). Remaining treatments were found at par with control ( $T_1$ ).

At 60 DAS significantly maximum number of branches were recorded in treatment  $T_2$  (25 ppm chitosan) followed by treatments  $T_{10}$  (100 ppm IBA),  $T_3$  (50 ppm chitosan),  $T_9$  (75 ppm IBA) and  $T_4$  (75 ppm chitosan) in a descending manner when compared with  $T_1$  (control) and remaining treatments under study.

At 75 DAS significantly maximum number of branches were recorded in treatment  $T_2$  (25 ppm chitosan) followed by the treatments  $T_{10}$  (100 ppm IBA),  $T_3$  (50 ppm chitosan),  $T_9$  (75 ppm IBA) and  $T_4$  (75 ppm chitosan) in a descending manner when compared with  $T_1$  (control) and remaining treatments under study. Treatments  $T_{11}$  (125 ppm IBA),  $T_5$  (100 ppm chitosan) and  $T_8$  (50 ppm IBA) also gave significantly more number of primary branches over  $T_1$  (control).

Chitosan increased the growth character viz., number of branches might be due to increased number of internodes or length of internodes because of increased cell number (Hong Yan and Shu Yu, 2001). Chibu *et al.* (2000) reported that there was an increase in number of branches for chitosan application in soybean.

IBA stimulate dry mass production through enhancement of cell division and chlorophyll accumulation and in turn reflected on the increase in vegetative growth (Amin *et al.*, 2007). This might be the reason for increase in number of branches in the present investigation.

A field experiment conducted by Farouk and Amany (2012) for improving the effect of chitosan on cowpea and suggested that foliar application of chitosan @ 250 ppm significantly enhanced number of branches.

Amin *et al.* (2013) revealed the response of chickpea (*Cicer arietinum* L.) to treatment with two plant growth regulators putrescine and Indole-3-butyric acid (IBA) @ 25, 50 and 100 mg l<sup>-1</sup> applied either alone or in combinations. Spraying of putrescine and IBA @ 100 mg l<sup>-1</sup> significantly increased the number of branches over control.

### Leaf area

Leaf area play a key role in absorption of radiation in the deposition of photosynthesis during the diurnal and seasonal cycle and in the pathways and rates of biogeochemical cycling within the canopy soil systems. Leaf area depends on the number and size of the leaves and hence, the total leaf area is important parameter for assessing the ability of plants to synthesis its dry matter. The photosynthesis capacity of plants is a function of leaf area development.

Data on leaf area plant<sup>-1</sup> were recorded at four stages viz., 30, 45, 60 and 75 DAS are furnished in table 1. Leaf area recorded at 45, 60 and 75 DAS gave significant results.

At 30 DAS the data regarding leaf area was found non significant, because foliar sprays of chitosan and IBA were given from this stage onwards.

At 45 DAS significantly maximum leaf area was noticed in treatment T<sub>2</sub> (25 ppm chitosan) followed by the treatments T<sub>10</sub> (100 ppm IBA), T<sub>3</sub> (50 ppm chitosan), T<sub>9</sub> (75 ppm IBA) and T<sub>4</sub> (75 ppm chitosan) when compared with treatment T<sub>1</sub> (control) and remaining treatments under study. While, treatment T<sub>11</sub> (125 ppm IBA) was found significantly superior over treatment T<sub>1</sub> (control). Remaining treatments were found at par with control (T<sub>1</sub>). The range of leaf area at 45 DAS was 4.95 – 8.02 dm<sup>2</sup>. The per cent increase in leaf area by treatment T<sub>2</sub> (25 ppm chitosan) over treatment T<sub>1</sub> (control) was 62.02.

At 60 DAS significantly maximum leaf area was noticed in treatment T<sub>2</sub> (25 ppm chitosan) followed by the treatments T<sub>10</sub> (100 ppm IBA) and T<sub>3</sub> (50 ppm chitosan) when compared with treatment T<sub>1</sub> (control) and rest of the treatments under study. Treatments T<sub>9</sub> (75 ppm IBA) was also produced significantly more leaf area over treatment T<sub>1</sub> (control). The range of leaf area at 60 DAS was 9.93 – 13.01 dm<sup>2</sup>. The per cent increase in leaf area by treatment T<sub>2</sub> (25 ppm chitosan) over treatment T<sub>1</sub> (control) was 31.02.

At 75 DAS significantly maximum leaf area was recorded in treatment T<sub>2</sub> (25 ppm chitosan) followed by treatments T<sub>10</sub> (100 ppm IBA) and T<sub>3</sub> (50 ppm chitosan) in a descending manner when compared with treatment T<sub>1</sub>

(control) and remaining treatments under study. Treatment T<sub>9</sub> (75 ppm IBA) was found significantly superior over treatment T<sub>1</sub> (control) and rest of the treatments. The range of leaf area at 75 DAS was 9.87 - 12.89 dm<sup>2</sup>. The per cent increase in leaf area by treatment T<sub>2</sub> (25 ppm chitosan) over treatment T<sub>1</sub> (control) was 30.59.

Data showed that leaf area increased from 30 to 60 DAS. But at 75 DAS leaf area decreased. It might be due to leaf fall at this stage.

Chibu and Shibayama (2001) indicate the existence of the higher chlorophyll content in the plant treated with chitosan. This contributed into the increase of the photosynthesis production and leaf area. The application of chitosan increased key enzymes activities of nitrogen metabolism and improved the transportation of nitrogen in the functional leave which enhanced leaf area. Similarly IBA stimulates dry matter production through enhancement of cell division and chlorophyll accumulation and in turn reflects on the increase in vegetative growth (Amin *et al.*, 2007). These might be the reasons for increase in leaf area in the present investigation.

Bideshki *et al.* (2013) conducted a field experiment to study the impact of 0 and 100 ppm indole-3-butyric acid (IBA) and 0, 0.1 and 0.5 mM salicylic acid (SA) on garlic and reported that 0.5 mM salicylic acid and 100 ppm IBA significantly enhanced leaf area.

Rabbi *et al.* (2016) examined different concentrations of chitosan viz., 0 (control), 25, 50, 75 and 100 ppm at 30 and 40 DAS on mungbean plant. Results showed that foliar application of chitosan @ 50 ppm significantly enhanced plant leaf area over control.

### Total dry weight

Total dry matter accumulation is one of the factors that determines economic yield in crop species where the seed is of economic importance. Leaf is the major organ where most of photosynthates are produced. The number of leaves and their arrangement on the main stem and side branches determine the structure of crop canopy which ultimately decides the dry matter production at each growth stage and its partitioning to reproductive organs during pre-flowering to maturity period has immense importance in determining the final productivity.

Data on dry weight plant were recorded at the four growth stages i.e. 30, 45, 60 and 75 DAS are presented in table 1. Data on dry matter showed significant variation at 45, 60 and 75 DAS. At 30 DAS the data regarding dry matter was found non significant, because foliar sprays of chitosan and IBA at different concentrations were given from this stage onwards.

At 45 DAS significantly maximum dry matter was recorded in treatment T<sub>2</sub> (25 ppm chitosan) followed by treatments T<sub>10</sub> (100 ppm IBA) and T<sub>3</sub> (50 ppm chitosan) in a descending manner when compared with treatment T<sub>1</sub> (control) and remaining treatments under study. Also, treatments T<sub>9</sub> (75 ppm IBA), T<sub>4</sub> (75 ppm chitosan) and T<sub>11</sub>

(125 ppm IBA) were found significantly superior over treatment  $T_1$  (control). Treatments,  $T_5$  (100 ppm chitosan),  $T_8$  (50 ppm IBA),  $T_6$  (125 ppm chitosan) and  $T_7$  (25 ppm IBA) were found at par with treatment  $T_1$  (control).

At 60 DAS significantly maximum dry matter was noticed in treatment  $T_2$  (25 ppm chitosan) followed by the treatments  $T_{10}$  (100 ppm IBA),  $T_3$  (50 ppm chitosan) and  $T_9$  (75 ppm chitosan) when compared with treatment  $T_1$  (control) and remaining treatments under study. Also, treatments  $T_4$  (75 ppm chitosan) and  $T_{11}$  (125 ppm IBA), were found significantly superior over treatment  $T_1$  (control) and rest of the treatments.

At 75 DAS significantly maximum dry matter was observed in treatment  $T_2$  (25 ppm chitosan) followed by the treatments  $T_{10}$  (100 ppm IBA) and  $T_3$  (50 ppm chitosan) and  $T_9$  (75 ppm chitosan) when compared with treatment  $T_1$  (control) and remaining treatments under study. Also, treatments  $T_4$  (75 ppm chitosan) and  $T_{11}$  (125 ppm IBA), were found significantly superior over treatment  $T_1$  (control).

Significant increase in dry matter from 45-75 DAS might be due to increase in the leaf area and photosynthetic capacity.

Increase in dry matter production by the application of IBA might be due to enhancement of cell division and chlorophyll accumulation (Amin *et al.*, 2007). Similarly the stimulating effect of chitosan on plant growth viz., dry matter may be attributed to an increase in the availability and uptake of water and essential nutrients through adjusting cell osmotic pressure and reducing the accumulation of harmful free radicals by increasing antioxidants and enzyme activities (Guan *et al.*, 2009). Higher area of leaves and chlorophyll content has contributed into the increase of the photosynthesis production which reflects a significant amount of dry weight (Chibu and Shibayama, 2001). These might be the reasons for increase in dry matter in the present investigation.

Shraiy and Hegazi (2009) studied the effect of acetylsalicylic acid (ASA) @ 10 and 20 ppm, indole-3-butyric acid (IBA) @ 50 and 100 ppm and gibberellic acid (GA) @ 50 and 100 ppm on pea (*Pisum sativum* L.). Application of ASA and IBA at 25 and 35 DAS significantly enhanced dry weight.

Mondal *et al.* (2013,b) carried out field experiment to investigate the effect of chitosan on mungbean plant. Five concentrations of chitosan viz., 0, 25, 50, 75 and 100 ppm were applied twice at 25 and 35 DAS. Results showed that 50 ppm chitosan enhanced dry weight of plant.

### Growth analysis

Growth analysis is one of the measures for accessing the seed yield of plant. The physiological basis of yield difference can be measured through an evaluation of difference in growth parameters and their impact on yield. The productivity of crop may be related with the parameters such as RGR, NAR and partitioning of total photosynthate into economic and non-economic sink. The analyzed data of RGR and NAR are presented in table 1.

### Relative growth rate (RGR)

The highest rate of RGR indicates the ability of maximum dry matter for development. The increment in RGR might be associated with maximum leaf area expansion and growth of stem and root. Increment in NAR is related with the increase in total dry weight of plant unit<sup>-1</sup> of leaf area.

Considering all the treatments under study, significantly maximum RGR was recorded in treatment with 25 ppm chitosan ( $T_2$ ) i.e., 0.0805 g g<sup>-1</sup> day<sup>-1</sup> at 30-45 DAS, 0.0412 g g<sup>-1</sup> day<sup>-1</sup> at 45-60 DAS and 0.0220 g g<sup>-1</sup> day<sup>-1</sup> at 60-75 DAS respectively. But it was lowest in control ( $T_1$ ) i.e., 0.0468 g g<sup>-1</sup> day<sup>-1</sup> at 30-45 DAS, 0.0333 g g<sup>-1</sup> day<sup>-1</sup> at 45-60 DAS and 0.0173 g g<sup>-1</sup> day<sup>-1</sup> at 60-75 DAS respectively.

At first stage i.e. 30-45 DAS range of RGR recorded was 0.0468-0.0805 g g<sup>-1</sup> day<sup>-1</sup>. Significantly maximum RGR was observed in treatment  $T_2$  (25 ppm chitosan) followed by treatment  $T_{10}$  (100 ppm IBA) and  $T_3$  (50 ppm chitosan) when compared with treatment  $T_1$  (control) and remaining treatments under study. Also, treatments  $T_9$  (75 ppm IBA),  $T_4$  (75 ppm chitosan),  $T_{11}$  (125 ppm IBA) and  $T_5$  (100 ppm chitosan), were found significantly superior over treatment  $T_1$  (control) and rest of the treatments.

At second stage i.e. 45-60 DAS range of RGR noticed was 0.0333-0.0414 g g<sup>-1</sup> day<sup>-1</sup>. Significantly maximum RGR was observed in treatment  $T_2$  (25 ppm chitosan) followed by treatments  $T_{10}$  (100 ppm IBA),  $T_3$  (50 ppm chitosan),  $T_9$  (75 ppm IBA),  $T_4$  (75 ppm chitosan),  $T_{11}$  (125 ppm IBA) and  $T_5$  (100 ppm chitosan) when compared with treatment  $T_1$  (control) and remaining treatments under study.

At third stage i.e. 60-75 DAS range of RGR recorded was 0.0173-0.0220 g g<sup>-1</sup> day<sup>-1</sup>. Significantly maximum RGR was observed in treatment  $T_2$  (25 ppm chitosan) followed by treatment  $T_{10}$  (100 ppm IBA),  $T_3$  (50 ppm chitosan),  $T_9$  (75 ppm IBA),  $T_4$  (75 ppm chitosan) and  $T_{11}$  (125 ppm IBA) when compared with treatment  $T_1$  (control) and remaining treatments under study. Also, treatment  $T_5$  (100 ppm chitosan) was found significantly superior over treatment  $T_1$  (control) and rest of the treatments.

Mondal *et al.* (2012) examined different concentrations of chitosan (0, 50, 75, 100 and 125 ppm) on okra. Spraying of chitosan @ 100 or 125 ppm at 25, 40 and 55 DAS significantly increased relative growth rate.

Wagh (2015) conducted a field experiment to investigate the impact of putrescine and IBA @ 0, 50, 75, 100, 125 and 150 ppm sprayed at 30 and 45 DAS on soybean. He reported that two foliar sprays of putrescine and IBA @ 100 ppm significantly increased relative growth rate.

### Net assimilation rate (NAR)

NAR is the rate of increasing the dry weight of a plant unit<sup>-1</sup> of active growing material. NAR is any attribute of the plant which is primarily concerned in carbon assimilation and thus has some claims to be taken as a measure of the internal factor for growth. NAR is closely associated with photosynthesis efficiency of leaves, but it

is not a pure measure of photosynthesis. NAR depends upon the excess dry matter gained, over the loss in respiration. It is increase in plants dry weight unit<sup>-1</sup> area of assimilation tissues unit<sup>-1</sup> time.

Considering all the treatments under study, significantly maximum NAR was recorded in treatment with 25 ppm chitosan (T<sub>2</sub>) i.e. 0.1499 g dm<sup>-2</sup> day<sup>-1</sup> at 30-45 DAS, 0.0715 g dm<sup>-2</sup> day<sup>-1</sup> at 45-60 DAS and 0.0482 g dm<sup>-2</sup> day<sup>-1</sup> at 60-75 DAS respectively. But it was lowest in control (T<sub>1</sub>) i.e. 0.0798 g dm<sup>-2</sup> day<sup>-1</sup> at 30-45 DAS, 0.0419 g dm<sup>-2</sup> day<sup>-1</sup> at 45-60 DAS and 0.0228 g dm<sup>-2</sup> day<sup>-1</sup> at 60-75 DAS respectively.

At first stage i.e. 30-45 DAS range of NAR recorded was 0.0798-0.1499 g dm<sup>-2</sup> day<sup>-1</sup>. Significantly maximum NAR was observed in treatment T<sub>2</sub> (25 ppm chitosan) followed by treatments T<sub>10</sub> (100 ppm IBA) and T<sub>3</sub> (50 ppm chitosan) when compared with treatment T<sub>1</sub> (control) and remaining treatments under study. Also, treatments T<sub>9</sub> (75 ppm IBA) and T<sub>4</sub> (75 ppm chitosan) were found significantly superior over treatment T<sub>1</sub> (control) and rest of the treatments.

At second stage i.e. 45-60 DAS range of NAR recorded was 0.0419-0.0715 g dm<sup>-2</sup> day<sup>-1</sup>. Significantly maximum NAR was observed in treatment T<sub>2</sub> (25 ppm chitosan) followed by treatments T<sub>10</sub> (100 ppm IBA), T<sub>3</sub> (50 ppm chitosan), T<sub>9</sub> (75 ppm IBA), T<sub>4</sub> (75 ppm chitosan) and T<sub>11</sub> (125 ppm IBA) when compared with treatment T<sub>1</sub> (control) and remaining treatments under study.

At third stage i.e. 60-75 DAS range of NAR recorded was 0.0228-0.0482 g dm<sup>-2</sup> day<sup>-1</sup>. Significantly maximum NAR was observed in treatment T<sub>2</sub> (25 ppm chitosan) followed by treatments T<sub>10</sub> (100 ppm IBA), T<sub>3</sub> (50 ppm chitosan), T<sub>9</sub> (75 ppm IBA), T<sub>4</sub> (75 ppm chitosan) and T<sub>11</sub> (125 ppm IBA) when compared with treatment T<sub>1</sub> (control) and remaining treatments under study. Also, treatment T<sub>5</sub> (100 ppm chitosan) was found significantly superior over treatment T<sub>1</sub> (control) and rest of the treatments.

Wagh (2015) carried out field experiment to investigate the effect of putrescine and IBA @ 0, 50, 75, 100, 125 and 150 ppm sprayed at 30 and 45 DAS on soybean. He observed that two foliar sprays of putrescine and IBA @ 100 ppm significantly increased net assimilation rate.

### Seed yield ha<sup>-1</sup>

Seed yield is a complex physiological character which is the sum total of all metabolic activities taking place in plant body. This includes various morphological aspects like increase in plant height, leaf size, leaf area, number of branches, dry matter, seed weight etc. These characters can be considered as yield contributing parameters.

Significantly maximum seed yield hectare<sup>-1</sup> was recorded in treatment T<sub>2</sub> (25 ppm chitosan) followed by the treatments T<sub>10</sub> (100 ppm IBA), T<sub>3</sub> (50 ppm chitosan), T<sub>9</sub> (75 ppm IBA) and T<sub>4</sub> (75 ppm chitosan) when compared with control and remaining treatments under study. While, treatments T<sub>11</sub> (125 ppm IBA), T<sub>5</sub> (100 ppm chitosan), T<sub>8</sub> (50 ppm IBA), T<sub>6</sub> (125 ppm chitosan) and T<sub>7</sub> (25 ppm IBA) were found at par with treatment T<sub>1</sub> (control).

Plant growth regulators are known to enhance the source-sink relationship and stimulate the translocation of photo-assimilates thereby helping in effective flower formation, fruit and seed development and ultimately enhance productivity of the crops. Growth regulators can improve the physiological efficiency including photosynthetic ability and enhance the effective partitioning of accumulates from source and sink in the field crops (Solamani *et al.*, 2001).

Bittelli *et al.* (2001) reported that foliar application of chitosan decreased transpiration in pepper plants and reduce water use while maintaining biomass production and yield. The significant effect of chitosan on plant growth and yield may be attributed to an increase in the key enzyme activities of nitrogen metabolism and increased photosynthesis which enhanced plant growth (Gornik *et al.*, 2008; Mondal *et al.*, 2012). The increase in cowpea yield due to chitosan application may be due to its effects in stimulating physiological processes, improving vegetative growth, followed by active translocation of photoassimilates from source to sink tissue (Sharifa and Abu-Munjefah 2013). Chitosan when externally supplied was observed to increase crop growth and ultimately the yield. It improves the nutritional status of plant system. Chitosan increases the absorption and translocation of nutrients in plant and ultimately influences yield.

Growth regulator IBA is proved to improve effective partitioning and translocation of accumulates from source to sink in the field crops. The plant growth regulators also increase mobilization of reserve food materials to the developing sink through increases in hydrolyzing and oxidizing enzyme activities and lead to yield increases. IBA increases the ability of cell division in meristematic zones of plant and hence, the ability of plant to absorb nutritive material which finally lead to the increase of grain yield (Ghodrat *et al.*, 2012).

The increase in the yield recorded in this investigation could be a reflection of the effect of growth regulators on growth and development, it might be due to marked increase in plant height, leaf area, dry weight, number of branches which gave a chance to the plant to carry more seeds and marked increase in the photosynthetic pigments content which could lead to increase in photosynthesis, resulting in greater transfer of assimilates to the seeds and causing increase in their weight.

Amin *et al.* (2013) studied the effect of two plant growth regulators putrescine and Indole-3-butyric acid (IBA) @ 25, 50 and 100 mg l<sup>-1</sup> applied either alone or in combinations. Spraying of putrescine and IBA @ 100 mg l<sup>-1</sup> significantly increased seed yield of chickpea (*Cicer arietinum* L.).

Rabbi *et al.* (2016) formulated an experiment to study the effect of chitosan (0, 25, 50, 75 and 100 ppm) on mungbean sprayed at 30 and 40 DAS. Results showed that application of chitosan @ 50 ppm significantly enhanced seed yield.

**Table 1. Effect of chitosan and indole-3-butyric acid on plant height, number of branches, leaf area and total dry weight**

Treatments	Plant height (cm)						Number of branches						Leaf area (dm <sup>2</sup> )						Total dry weight (g)														
	30		45		75		30		45		75		30		45		60		75		30		45		60		75						
	DAS	DAS	DAS	DAS	DAS	DAS	DAS	DAS	DAS	DAS	DAS	DAS	DAS	DAS	DAS	DAS	DAS	DAS	DAS	DAS	DAS	DAS	DAS	DAS	DAS	DAS	DAS	DAS					
T <sub>1</sub> (Control)	17.33	29.77	44.30	48.62	2.42	2.68	2.92	3.04	1.54	4.95	9.93	9.87	2.43	6.93	12.73	17.33	17.47	35.50	49.53	54.40	2.48	4.22	4.48	4.66	1.62	8.02	13.01	12.89	2.83	12.83	22.90	32.28	
T <sub>2</sub> (25 ppm Chitosan)	16.50	33.60	47.86	53.00	2.55	3.79	4.13	4.34	1.72	7.50	12.14	11.95	2.63	11.00	20.63	28.47	18.00	32.30	46.93	51.96	2.45	3.57	3.98	4.19	1.65	6.77	11.03	10.94	3.07	10.13	19.74	26.50	
T <sub>3</sub> (50 ppm Chitosan)	17.30	31.57	46.12	50.57	2.76	3.42	3.62	3.95	1.58	6.33	10.66	10.55	2.80	9.23	18.53	25.23	T <sub>4</sub> (75 ppm Chitosan)	17.67	30.80	45.43	49.87	2.62	3.19	3.47	3.67	1.63	5.97	10.25	10.13	2.63	8.73	15.96	24.61
T <sub>5</sub> (100 ppm Chitosan)	17.53	30.03	44.26	49.04	2.68	3.02	3.38	3.55	1.66	5.47	10.04	9.96	2.73	7.86	13.73	19.23	T <sub>6</sub> (125 ppm Chitosan)	18.23	31.23	45.77	50.27	2.53	3.28	3.54	3.84	1.71	6.12	10.44	10.36	2.53	9.06	18.30	23.64
T <sub>7</sub> (25 ppm IBA)	16.97	32.53	47.33	52.53	2.38	3.66	4.12	4.28	1.62	7.08	11.37	11.23	2.87	10.32	20.12	28.00	T <sub>8</sub> (50 ppm IBA)	17.40	33.80	48.90	53.63	2.72	3.96	4.32	4.48	1.66	7.72	12.56	12.43	2.60	11.76	21.68	29.12
T <sub>9</sub> (75 ppm IBA)	18.30	31.60	46.13	51.40	2.66	3.50	3.78	4.03	1.60	6.58	10.88	10.78	2.83	9.27	19.53	25.50	T <sub>10</sub> (100 ppm IBA)	0.412	0.566	0.925	1.196	0.148	0.189	0.257	0.196	0.834	0.474	0.320	0.252	0.236	0.742	1.107	2.151
T <sub>11</sub> (125 ppm IBA)	-	1.670	2.729	3.529	-	0.558	0.758	0.580	-	1.398	0.944	0.742	-	2.188	3.266	6.347	SE (m±)																
CD at 5%																																	

**Table 2. Effect of chitosan and indole-3-butyric acid on RGR, NAR, seed yield ha<sup>-1</sup>, harvest index and B:C ratio**

Treatments	RGR						NAR						Seed yield ha <sup>-1</sup> (q)	Harvest index (%)	B:C ratio
	30-45 DAS		45-60 DAS		60-75 DAS		30-45 DAS		45-60 DAS		60-75 DAS				
	DAS	DAS	DAS	DAS	DAS	DAS	DAS	DAS	DAS	DAS	DAS	DAS			
T <sub>1</sub> (control)	0.0468	0.0333	0.0173	0.0798	0.0419	0.0228	23.56	27.12	2.19						
T <sub>2</sub> (25 ppm Chitosan)	0.0805	0.0414	0.0220	0.1499	0.0715	0.0482	30.10	41.88	2.75						
T <sub>3</sub> (50 ppm Chitosan)	0.0739	0.0410	0.0217	0.1251	0.0648	0.0433	29.26	36.57	2.63						
T <sub>4</sub> (75 ppm Chitosan)	0.0607	0.0405	0.0210	0.1114	0.0639	0.0414	28.36	33.52	2.52						
T <sub>5</sub> (100 ppm Chitosan)	0.0592	0.0392	0.0199	0.1037	0.0590	0.0365	26.67	31.28	2.33						
T <sub>6</sub> (125 ppm Chitosan)	0.0585	0.0359	0.0180	0.1016	0.0524	0.0304	25.84	29.66	2.23						
T <sub>7</sub> (25 ppm IBA)	0.0496	0.0344	0.0175	0.0851	0.0471	0.0258	25.26	28.63	2.27						
T <sub>8</sub> (50 ppm IBA)	0.0591	0.0389	0.0190	0.1026	0.0589	0.0342	26.13	29.93	2.27						
T <sub>9</sub> (75 ppm IBA)	0.0653	0.0407	0.0213	0.1161	0.0640	0.0423	28.83	35.46	2.42						
T <sub>10</sub> (100 ppm IBA)	0.0789	0.0411	0.0219	0.1379	0.0664	0.0450	29.56	37.49	2.41						
T <sub>11</sub> (125 ppm IBA)	0.0601	0.0401	0.0206	0.1052	0.0601	0.0377	27.60	32.81	2.18						
SE (m±)	0.00420	0.00185	0.00060	0.00841	0.00580	0.00386	1.199	1.839	-						
CD at 5%	0.01238	0.00546	0.00176	0.02482	0.01712	0.01138	3.536	5.426	-						

## Harvest index (HI)

Significantly maximum harvest index was recorded in treatment T<sub>2</sub> (25 ppm chitosan) and minimum in T<sub>1</sub> (control). The range of increased harvest index was 27.12 in control to 41.88 % in above treatments.

Similarly, harvest index was also significantly increased in treatments receiving T<sub>2</sub> (25 ppm chitosan) followed by treatments T<sub>10</sub> (100 ppm IBA) and T<sub>3</sub> (50 ppm chitosan) in a descending manner when compared with treatment T<sub>1</sub> (control) and other remaining treatments under study. Treatments T<sub>9</sub> (75 ppm IBA) and T<sub>4</sub> (125 ppm IBA) also found their significance over treatment T<sub>1</sub> (control). While, treatments T<sub>11</sub> (125 ppm IBA), T<sub>5</sub> (100 ppm chitosan), T<sub>8</sub> (50 ppm IBA), T<sub>6</sub> (125 ppm chitosan) and T<sub>7</sub> (25 ppm IBA) were found at par with T<sub>1</sub> (control).

Harvest index is the proportion of biological yield represented by economic yield. It is the coefficient of effectiveness or migration coefficient. Harvest index reflects the proportion of assimilate distribution between the economic and total biomass (Donald and Hamblin, 1976). Increase in harvest index might be the result of co-ordinated interplay of growth and development characters.

The highest per cent increase in yield over control was observed in treatment sprayed with 25 ppm chitosan i.e. 27.76 %. Next to this treatment foliar spray of 100 ppm IBA also enhanced yield by 25.46 % over control.

From overall results, it can be stated that foliar application of growth regulators such as chitosan and IBA with different concentrations improved the morpho-physiological parameters might have helped in attaining better seed yield in the present investigation. But, considering the B : C ratio foliar application of 25 ppm chitosan (T<sub>2</sub>) was found most effective treatment having B : C ratio of 2.75 as compared to 2.19 in control.

Finally it is inferred that, spraying plants at vegetative stage (30 DAS) with 25 ppm chitosan could be considered as the most suitable time and most suitable concentration to expect promising improvement regarding the growth parameters, physiological characters and yield quantity and quality of soybean.

## REFERENCES

- Amin, A. A., EL-Sh. M. Rashad and H. M. H. EL-Abagy, 2007. Physiological effect of indole - 3 - butyric acid and salicylic acid on growth, yield and chemical constituents of onion plants. *J. Appl. Sci. Res.* **3**(11): 1554-1563.
- Amin, A. A., F.A. Gharib, H.F. Abouziena and Mona G. Dawood, 2013. Role of indole-3-butyric acid or/and putrescine in improving productivity of Chickpea (*Cicer arietinum* L.) Plants. *Pakistan J. Biol. Sci.* **16**: 1894-1903.
- Anonymous, 1985. Legume inoculants and their use. Nitrogen fixation for Tropical Agriculture Legumes Project (NIFTAL), FAO, Rome.
- Bideshki Abumoslem, Mohammad Javad Arvin and Mehrnoosh Darini, 2013. Interactive effects of Indole- 3-butyric acid (IBA) and salicylic acid (SA) on growth parameters, bulb yield and allicin contents of garlic (*Allium sativum*) under drought stress in field. *Intl. J. Agron. Plant. Prod.* **4**(2): 271-279.
- Bittelli, M., M. Flury, G.S. Campbell, E.J. Nichols, 2001. Reduction of transpiration through foliar application of chitosan. *Agri. and Forest Meteorology.* **107**(3): 167-175.
- Chibu, H., H. Shibayama, M. Mitsutomi and S. Arima, 2000. Effects of chitosan application on growth and chitinase activity in several crops. *Marine Highland Bio sci. Center Rep.,* **12**: 27-35.
- Chibu, H., H. Shibayama, M. Mitsutomi and S. Arima, 2002. Effects of chitosan application on the shoot growth of rice and soybean. *Japanese J. Crop Sci.* **71**: 206-211.
- Chibu, H. and H. Shibayama, 2001. Effects of chitosan application on the growth of several crops. in: T. Urugami, K Kurita and T. Fukamizo (Eds.), *Chitin and chitosan in life science*, Yamaguchi, Japan. pp. 235-239.
- Donald, C.M. and J. Hamblin, 1976. Growth and development in physiology of crop plants, 2<sup>nd</sup> Ed., Scientific publishers Jodhpur, pp.198-199.
- Farouk, S. and A. Ramadan Amany, 2012. Improving growth and yield of cowpea by foliar application of chitosan under water stress. *Egyptian J. Bio.* **14**: 14-26.
- Ghodrat, V., M.J. Roustae and M.S. Tadaion, 2012. Effect of priming with indole-butyric acid (IBA) on germination and growth of wheat under saline conditions. *IJACS.* **4**(6):289-292.
- Gooday, G.W. 1990. Physiology of microbial degradation of chitin and chitosan. *Biodegr.* **1**: 177-190.
- Gornik, K., M. Grzesik and B.R. Duda, 2008. The effect of chitosan on rooting of grape vine cuttings and on subsequent plant growth under drought and temperature stress. *J. Fruit Ornamental Plant Res.* **16**: 333-343.
- Guan, Y.J., J.Hu, X.J. Wang and C.X. Shao, 2009. Seed priming with chitosan improves maize germination and seedling growth in relation to physiological changes under low temperature stress. *J. Zhejiang University Sci B.* **10**(6): 427-433.
- Hong Yan, W. and L. Shu Yu, 2001. The effect of chitosan and sodium alginate on the growth and photosynthesis of soybean. *J. NortheastAgric: Univ. China.* **8**: 156-160.
- Mondal, M.M.A., M.A. Malek, A.B. Puteh, M.R. Ismail, M. Ashrafuzzaman and L. Naher, 2012. Effect of foliar application of chitosan on growth and yield in okra, *Australian J. Crop Sci.* **6**(5): 918-921.
- Mondal, M.M.A., A.B. Puteh, N.C. Dafader, M.Y.Rafii and M.A. Malek, 2013 a. Foliar application of chitosan improves growth and yield in maize. *J. Food Agriculture & Environment.* **11**(2):520-523.
- Mondal, M. M. A., M.A. Malek, A.B. Puteh and M.R. Ismail, 2013 b. Foliar application of chitosan on growth and yield attributes of mungbean [*Vigna radiata* (L.) Wilczek]. *Bangladesh J. Bot.* **42**(1): 179-183.
- Pansee, V.G. and P.V. Sukhamte, 1954. Statistical method for agriculture works, ICAR New Delhi, pp. 107-109.
- Rabbi, F., M. Rahman, M.M.A. Mondal, S.K. Bhowal and A. Haque, 2016. Effect of chitosan application on plant characters, yield attributes and yield of mungbean. *Res. J. Agri. and Environ. Management.* **5**(3): 095-100.
- Shraiy, Amal M. El. and Amira M. Hegazi, 2009. Effect of acetylsalicylic acid, indole-3- butyric acid and gibberellic acid on plant growth and yield of pea (*Pisum Sativum* L.), *Aust.J. Basic and Appl. Sci.* **3**(4): 3514-3523.
- Solamani, A., C. Sivakumar, S. Anbumani, T. Suresh and K. Arumugam, 2001. Role of plant growth regulators on rice production. A review. *Agric. Rev.* **23**: 33-40.
- Sharifa, S. and Abu-Muriefah, 2013. Effect of chitosan on common bean (*Phaseolus vulgaris* L.) plants grown under water stress conditions. *Intl. Res. J. Agric. Sci. and Soil Sci.* **3**(6): 192-199.
- Wagh, Y. A. 2015. Influence of putrescine and indole-3-butyric acid on growth and productivity of soybean. M.Sc. (Agri.) thesis (Unpublished) submitted to Dr. P.D.K.V., Akola.
- Walker, R. 2004. Evaluation of potential chitosan to enhance plant defence. *RIROC.* **(4)**:1-7.