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SEED TREATMENT WITH SALICYLIC ACID IMPROVES GROWTH, BIOCHEMICAL ATTRIBUTES AND QUALITY SEED PRODUCTION OF PEA

Qamar Shahzad Anjum[1,*], Muhammad Amjad[1], Khurram Ziaf[1] and Amer Habib[2]

[1]Vegetable Seed Lab., Institute of Horticultural Sciences, University of Agriculture, Faisalabad, Pakistan;
[2]Seed Pathology Lab., Department of Plant Pathology, University of Agriculture, Faisalabad, Pakistan
[*]Corresponding author's e.mail: qamar2005.uaf@gmail.com

The use of different growth regulators can modulate plant growth and increase yield of various crops. Salicylic acid (SA) has been successfully used as seed treatment in improvement of plant growth and yield. This study was conducted to optimize different concentrations of salicylic acid (0, 50, 100, 150, 200 & 250 mg/liter) for seed treatment of pea cultivars (Meteor & PF-400). Results implied a positive impact of salicylic acid seeds treatment in improving the studied attributes. Seed treatment with 200 mg/L salicylic acid produced maximum root length (11.25 cm), flowers per plant (44.7), average seed yield per plant (66.3 g) in mature plants of cv. Meteor which were significantly different from control *i.e.*, 8.3 cm root length, 23.2 flowers per plant and 30.2 g seed yield per plant of control. Meteor plants from this treatment also maximally accumulated total phenolics (17.4 mg/g F.wt.) and total soluble proteins (3.49 mg/g F.wt.) in leaves which were significantly better than control (8.3 mg/g F.wt. total phenolics and 2.83 mg/g F.wt. total soluble proteins). Similarly, maximum shoot length (64.3 cm), average seed yield per plant (68.2 g), chlorophyll contents (3.53 mg/g F.wt.), carotenoid contents (0.83 mg/g F.wt.), total phenolics (15.6 mg/g F.wt.) and total soluble proteins (3.58 mg/g F.wt.) in leaves of PF-400 plants were recorded in response to seed treatment with 200 mg/L salicylic acid. These results were also statistically significant as compared to lower concentrations of SA and control (shoot length 40.3 cm; average seed yield per plant 26.9 g; chlorophyll contents 2.59 mg/g F.wt.; carotenoid contents 0.56 mg/g F.wt.; total phenolics 8.9 mg/g F.wt. and total soluble proteins 2.69 mg/g F.wt. in control plants). Moreover, seeds harvested from plants of both pea cultivars grown from 200 mg/L SA treatment showed maximum final germination percentage, seedling length and seedling vigor index. Furthermore, treatment of pea seeds with 250 mg/L SA produced, longest main shoot (60.2 cm), maximum number of pods per plant (40.0) of Meteor, while maximum root length (11.2 cm), flowers (48.8) and pods per plant (45.0) of PF-400. It can be concluded that treatment of pea seeds (Meteor & PF-400) with 200 or 250 mg/L salicylic acid can be used as a tool to improve growth of pea plants, seed yield and also quality of the produced seed.
Keywords: *Pisum sativum*, Meteor, PF-400, salicylic acid, seed soaking, seedling vigor index, seed yield
Abbreviations: Salicylic acid (SA); Fresh weight (F.wt.), Indole acetic acid (IAA), Gibberellic acid (GA_3), Reactive oxygen species (ROS), Hectare (ha).

INTRODUCTION

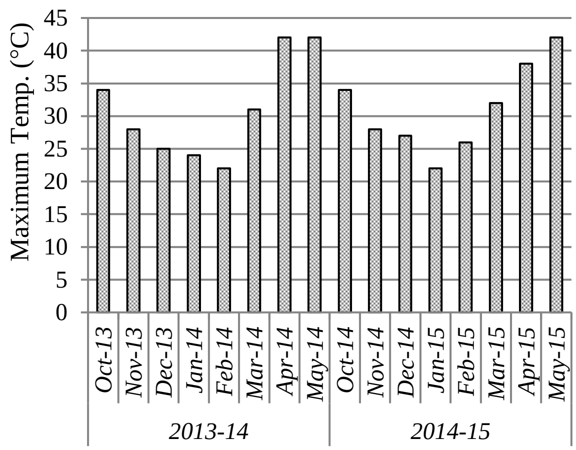
Pea is an important rabi crop in plain areas of Pakistan. However, in hilly areas it is cultivated in spring or summer season to meet the cool temperature requirements (a mean temperature 12 to 20°C). In Pakistan, 1465 thousand tons of peas were harvested from an area of 26569 hectares during the year 2017-18. Punjab province has the highest share regarding area (81.3%) and production (97.7%) for national level. Regarding area under cultivation, share of Sindh, Khyber Pakhtunkhwa and Baluchistan is 7.3, 7.3 and 4.1% respectively. Share of Sindh, Khyber Pakhtunkhwa and Baluchistan for production is 0.5, 1.0 and 0.8%, respectively (MNFSR, 2018). Pea improves soil fertility and reduces the nitrogen requirement of succeeding crop. Average per unit area yield of pea in Pakistan is low in comparison with yield of other countries such as India (Anonymous, 2016), which can be attributed to numerous factors, *viz.*, poor quality seed, insect-pests, pathogens, low fertilizers use efficiency and negligence of farmers for maintaining healthy crop stand.

Salicylic acid [$C_6H_4(OH)COOH$] is a phenolic compound which regulates several plant physiological and developmental processes *i.e.,* photosynthetic activity, stomatal conductance, oxidative phosphorylation, electron transport in mitochondria, conferring innate immunity to plant against oxidative stresses. It plays an important role in flowering and binds the activity of amino-oxyacetic acid (a flowering inhibitor) under stress conditions (Wada *et al.*, 2010). It increases nitrogenase and nitrate reductase activity, thereby improves fertilizer use efficiency (Hayat *et al*., 2012). It plays a regulatory role for flowering with involvement in photoperiod pathways (Martinez *et al.*, 2004) as well as for senescence related genes (Kinoshita *et al.*, 1999; Robatzek and Somssich, 2001; Miao *et al.*, 2004; Schenk *et al.*, 2005). Moreover, it regulates the activity of apoplastic proteins, catalase (CAT), superoxide dismutase (SOD), ascorbate peroxidase (APO), peroxidase (POX), guaiacol peroxidase (GPO), ACC-oxidase and ortho-hydroxycinnamic acid. Furthermore, it also plays a regulatory role in plant growth and development (Rady and Mohamed, 2015). In addition to this, *Phaseolus vulgaris* seed treatment with salicylic acid significantly enhances germination rate, germination percentage, seedling vigor and endogenous level of IAA and GA_3 (Gharib and Hegazi, 2010).

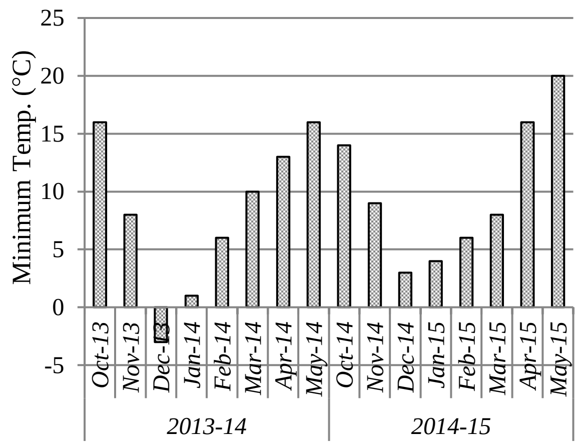
However, little work has been done to explore the impact of salicylic acid seed treatment on growth, seed yield and biochemical attributes of pea. Considering the low productivity challenge and potential of salicylic acid in improving plant growth and productivity, this study was conducted to evaluate its effect as seed treatment on vegetative and reproductive growth in relation to the improvement of biochemical attributes of pea plants as well as quality seed production.

MATERIALS AND METHODS

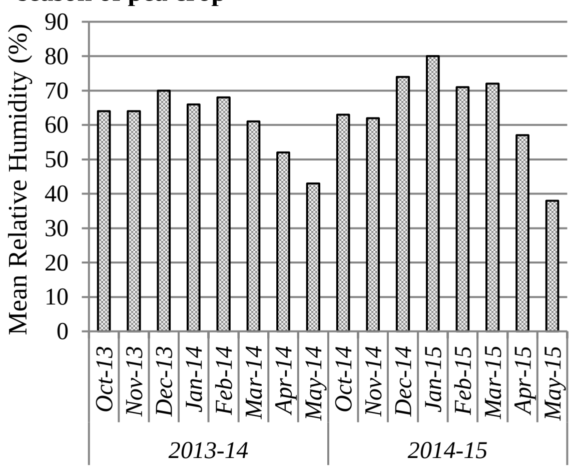
This Experiment was conducted at Vegetable Research Area and Vegetable Seed Laboratory, Institute of Horticultural Sciences as well as Bioassay Section Medicinal Laboratory, Department of Biochemistry, University of Agriculture Faisalabad, Pakistan (31.4336 ºN, 73.0683 ºE). Seeds of two pea varieties (Meteor & PF-400) were obtained from Ayub Agricultural Research Institute (AARI), Faisalabad. Pea seeds, after treatment with salicylic acid (mentioned below) were sown during November, 2013-14 and 2014-15 on well prepared raised beds in field at a distance of 20 cm (plant-plant) x 150 cm (row-row) keeping plant population approximately 13,400 plants/acre. Urea, DAP (Diammonium phosphate) and SOP (Sulphate of potash) were used as sources of NPK, which were applied @ 80:120:100 kg/ha. Total phosphorus (P) and potassium (K) were applied at the time of seed bed preparation, whereas half nitrogen (N) was



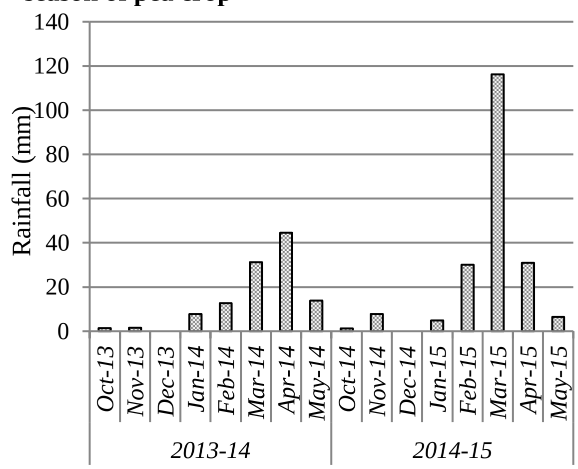
Maximum temperature (°C) during growing season of pea crop



Minimum temperature (°C) during growing season of pea crop



Mean relative humidity (%) during growing Season of pea crop



Rainfall (mm) during growing season of pea crop

applied as basal dose at seed bed preparation and remaining half was applied before flowering. Irrigation was applied as per requirement and was stopped about 15-20 days before harvesting, which was done in 2nd week of April.

***Treatments*:** Seeds of both pea varieties were soaked in 50, 100, 150, 200 or 250 mg/liter salicylic acid, while untreated (unsoaked) seeds served as control (0 mg/liter). After soaking for 12 hours, seeds were dried at room temperature (24±2°C) and sown in the field.

***Weather data*:** Data of weather conditions prevailing in growing region (Faisalabad) during growth season of pea crop for 2013-14 and 2014-15 is has been presented.

***Biochemical attributes*:** Biochemical attributes in leaf samples including chlorophyll, carotenoid, total phenolic and total soluble protein contents were determined after 50 days of sowing to analyze the biochemical changes in response to salicylic acid seed treatments and were expressed as mg/g F.wt. Chlorophyll and carotenoid contents were assayed by following the method of Arnon (1949) and Davies (1976) by recording absorbance of the samples at 645 nm, 663 nm and 480 nm using double beam spectrophotometer (Hitachi-120, Japan). Total phenolic contents were determined following the protocol of Julkenen-Titto (1985) and recorded absorbance of samples at 750 nm using double beam spectrophotometer (Hitachi-120, Japan). Total soluble proteins were calculated following the protocol devised by Lowry *et al.* (1951). The optical density (OD) was noted at 620 nm in a double beam spectrophotometer (Hitachi-120, Japan).

***Statistical analysis*:** Experiment was laid out according to Randomized Complete Block Design with 2-factor factorial arrangement (factor-1: variety and factor-2: concentration of salicylic acid) and three replications per treatment. Data are presented as mean of two years values. Data were statistically analyzed using analysis of variance (ANOVA) technique (Steel *et al*., 1997). Tukey's test ($P \leq 0.05$) was used to compare the significant difference among the mean values of treatments.

RESULTS

Vegetative and Reproductive Parameters

Main shoot and root length (cm): Seed treatment with salicylic acid increased main shoot length (Fig. 1) and root length (Fig. 2) of plants of both pea varieties *i.e.*, Meteor and PF-400, in a dose dependent manner. Main shoot length of Meteor and PF-400 plants under control treatment was 48.7 cm and 40.3 cm respectively. Longest main shoot of Meteor plants (60.2 cm) was recorded in response to seed treatment with 250 mg/L salicylic acid, whereas PF-400 plants (64.3 cm) were observed as the result of seed treatment with 200 mg/L salicylic acid, which were statistically significant than control. However, these results were statistically at par to 200 mg/L SA seed treatment for Meteor. Similarly, results of Meteor under control and SA 50 mg/L seed treatment were statistically at par. Results of PF-400 plants under SA 200 mg/L seed treatment were significantly different from all treatments. Increment in main shoot length as compared to respective untreated plants was 23.6% for Meteor and 59.5% for PF-400 plants in response to 250 mg/L and 200 mg/L salicylic acid seed treatments, respectively.

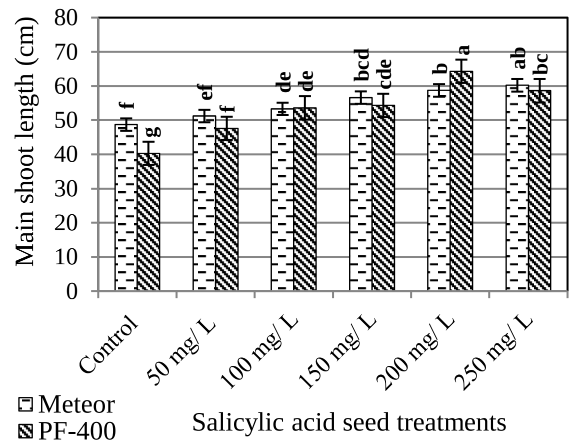


Figure 1. Effect of salicylic acid seed treatment on main shoot length of pea cultivars

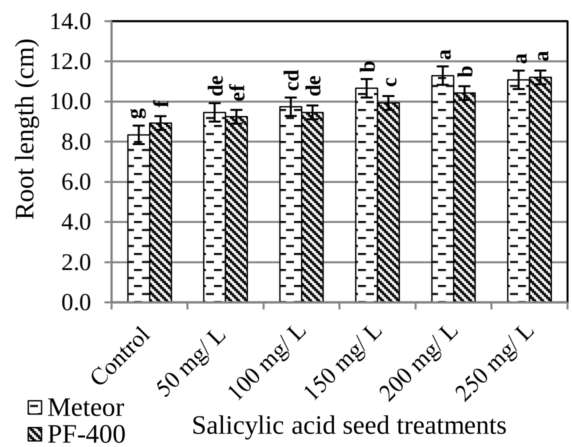


Figure 2. Effect of salicylic acid seed treatment on root length of pea cultivars

Minimum root length of Meteor (8.3 cm) and PF-400 (8.9 cm) plants was noticed for plants raised using untreated seeds (Fig. 2). Statistically significant maximum root length of Meteor plants (11.25 cm) was recorded in response to seed treatment with 200 mg/L salicylic acid, while that of PF-400 plants (11.2 cm) was recorded in response to the seed treatment with 250 mg/L. Response of Meteor plants to 200 mg/L and 250 mg/L salicylic acid seed treatments was statistically alike; however, these results were statistically significant than lower concentrations of salicylic acid. Results of PF-400 plants for SA 250 mg/L were significantly better than lower concentrations of SA as well as control. This maximum increase in root length was 35.5% for Meteor (200 mg/L) and 25.8% for PF-400 plants (250 mg/L) when compared to their respective control plants *i.e.,* plants raised from untreated seeds.

Number of flowers and pods per plant: Plants with salicylic acid seed treatment also showed better reproductive growth than plants without seed treatment. Minimum number of flowers per plant of both varieties *i.e.,* Meteor (23.2) and PF-400 (27.8) were recorded of plants raised from untreated seeds (Fig. 3). Statistically significant and the best results for Meteor and PF-400 plants as compared to respective control were found for 200 and 250 mg/L SA seed treatments respectively. Maximum number of flowers per plant of Meteor (44.7) and PF-400 (48.8) was produced in response to seed treatment with 200 and 250 mg/L salicylic acid, respectively. Results of 200 and 250 mg/L SA were statistically different for PF-400, while at par for Meteor plants. All the treatments of SA were statistically different from control. This maximum enhancement in number of flowers per plant as compared to respective control was 92.7% for Meteor (200 mg/L) and 75.5% for PF-400 (250 mg/L).

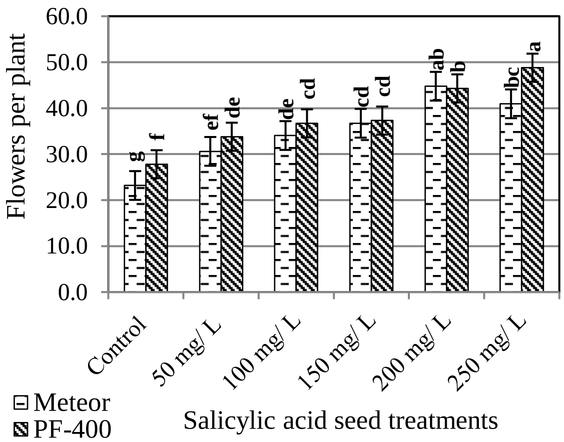


Figure 3. Effect of salicylic acid seed treatment on number of flowers per plant of pea cultivars.

Similarly, pods production of pea plants with SA seed treatment was enhanced as compared to control (Fig. 4). Meteor and PF-400 plants under control treatment produced minimum number of pods per plant *i.e.,* 20.3 and 21.8 respectively. Although all concentrations of SA produced statistically significant results than control for both pea varieties, however increase in pods production was more prominent for higher concentrations of *i.e.,* 150, 200 and 250 mg/L. Statistically best significant improvement in pods production for both varieties was in response to SA 250 mg/L seed treatment, which produced maximum number of pods per plant of both Meteor (40.0) and PF-400 (45.0). This enhancement was 97 and 106.4% as compared to their plants without seed treatment. Results of PF-400 plants for 200 and 250 mg/L salicylic acid seed treatments were at par statistically.

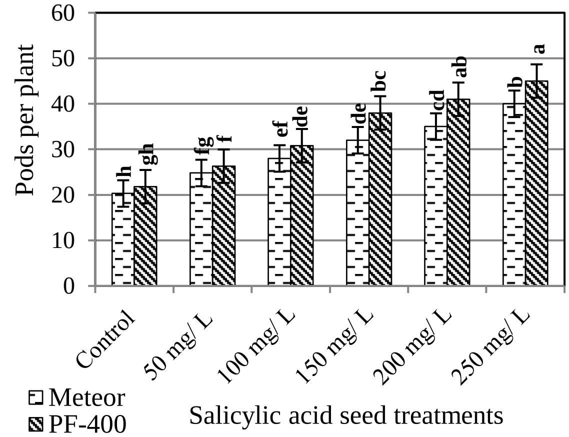


Figure 4. Effect of Salicylic acid seed treatment on number of pods per plant of pea cultivars

***Average seed yield per plant*:** Results of all the treatments for both varieties were statically different. Pea plants under control treatment produced minimum average seed yield *i.e.,* 30.2 g of Meteor and 26.9 g of PF-400 plants. Statistically most significant results among all treatments for both varieties were recorded in response to SA 200 mg/L seed treatment. Pea plants under this treatment produced highest seed yield per plant *i.e.,* 66.3 g of Meteor and 68.2 g of PF-400 plants as compared to plants under respective control. This highest increase in yield was 119.5% for Meteor and 153.5% for PF-400 plants in response to this SA seed treatment.

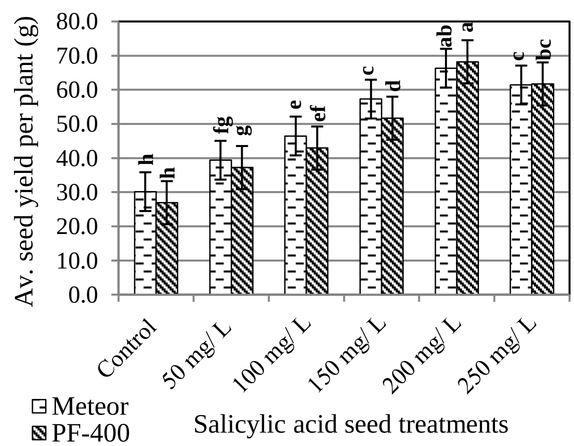


Figure 5. Effect of salicylic acid seed treatment on average seed yield per plant of pea cultivars

Biochemical Attributes

***Chlorophyll and carotenoid contents*:** Improvement in vegetative parameters of pea plants after salicylic acid seed treatment has been supported by concomitant statistically significant increase in accumulation of chlorophyll and carotenoid contents. Statistically best significant improvement in accumulation of photosynthetic pigments in Meteor and PF-400 plants than control was the result of SA 250 and 200 mg/L treatment, respectively. As Meteor plants with 250 mg/L seed treatment accumulated highest concentration of chlorophyll contents *i.e.,* 3.32 mg/g F.wt. among all SA treatments and compared to plants without seed treatment (2.41 mg/g F.wt.) (Fig. 6). Similarly, minimum (2.59 mg/g F.wt.) and maximum (3.53 mg/g F.wt.) chlorophyll contents accumulation in PF-400 plants was recorded in plants without seed treatment and plants with 200 mg/L salicylic acid seed treatment respectively. Results of 200 and 250 mg/L SA treatments were statistically at par for Meteor as well as PF-400. Maximum enhancement in chlorophyll contents as compared to plants under respective control treatment was 37.8% for Meteor (250 mg/L), while 36.3% for PF-400 plants (200 mg/L). Enhancement in chlorophyll contents with better source sink relationship and distribution of phyto-assimilates helped plants to achieve improved growth and development.

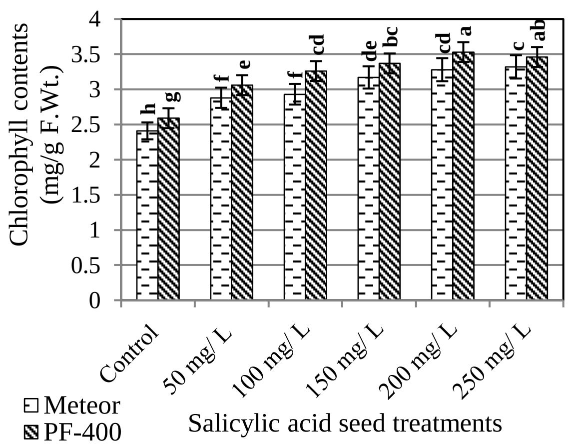


Figure 6. Effect of salicylic acid seed treatment on chlorophyll contents of pea leaves

In the same way, improvement in accumulation of carotenoid contents accumulation was observed in response to salicylic acid seed treatments depending upon the concentration used (Fig. 7). Plants of both pea varieties from untreated seeds accumulated minimum carotenoid contents *i.e.,* 0.62 mg/g F.wt. in Meteor and 0.56 mg/g F.wt. in PF-400 plants. Maximum accumulation of this constituent in Meteor (0.78 mg/g F.wt.) and PF-400 plants (0.83 mg/g F.wt.) was recorded as the result of 250 and 200 mg/L salicylic acid seed treatments respectively, which was statistically the most significant among all treatments. This increment of carotenoids accumulation as compared to respective plants under control treatment was 25.8% for Meteor and 48.2% for PF-400 plants in response to said treatments.

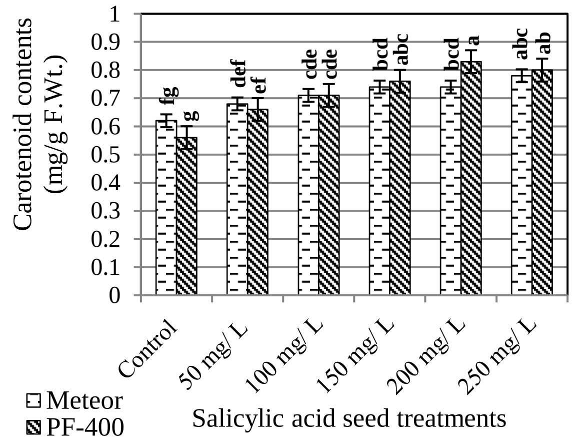


Figure 7. Effect of salicylic acid seed treatment on carotenoid contents of pea leaves

***Total phenolics and total soluble protein contents*:** All treatments of SA produced statistically different results than control for both pea varieties. It was observed that untreated PF-400 plants accumulated more total phenolic contents (8.9 mg/g F.wt.) than untreated Meteor plants (8.3 mg/g F.wt.) (Fig. 8). Although all treatments of SA produced significantly different results than control, however results of 200 mg/L and 250 mg/L for both varieties were statistically at par to SA 150 mg/L seed treatment. Maximum enhancement in synthesis of total phenolic contents as compared to control of both pea varieties was recorded as the result of 200 mg/L salicylic acid seed treatment *i.e.,* 17.4 mg/g F.wt. for Meteor and 15.6 mg/g F.wt. for PF-400 plants.

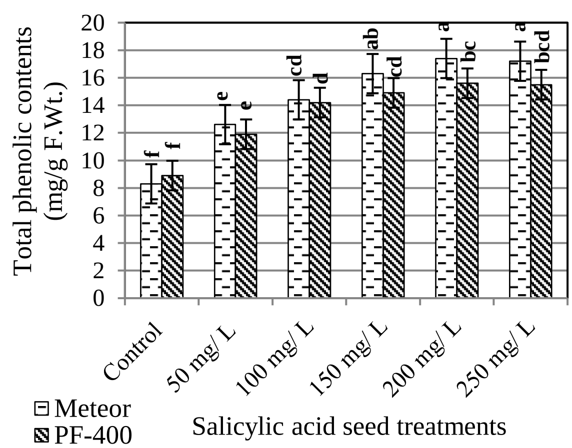


Figure 8. Effect of salicylic acid seed treatment on total phenolic contents of pea leaves

This maximum enhancement as compared to control was 109.6% for Meteor and 75.3% for PF-400 plants by this treatment. Total phenolic contents are involved in manipulation of growth processes, scavenging of reactive oxygen species (*ROS*) and free radicals, therefore their improvement is aligned with stimulation of growth parameters. Response of Meteor plants to salicylic acid 200and 250 mg/L seed treatments was statistically alike, whereas of PF-400 plants for same treatments was at par statistically.

Statistically significant improvement was recorded for synthesis of total soluble protein contents in pea plants in response to SA seed treatments than control (Fig. 9). Without SA seed treatment, plants of both pea varieties accumulated least total soluble protein contents *i.e.,* 2.83 mg/g F.wt. in Meteor and 2.69 mg/g F.wt. in PF-400 plants. As the salicylic acid concentration used for seed treatment was increased, plants accumulated more total soluble proteins; however, PF-400 plants with 50 mg/L seed treatment showed better results than plants with 100 and 150 mg/L salicylic acid seed treatments. Statistically most significant results as compared to all SA treatments and control were recorded for 200 mg/L SA treatment. Plants under this treatment showed maximum accumulation of total soluble protein contents in leaves of both Meteor (3.49 mg/g F.wt.) and PF-400 (3.58 mg/g F.wt.). This maximum enhancement when compared to respective plants without seed treatment was 23.3% for Meteor and 33.1% for PF-400 plants as the result of said treatment. Accumulation of total soluble proteins in plants of both varieties in response to SA 200 and 250 mg/L seed treatments was statistically at par.

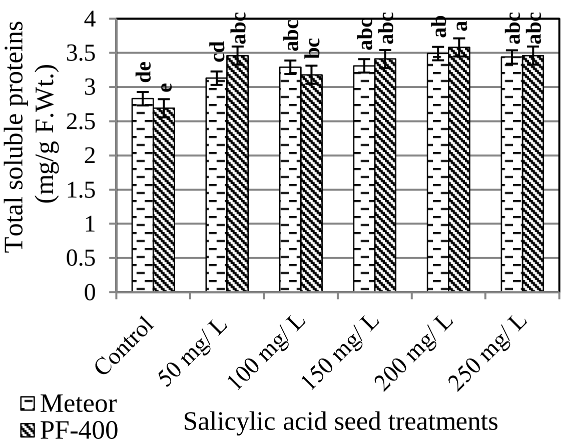


Figure 9. Effect of salicylic acid seed treatment on total soluble proteins of pea leaves

Quality Parameters of Harvested Seed

***Final germination percentage and seedling length (mm)*:** All SA treatments were statistically significant than control, yet few SA treatments were at par among themselves. Seeds of both pea varieties harvested from plants with salicylic acid seed treatments were better in quality as compared to seeds of control plants. Final germination percentage and seedling length of seeds harvested from plants with salicylic acid seed treatments improved considerably. Pea seeds harvested from control plants (without seed treatment) showed minimum final germination percentage *i.e.,* 76% for Meteor seeds and 78% for PF-400 seeds (Fig. 10). Maximum final germination percentage of Meteor seeds (97%) was statistically alike for seeds harvested from plants with 200 and 250 mg/L salicylic acid seed treatments. Similarly, highest final germination percentage of PF-400 seeds was 98% for seeds harvested from plants with 200 and 250 mg/L seed treatments. These results were statistically best as compared to the results of seeds harvested from control plants. This maximum improvement in final germination percentage was 27.6% for Meteor seed and 25.6% for PF-400 seeds as compared to their seeds harvested from plants under control treatment.

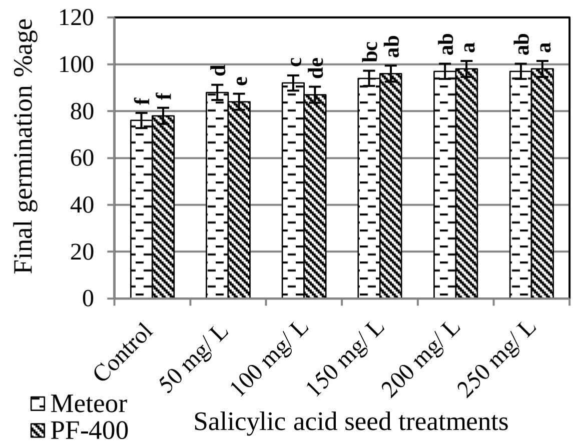


Figure 10. Effect of salicylic acid seed treatment on final germination percentage of harvested pea seeds

Similarly, results of seeds harvested from plants under salicylic acid treatments were statistically significant than seeds from control plants. As seeds harvested from pea plants under SA seed treatments produced longer seedlings than seeds of control plants (Fig. 11). Seeds of plants under control treatment produced least length of emerged seedlings *i.e.,* 83 mm of Meteor and 88 mm of PF-400 seedlings. Statistically most significant results for both pea varieties were recorded for seeds harvested from plants under SA 200 mg/L treatment. Seeds harvested from plants of this treatment produced longest seedlings of both varieties *i.e.,* 103 mm of Meteor and 109 mm of PF-400. This maximum increment was 24% for Meteor seedlings and 23.8% for PF-400 seedlings as compared to seedling grown from seeds harvested from control plants.

***Seedling vigor index*:** Better seedling vigor index of pea seeds harvested from plants under different SA seed treatments was observed than seeds of plants under control. Statistically different results recorded in response to SA treatments as compared to seeds of control validate the positive role of salicylic acid in quality seed production of pea plants (Fig. 12). Pea seeds harvested from control plants showed least vigor index *i.e.,* 63.0 for Meteor and 68.6 for PF-400 seedlings. However, plants of both pea varieties with 200 mg/L SA seed treatment produced best quality seeds, which showed highest seedling vigor index *i.e.,* 99.8 for Meteor and 107.4 for PF-400 seedlings as compared to seedling grown from seeds of plants without seed treatment. These results were most significant statistically among all recorded results. This maximum improvement was 58.4% for Meteor and 56.6% for PF-400 seedlings as compared to seeds harvested from control treatment.

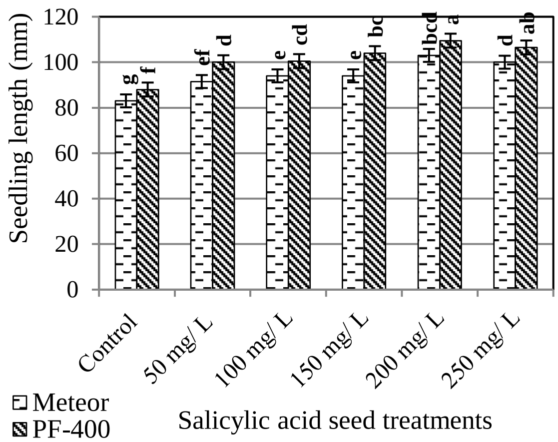


Figure 11. Effect of salicylic acid seed treatment on seedling length of harvested pea seeds

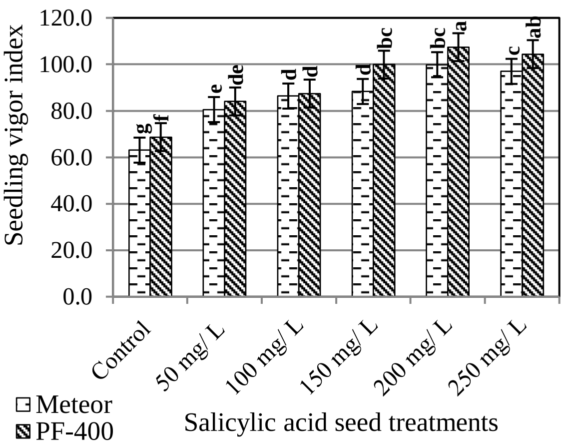


Figure 12. Effect of salicylic acid seed treatment on seedling vigor index of harvested pea seed

DISCUSSION

The present study explores the impact of salicylic acid on growth of two pea cultivars *i.e.,* Meteor and PF-400, when applied as seed soaking in relation to improvement of metabolic machinery of pea plants. Results of our investigation showed that soaking of pea seeds in salicylic acid solutions of different concentrations significantly improved vegetative traits *i.e.*, length of main shoot and root, reproductive characters as flowers, pods and average seed yield per plant with concomitant improvement in photosynthetic pigments (chlorophyll & carotenoid contents) and biochemical constituents (total phenolics & total soluble protein contents). Seeds produced by plants raised from SA treated seeds showed better final germination, seedling length and seedling vigor index than seeds of plants from control treatment.

Soaking of pea seeds in salicylic acid significantly improved vegetative growth of plants (Fig. 1, 2) and crop establishment as was previously reported for seed treatment of okra (Hussein, 2015) and common bean (Rady and Mohamed, 2015) with SA. Improved growth of pea plants may be explained with better root growth, which improved nutrient uptake from soil and deposition as dry matter (Rafique *et al*., 2011; Ahmad *et al*., 2015). A parallel statistically significant increase in accumulation of photosynthetic pigments (Fig. 6, 7) was recorded in response to SA treatments, which might have played role in growth promotion of pea plants. Earlier findings of Agami and Mohamed (2013), Moussa and Khodary (2003) and Krantev *et al*. (2008) validate the up-regulation of photosynthetic contents in response to seed treatment with SA. Seed soaking in this study, might have increased SA endogenous level due to *de novo* synthesis in treated seeds, which in turn activated various enzymatic reactions to improve germination and depicted in later on growth of plants (Szalai *et al*., 2011; Szalai *et al*., 2016). Growth improvement may also be due to increased level of cell division, differentiation and elongation within the apical meristem of growing plants (El-Khallal *et al*., 2009; Delavari *et al*., 2010).

Significant increment, efficient distribution and utilization of growth promoting hormones *i.e.*, IAA, GA_3, cytokinin and zeatin etc., in plants is also well documented in response to SA application (Thomas and Howarth, 2000; Hanieh *et al*., 2013; Rady and Mohamed, 2015; Shinwari *et al*., 2015; Soliman *et al*., 2016; Imriz, 2019), which might have improved photosynthetic machinery, ultimately resulted in growth promotion of pea plants under this study. Better reproductive growth and seed yield was recorded as the result of pretreatment of pea seeds with SA (Fig. 3, 4, 5). Rehman *et al*. (2015) (maize) and Rady and Mohamed (2015) (common bean) also reported improved vegetative growth and yield of plants raised from SA treated seeds as compared to control. In this connection, SA seed soaking is also relevant to increased uniform distribution of photoassimilates to reproductive parts and growing seeds, thereby increasing seed quantity and quality (Mahboob *et al*., 2015) as shown by the pea plants with different concentration of SA used as seed treatment. Enhanced reproductive growth and yield recorded under this study is similar to the finding of Heidari *et al*. (2015) for bean plants and can possibly be related to increased endogenous levels of SA, thus acting as stimulator of plant growth (Martinez *et al*., 2004).

Results of this study for better accumulation of chlorophyll and carotenoid contents in pea plants with pretreatment of SA are in corroboration with the findings of Baninasab (2010) in cucumber, Rafique *et al*. (2011) in pumpkin, Nimir *et al*. (2015) in sorghum as well as Rady and Mohamed (2015) in common bean. Similarly, findings of Karlidag *et al*. (2009) for strawberry, Shinwari *et al*. (2015) for rice and Fahad and Bano (2012) for maize also support the results of up-regulation of biochemical attributes as observed in present work. Another possible reason of growth promotion of pea plants may also be due to better accumulation and activity of total soluble sugars, total flavonoids, nitrate reductase, carbonic anhydrase and carboxylating enzymes within plant body (Khodary, 2004; Rady, 2011; Hassanein *et al*., 2014). Likewise, many studies indicated positive impact of treatment with salicylic acid in enhancing the CO_2 assimilation in plant body due to efficient role of carbonic anhydrase and nitrate reductase with efficient uptake and deposition of nitrogen, phosphorus, potassium and iron (Hayat *et al*., 2005; Gharib, 2006; Espin *et al*., 2011; Rady and Mohamed, 2015), which might have ultimately reflected as increased yield per plant as observed in this study. Improved quality of produced seed in response to SA seed soaking treatments might also be due to better assimilation of nutrients in seeds (Fig. 10,11, 12).

In addition to chlorophyll and carotenoid contents, total phenolics and total soluble protein contents significantly increased in leaves of pea plants pretreated with SA as seed treatment (Fig. 8, 9). Phenolic and total soluble protein contents are involved in various growth regulatory processes and are integral part of antioxidative system of plants to quench *ROS* produced during normal cellular processes. Improvement in accumulation of these bioactive molecules has been related to enable plants to utilize energy resources for growth enhancement rather than overcoming damages caused by scavenging free radicals. It has been observed earlier that salicylic acid treatment increased the synthesis of phenolic secondary metabolites in pea plants which is important for plant growth (McCue *et al*., 2000). This manipulation of biochemical attributes has been well represented in growth promotion and seed yield of both pea varieties. Treatment of pea seeds with SA improved biochemical components of plant metabolic machinery, which acted as signal inducer for growth and development of plants. These results are consistent with those of Singh and Usha (2003) in wheat, Rafique *et al*. (2011) in pumpkin, Misra *et al*. (2014) in *Catharanthus roseus*, Nimir *et al*. (2015) in sorghum and Roychoudhury *et al*. (2015) in mung bean, who observed better growth of plants in response to seed treatment with SA. Another possible mode of SA seed soaking may be improved physiological processes *i.e.*, source/ sink regulation, enzymatic activities, protein synthesis, polyamines and increased antioxidant capacity of pea plants, which ultimately regulated growth and development (Tiburcio *et al*., 2014; Pal *et al*., 2015; Soliman *et al*., 2016; Szalai *et al*., 2016). Similar results for improvement in accumulation of these bioactive compounds have been recorded by Ali *et al*. (2007) in ginseng roots. Increase in synthesis of soluble proteins can be assumed as a step of defense mechanism against cellular oxidative stress generated during regular metabolic functioning. Efficient accumulation of bioactive compounds in pea plants has been well reflected in the form of improved seed quality, seed yield and quality related attributes.

***Conclusion*:** Treatment of seeds of pea cultivars *i.e.*, Meteor and PF-400 with salicylic acid improved vegetative and reproductive growth of plants as compared to plants without seed treatment by manipulating biochemical attributes. Therefore, it can be concluded that treatment of Meteor and PF-400 seeds with 200 mg/liter salicylic acid (200 mg salicylic acid dissolved in one liter of water) can be utilized as a tool to get maximum seed yield. Similarly, statistically significant results for vegetative attributes have also been recorded in response to 250 mg/liter salicylic acid seed treatment than control. Pea seeds of cultivars Meteor and PF-400 may be treated with 200 or 250 mg/liter salicylic acid to improve vegetative growth, reproductive characters and yield of good quality seed in relation to better biochemical metabolic functioning of plants.

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