EFFECT OF EXOGENOUS APPLICATION OF PLANT GROWTH REGULATORS ON PHYSIOLOGY AND SECONDARY METABOLITE ACCUMULATION OF COLUBRINA ASIATICA L.

Manasi Patil[1], Nivas Desai[2], Ahilya Waghmode[1] and D.K. Gaikwad[3]

1. Sadguru Gadage Maharaj College, Karad, Dist- Satara, MS India
2. Shri Pancham Khemraj Mahavidyalaya, Sawantwadi MS India
3. Dr. Babasaheb Ambedkar Marathwada University Sub Capmus, Osmanabad, MS India

Email:-manasipatil202@gmail.com

Abstract

Colubrina asiatica L. seedlings (1 year old) was raised in garden soil. The effect of foliar spray of 100 ppm of Salicylic acid (SA), 6-Benzylaminopurine (6BA), Gibberellic acid (GA_3) and Indole-3-butyric acid (IBA) on physiology and secondary metabolites was analyzed. The alleviation in photosynthetic pigments and in secondary metabolites was observed due to all the foliar sparys and it was more pronounced in GA_3. The foliar application of plant growth regulators (PGRs) also Enhances the secondary metabolites in *C. asiatica* leaves. The foliar application of PGRs will certainly help to improve the medicinal potential of *C. asiatica* plants.

Introduction

Plant growth and developmental processes are very much regulated by certain chemical substances called Growth regulators. Growth regulators are known to improve the physiological efficiency including photosynthetic ability and can enhance the effective partitioning of accumulates from source and sink in the field crops (Ramesh and Ramprasad, 2013). Secondary metabolites are responsible for the medicinal value of the plants but they have very limited distribution than primary metabolites. Research on plant secondary metabolites has increased during last 50 years due to value necessity of the daily lives including health care on these plant products (Mulabagal and Tsay, 2004). Plant growth regulators (PGRs) have been used as proficient elicitors to stimulate production of plant secondary metabolites. Plant growth regulators include hormonal substances of natural occurrence (phytohormones) as well their synthetic analogues (Basra, 2000). The plant reaction to plant growth regulators may di er with species, age of plant, varieties, environmental conditions, stage of development, physiological and nutritional status and endogenous hormonal balance (Naeem et al., 2011). A number of studies have been performed and confirmed in vast plants to find out the e ects of unlike plant growth regulators on secondary metabolite production (Shilpashree and Rai, 2009). *Colubrina asiatica* (L.) is a member of family Rhamnaceae, climbing shining shrub, commonly known as Indian sankewood, leatherleaf. This is a promising medicinal plant. *C. asiatica* is often planted as an ornamental and to be used for medicinal purposes (USDA-ARS, 2014). Medicinal oil is prepared from seeds along with other ingredients, and is used to treat rheumatism and numbness in adults and also in treating weak legs in children (Selvam, 2007).

Thus, the present study was aimed to study the effect of PGRs on the physiology and biochemistry of *C. asiatica* plants.

Material and Methods

The present investigation was carried out during the year 2019-20 at SGM College, Karad district Satara. The experiment was laid in completely randomized design with pre-sowing soaking treatments using PGR. The PGR used were GA_3, SA, 6BA and IBA at 100 ppm. The experiment constituted of 4 treatment with three replications. Solutions of PGRs were prepared and sprayed on the foliage of plants twice in a week for 30 days with the help of hand sprayer as per treatment while in untreated control distilled water was sprayed. The photosynthetic pigments chlorophyll a and b were analysed as per the method of Arnon (1949). The olyphenols were analysed according to method of Folin and Denis (1915) and total flavonoids content was estimated by $AlCl_3$ method described by Luximan – Ramma et al. (2002). The alkaloid content was determined as per the method described by Singh et al. (2004). the quantitative test of alkaloids, cardiac glycosides, coumarins, phenols, saponins, sterols, tannins, Xanthoproteins was carried out using the methods of Trease and Evans(1985); Brindha et al., (1981) and Lala(1993).

Results and Discussion

Effect of foliar spray of 6-BA, GA_3, IBA, SA at 100 ppm concentrations on chlorophyll content in leaves of *C. asiatica* at 30 days is recorded in Table 1. It is clear that the chlorophyll a and total chlorophylls increases due to foliar sprays of 6-BA, GA_3, IBA, while application of SA showed reduction in chlorophyll. The concentration of chlorophyll b exhibits induction in response to PGR sprays and the effect is more pronounced in GA_3 treated plants, but the reduction of Chl-b was noticed in SA treated plants. The similar kind of results were obtained by various workers in other plants treated with GA. An increase in chlorophyll content with GA was also reported by Khan et al., (1996) and Hayat et al., (2001) in mustard. Gibberellic acid increased pigment concentration in maize (Kaya et al., 2006). The photosynthetic pigments chlorophyll (a and b) was highest for GA at 150 ppm in croton plants (Eid and Abou-Leila, 2006). SA also caused inhibitory action on chlorophyll contents. Pancheva et al. (1996) observed a reduction in chlorophyll content in barley leaves by application of SA. With increase in concentration of SA caused decrease in chlorophyll content in wheat and moong seedlings (Moharekar et al., 2003).
Our observations indicate there is increase in chlorophyll content due to foliar application of growth regulators. This increase would undoubtably helps to improve the photosynthetic efficiency. The applications of growth regulators may prove beneficial for improvement of growth and productivity of medicinally as well as economically important *C. asiatica* plant.

The polyphenol content as well as flavonoid content was slightly elevated with respect to all the applied growth regulators. The foliar application of these growth hormones exerts positive influence on accumulation of polyphenols. Among these GA_3, SA and BA exhibit marked accumulation of polyphenols. An increase in total phenolic compounds in wheat seedling was reported by Sari and Etebarian (2009) due application of SA. Patil and Gaikwad (2013)observed foliar application of PGRs (6-BA, GA, SA, CCC, Cysteine and Methionine) shows a positive influence on polyphenols and flavonoid in *Simarouba glauca*. Duxbury et al. (2004) studied effect of chitosan and 5- chlorosalicylic acid on total phenolic content of grapes and wine. Exogenous application of SA significantly enhanced flavonoid content of *Taraxacum officinale* (Dandellon) (Kim et al., 2009). As compared to control alkaloid content increases by 2 to 3 folds in all stressed as well as sprayed plants. Pitta-Alvarez et al. (2000) and Spolansky et al. (2000) reported that SA can be used as effective strategy to increase the production of important alkaloids.

The qualitative screening of phytochemical constituents of leaf extract of *C. asiatica* reveals the presence of phenols, coumarins, sterols, alkaloid, tannins, xanthoproteins, cardiac glycosides, saponins and terpenoids (Table 2). Phenols, sterols, alkaloids, tannins and saponins were accumulated in the treated plant as compared to control plants.

Table.- Effect of PGRs on physiological and biochemical parameters in the leaves of *C. asiatica*

	Control	GA	SA	6-BA	IBA
Chl-a (mg g-1 fresh weight)	0.92±0.03	1.31 ±0.02	0.90±0.01	1.14±0.01	1.26±0.02
Chl-b (mg g-1 fresh weight)	1.44±0.13	1.99 ±0.10	1.21±0.08	1.74±0.07	1.57±0.03
Total Polyphenols (mg g-1 fresh weight)	0.23±0.01	0.47±0.03	0.32±0.02	0.37±0.03	0.39±0.03
Total Flavonoids (mg g-1 fresh weight)	0.18±0.02	0.39±0.03	0.27±0.02	0.31±0.02	0.33±0.02
Total Alkaloids (mg g-1 fresh weight)	0.20±0.01	0.64±0.03	0.46±0.02	0.49±0.01	0.45±0.02

Table.2. Effect of PGRs on secondary metabolites in the leaves of *C. asiatica*

	Control	GA	SA	6-BA	IBA
Phenols	++	+++	++	+++	+++
Coumarines	+	+++	++	+++	++
Saponins	++	+++	+++	+++	+++
Cardiac Glycosides	+	++	+++	++	+++

Sterols	++	++	+++	++	+++
Terpenoids	+	+++	++	+++	++
Tannins	++	+++	++	++	+++
Alkaloids	+	+++	++	++	+++
Xanthoproteins	+	+++	+++	++	++

Conclusion

The present study on the effect of some plant growth regulators on the physiological and biochemical parameters revealed the superiority of application GA3 for majority of biochemical parameters. The foliar application of PGRs results in the induction of synthesis of several bioactive compounds which will improve medicinal potential of *Colubrina asiatica* plants.

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