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RESEARCH PAPER

Exploring the effects of indole butyric acid (IBA) on *in vitro* growth of potato (*Solanum tuberosum*)

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Key Message: In this study three doses of indole butyric acid (IBA) was assessed on the *in vitro* growth of potato (*Solanum tuberosum*). Here 1 ml of IBA emerged as the optimal concentration for promoting root length, root number, and leaf growth in Asterix variety potato.

Abstract

In this study the effect of different concentration of indole butyric acid (IBA) was examined on *in vitro* propagation of potato (*Solanum tuberosum*). Three doses of IBA (1 ml, 2 ml, and 3 ml) were applied in liquid medium to potato explants of Asterix variety. The main purpose of this study was to optimize the dosage of IBA that would develop the best results for potato micropropagation. The experiments were conducted at Gilgit Baltistan's Agriculture Lab. Various parameters such as number of roots, roots length, and total weight were measured after exposing Asterix explants to varying concentrations of IBA. All tests were conducted under aseptic conditions. Analysis of variance (ANOVA) was used for analyzing the collected data. Throughout the study period, the results indicated significant differences ($p>0.05$) in potato growth among different concentrations of IBA. Among the different treatments, the Asterix potato variety exhibited the longest root length (11.1 cm) treated with 1 mL of IBA compared to the other concentrations. The IBA dose of 1 ml also proved to be the most effective in promoting the number of roots (9) in the Asterix variety. In this experimentation, 1 ml of indole-3-butyric acid (IBA) addition to the solution resulted in a major rise in the number of leaves (9) in Asterix plants. These findings have significant implications for future micropropagation and cultivation of potato plants. The ideal concentration of 1 ml of indole butyric acid (IBA) to promote root population can be used to develop effective and inexpensive protocols for *in vitro* potato propagation.

Keywords: Efficient protocols, Indole butyric acid (IBA), *In vitro* propagation, *Solanum tuberosum*

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Introduction

Potato (*Solanum tuberosum*) is the world's most significant crop, cultivated on over 18 million hectares worldwide (Zaman et al., 2014; Zaman et al., 2016). It ranks as the 4th most important crop after wheat, maize, and rice. Potatoes holds an important position globally, as it is most important tuber crop (Jacobs et al., 2011). The crop is grown in more than 125 countries, and over 1 billion people consume them daily in various forms (Wu et al., 2018). Potatoes have experienced a significant surge in popularity within developing nations, attributed to their simplicity in cultivation, nutritional richness, substantial yields, and practical manageability (Zaheer & Akhtar, 2016). Potatoes are an essential food for humans. They can be eaten raw, added to other foods as a snack, or processed into a variety of potato-based goods and starch derivatives (Ahmad et al., 2012; Zaheer & Akhtar, 2016). However, the development of an economically profitable, dynamic, and sustainable potato subsector in developing countries necessitates overcoming persistent challenges of low production (Birch et al., 2012). To accomplish this goal, it is imperative to implement strategies focused on improving the quality of planting materials, developing potato cultivars that require less water, have higher levels of resistance to pests and diseases, and are more able to adjust to changing climate conditions. It is also critical to adopt farming practices that promote the sustainable use of natural resources (Dolničar, 2021).

In the broader framework of potato and agriculture development, it becomes clear that providing smallholder farmers with improved access to inputs for production, capital, and markets is essential. These approaches perfectly capture the variety of potato management practices used in developing and tropical areas. These kinds of advancements have greatly increased total production capacities and potato yields (Devaux et al., 2021). The discovery of auxins as plant growth regulating chemicals, its physiological effects and practical applications form an attracting story of horticultural research (Ghaffoor et al., 2003; Shah et al., 2015; Jan et al., 2015). A synthetic auxin that plays a crucial role in plant growth and development is indole butyric acid (IBA). In plant tissue culture to persuade root development and to speed up the success rate of *in vitro* propagation, IBA is greatly used. It stimulates the formation of adventitious roots, cell division and differentiates within plant tissues and maintains hormonal stability during the initial stages of plant development. In a range of plant species, including potatoes, use of IBA in tissue culture systems has displayed key potential (Frick & Strader, 2018).

Since indole butyric acid (IBA) has the ability to increase the efficiency and consistency of potato micropropagation, researchers have been paying more attention to how IBA affects potato *in vitro* growth (Hajare et al., 2021). The development of roots, the stimulation of shoot growth, and the effective establishment of plantlets are important considerations when figuring out the appropriate IBA dosage for *in vitro* processes. Numerous investigations have highlighted the impact of IBA on a number of features of potato *in vitro* propagation, including acclimation, shoot expansion, and root formation (Ghaffoor et al., 2003; Hajare et al., 2021). Extensive research is imperative to elucidate the mechanisms underlying the effects of IBA on various stages of potato micropropagation. Furthermore, a thorough comprehension of the elements such as potato cultivars, media composition, and environmental conditions that impact the response to IBA therapy is still lacking (Gildemacher et al., 2009). Therefore, the purpose of this study was to look at how IBA affects the *in vitro* propagation of potatoes. Our study aims to assess the effects of varying IBA concentrations and application techniques on potato plantlet growth, rooting effectiveness, and subsequent acclimatization.

Materials and Methods

Planting material and cultural conditions

A variety Asterix was used during this experiment, which is famous in Gilgit Baltistan. The plant material was collected from the tissue culture laboratory at the Agricultural Research Center, Directorate of Agriculture Research, Gilgit Baltistan. These potatoes were subjected to treatment with 100 ml of MS media, which contained growth regulator (IBA) at different concentrations, namely 1 ml, 2 ml, and 3 ml. The cultures were then incubated at a temperature of 25 ± 1 °C under a 16-hour light cycle, providing illumination with white fluorescent tubes (Philips TL 40W/54) at intensity of 2,000 lux.

Nutrient media preparation

The formulation of nutrient media is essential to the process of growing plant specimens. The Murashige and Skoog (MS) medium, which was first created by Hartman and Kester (1968), was used in this experiment along with different IBA concentrations. In order to prepare the media for this investigation, Murashige and Skoog (MS) 1962 were used as the growing medium. There were two different kinds of media used: liquid and solid. The growth of plant specimens was facilitated by the careful administration of these medium under well regulated laboratory conditions, which served as vital support for micro-scale plants. To create a standard solid medium, a liquid solution was augmented with 10-20% agar, resulting in a specific medium conducive to plant growth known as agar medium. This solid medium served as a semi-solidifying agent, facilitating explant growth. In contrast, the liquid medium contained all requisite nutrients essential for plant growth. It comprised 4.43 g of Murashige and Skoog (MS) medium, 30 g of sucrose, and 10 ml of an iron stock solution. The optimal conditions for plant growth and development were observed in the liquid medium.

Growth regulator preparation

Initially, 0.005 grams of IBA were placed in a petri dish to make a stock solution of the plant growth regulator. A few drops of NaOH (Sodium hydroxide) were added to aid in the dissolution of IBA. After that, 5 ml of distilled water was added to a conical flask, followed by growth regulator solution and an additional 5 ml of distilled water. The resultant mixture was stirred for 2-3 minutes in an electrical orbital shaker, until total suspension was attained. When solution was prepared completely, it was moved to a glass container and sealed compactly with a rubber band in order to protect it from sunlight. In order to ensure its constancy and prevention from degeneration, the container was stored in a refrigerator at -20 °C.

Analysis of data

The Analysis of Variance (ANOVA) technique was used to analyze data statistically. The Least Significant Difference (LSD) test was used to calculate differences between treatment means at the 5% probability level, using CPSS (Gomez & Gomez, 1984).

Results

Effect of IBA on root length

Data regarding root length of potato showed a significant difference ($P < 0.05$) among all treatments (Fig. 1). Plants treated with 1ml of IBA have maximum root length which is non-significantly ($P > 0.05$) with the plants treated with 2 ml of IBA. While the minimum root length was observed in control plants (Fig. 1).

Effect of IBA on number of roots

Data regarding effect of IBA on number of roots showed a significant difference ($P < 0.05$) among all treatments (Fig. 2). Plants treated with 1ml of IBA have maximum number of roots. While the minimum number of roots was observed in control plants (Fig. 2).

Effect of IBA on leaves number

In this experimentation, 1 ml of Indole-3-butyric acid (IBA) addition to the solution resulted in a major rise in the number of leaves in Asterix plants. Plants had a total leaf count of 9, when treated with 1ml of IBA. This treatment was recognized as the average dose for initiating leaf propagation, with a concentration of 1 ml of IBA (Fig. 3). The diagram suggests that higher IBA concentrations might result in rise in leaf number. These conclusions suggest that IBA, particularly at concentrations of 1 ml has an optimistic influence on the leaf population of Asterix.

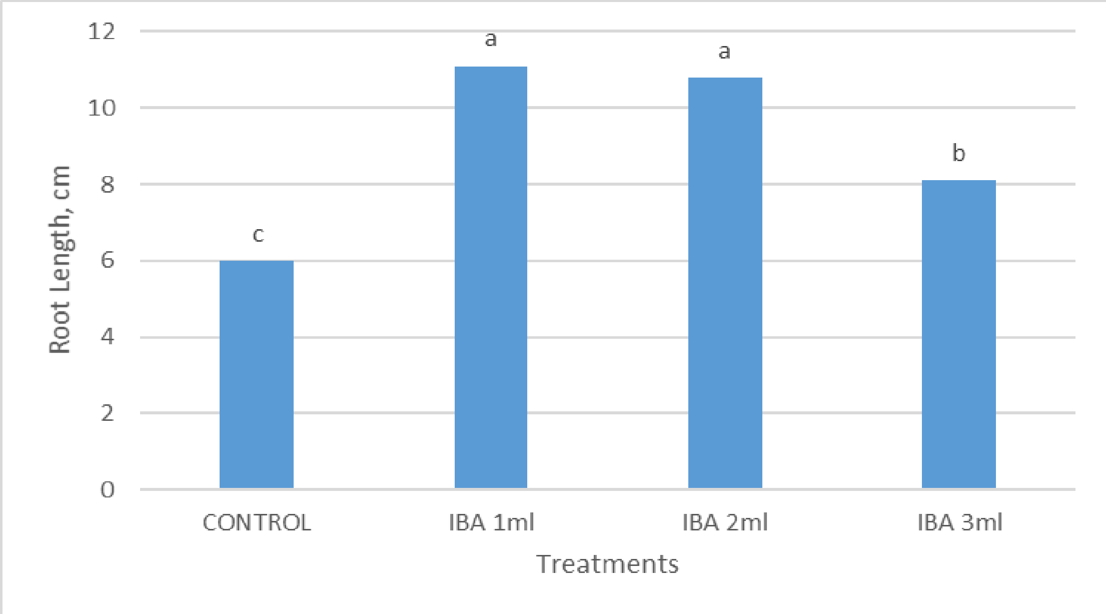

Fig. 1 Effect of.different concentrations of IBA on root length of potato. Means with different letters are significantly different (P≤0.05) from each other

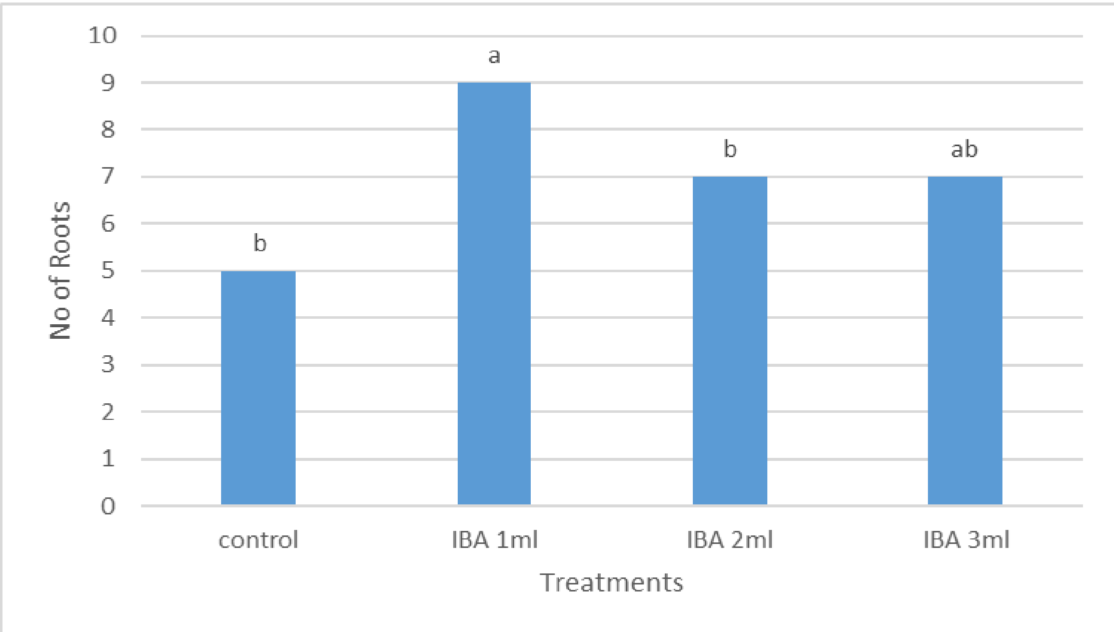

Fig. 2 Effect of different concentrations of IBA on number of roots of potato. Means with different letters are significantly different (P≤0.05) from each other

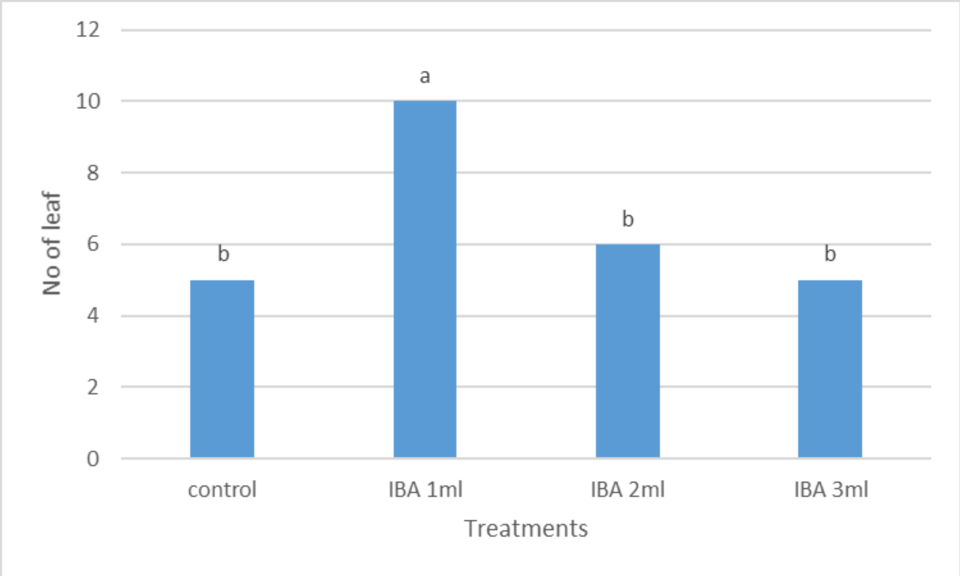

Fig. 3 Effect of different concentrations of IBA on number of leaves of potato. Means with different letters are significantly different (P≤0.05) from each other

Discussion

Potatoes are an important food staple that are high in nutrients and a staple in many people's diets. Potatoes are grown in over 125 countries and are the most popular tuber and root crop in the world. One billion people eat potatoes every day in one form or another. For innumerable people, the potato has come to represent nourishment, and its absence could lead to dietary deficits.

Perera and Dahanayale (2017) considered the effects of numerous concentrations of IBA (indole-3-butyric acid) and the ripeness level of single node stem cuttings for the asexual development of *Exacum ritigalensis*. Before being washed twice with distilled water the plant parts of plant were disinfected with 20% Clorox solution for 20 minutes. The cuttings were subsequent treated with various IBA concentrations (0, 2, 2.5, 5, 10, and 15 mg/L) and the survival rate was tested for two months. Following this period, it was discovered that semi-hardwood single node stems, each with half a single leaf or double leaves, had the highest rooting formation when treated with 10 mg/L and 15 mg/L of IBA in contrast to other treatments. The control group showed 100% survival rate using 3 buds per cutting.

Gurel and Guisen (1998) in initial research selected two diverse cultivars for investigation on vegetative propagation of almond (*Amygdalus communis* L.). Various levels of IBA (0, 0.1, and 0.5 mg/L) were tested to detect shoot growth and development at three sequential stages: initiation, transplantation, and development. The medium with a low IBA concentration (0.1 mg/L) appeared to be effective only for the multiplication and transplantation stages. The combination of 0.1 mg/L IBA demonstrated its importance and positive impact on shoot development during the last two stages. According to Overvoorde et al. (2010), auxins accelerate the development and growth of roots by promoting cell division in the root zone. Root initiation is influenced by the interplay of external and internal factors, as described by Rogg et al. (2001). IBA is the most commonly used and potent auxin, displaying high stability and low toxicity compared to other types and levels of endogenous PGRs.

The findings from this study have markedly enhanced our understanding of the involvement of IBA in potato micropropagation. Moreover, the insights gained have been instrumental in formulating the procedures utilized in commercial potato tissue culture systems. Additionally, this research holds promise for enhancing the efficacy, rapidity, and success rate of potato propagation. Consequently, there is potential to augment the availability of superior planting material and elevate overall potato production, thereby contributing to sustainable agricultural methodologies.

Conclusion

The *in vitro* propagation of the potato variety Asterix was the main focus of this study. The findings indicate that the therapy has a positive impact on root development, with the 1 ml/L concentration of indole butyric acid (IBA) showing better effects than other treatments. Additionally, our research highlights the fact that the 1 mL (IBA) treatment promotes strong root elongation as well as a significant rise in the maximum number of roots. Significantly, our results show that, in comparison to other criteria, indole butyric acid (IBA) has a greater impact on the root population.

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