

Asian Journal of Research in Crop Science

Volume 9, Issue 3, Page 50-56, 2024; Article no.AJRCS.121907 ISSN: 2581-7167

Effect of Chemical Treatments for Breaking Dormancy and Enhancing Sprouting in Freshly Harvested Potato Tubers in Nepal

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Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

Article Information

DOI: https://doi.org/10.9734/ajrcs/2024/v9i3288

Open Peer Review History:

This journal follows the Advanced Open Peer Review policy. Identity of the Reviewers, Editor(s) and additional Reviewers, peer review comments, different versions of the manuscript, comments of the editors, etc are available here: https://www.sdiarticle5.com/review-history/121907

Original Research Article

Received: 26/06/2024 Accepted: 29/08/2024 Published: 05/09/2024

ABSTRACT

This study aims to improve low potato production in Dolakha, Nepal, by identifying effective chemical treatments to break dormancy in freshly harvested tubers. The experiment was performed in Complete Randomized Design (CRD) with three replications. Eight treatments were used: gibberellic acid (GA3) at 50 ppm and 100 ppm, benzyl amino purine (BAP) at 50 ppm and 100 ppm, sugar solutions (0.5% ethanol + 1% sugar, 0.5% ethanol + 10% sugar, and 50% sugar) and distilled water. Medium-sized Ms-42.3 tubers were soaked in these treatments for two hours before being stored in a dark room. The results demonstrated that GA3 at 100 ppm was the most effective, reducing the days to first sprout emergence to 1.8 days and breaking dormancy in 2.63 days. This treatment also resulted in the longest sprouts, measuring up to 17.70 mm at 50 days. GA3 at 50 ppm similarly improved sprout emergence and length. BAP treatments (50 ppm and 100 ppm) and

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Cite as: Luitel, Prabesh, Diksha Sigdel, and Laxmi Kanta Paudel. 2024. "Effect of Chemical Treatments for Breaking Dormancy and Enhancing Sprouting in Freshly Harvested Potato Tubers in Nepal". Asian Journal of Research in Crop Science 9 (3):50-56. https://doi.org/10.9734/ajrcs/2024/v9i3288.

50% sugar solution also significantly shortened the time to sprout emergence and dormancy breaking compared to the control. Specifically, BAP at 100 ppm reduced the time to first sprout emergence to 3.63 days and was effective in promoting sprout density and length. Sugar solutions, particularly 50% sugar, showed promise in early sprout emergence and growth. In contrast, the control treatment (distilled water) yielded the lowest sprout density, with a maximum of 15.20 days to first emergence and sprouts measuring up to 4.53 mm at 50 days. This study provides valuable insights into effective sprouting techniques, potentially boosting agricultural productivity in similar regions.

Keywords: Potato tubers; Complete Randomized Design (CRD); gibberellic acid (GA3); ethylene; cytokinin; dormancy.

1. INTRODUCTION

Potato (Solanum tuberosum), an annual plant in the nightshade family (Solanaceae), originated from South America [1]. It is abundant in minerals like potassium, phosphorus, and magnesium, as well as nutrients like protein, carbs, and vitamins (particularly B1, B3, B6, and C) [2]. It is the fourth most important crop after rice, maize, and wheat in terms of production and productivity in Nepal [3]. In Nepal, cultivated in 1, 98,788 ha with 332523 mt production and 16.73 mt/ha productivity contributing 2.17% to the GDP and 6.57% to the AGDP whereas Dolakha accounts for 3069 ha area, yielding 55917 mt annually with the productivity of 18.22 mt/ha [4].

Dormancy of potato tuber is a physiological state characterized by a period during which autonomous growth of the sprout is suspended even under optimal sprouting conditions i.e. darkness, 15 to 20°C, and relative humidity of about 90 %. [5]. During dormancy, potato tubers do not undergo biological processes, resulting in delayed sprouting [6]. Various factors influence tuber dormancy such as temperature, moisture, and genetics [7]. After harvest, tubers stay dormant until biochemical changes during storage break this dormancy. Once dormancy is broken, sprouts begin to grow, and the tubers supplvina nutrients to support the start development of these sprouts and the formation of roots [8,9].

Chemicals such as cytokinin, ABA, and ethylene dormant. contribute to keeping potatoes However, treatments like heat and GA3 can help potatoes sprout earlier and improve their growth [10] & [11]. Proper cleaning and concentration are crucial for GA3 treatment, as higher endogenous gibberellin levels are observed before breaking dormancy [12]. GA3 can treat old and bruised tubers, improving performance and productivity [13]. Exogenous cytokinins can also break dormancy, with BAP widely used to stimulate cell division and morphogenesis [14]. Higher concentrations of BAP have shown faster dormancy breaking and better germination rates in potato varieties [15].

In vitro studies show that ethanol treatment breaks potato tuber dormancy by inducing the apical bud's growth. Primary alcohols can break dormancy in tubers, but secondary alcohols cannot. Additionally, the effect of ethanol on sprouting and gene expression in tuber tissue is blocked by a substance that inhibits alcohol dehydrogenase [16]. The concentration of ethanol (95%) varies based on the dormancy stage. The sprouting rate increases with 1% or 8% sucrose in the medium. Sprouts visible in 3 days with ethanol treatment, reaching close to 100% sprouting by day 5 or 6 [17]. In contrast, sugar solution treatment results in 50% sprouting in the dark, 50% in semi-dark, and 100% in light within 12-16 days. Sprouts start within 16 days of treatment [18].

Due to insufficient technology and dependency on conventional agriculture, potato production is still low compared to the attainable yield and yield of neighboring countries like India and China [19]. This research aims to identify effective treatments for breaking dormancy in freshly harvested potato tubers, enabling their use as seed potatoes in Dolakha. This will help farmers improve crop rotation, ensure better germination, and meet early market demands.

2. MATERIALS AND METHODOLOGY

2.1 Experimental Site

This research was conducted under laboratory conditions at the Potato Crop Development Centre, Dolakha, Nepal.

2.2 Experimental Design

The experiment was performed in Complete Randomized Design (CRD) with three

replications and eight treatments using different concentrations of GA3, BAP, ethanol, and sugar. To test sprouting, we used medium-sized Ms-42.3 tubers (25-35 grams). After cleaning them, we placed 15 potatoes on paper plates in a storeroom and treated them with chemicals.

2.3 Sample Collection Preparation of Chemical Treatment

The tubers required for the research experiment were collected locally from Bhimeswor municipality, Dolakha, and were harvested in early March. Ms-42.3 variety was used because it's widely grown and preferred by local farmers. After cleaning and sorting, medium-sized tubers (25-35 grams) were chosen. The treatments were made by mixing different chemicals with distilled water, except for GA3 and BAP, which were first dissolved in a strong base (NAOH) before being mixed with water. Tubers were washed with tap water and then soaked in the chemical treatments for two hours. For the control group, tubers were soaked in distilled water for two hours, then dried and stored in a dark room for observation.

2.4 Treatments Details

The treatments are as follows: T1 is the control, T2 and T3 use 50 ppm and 100 ppm Gibberellic acid, T4, and T5 use 50 ppm and 100 ppm cytokinin (Benzyl Amino Purine), and T6, T7, and T8 use 0.5% ethanol + 1% sugar, 0.5% ethanol + 10% sugar, and a 50% sugar solution, respectively.

2.5 Data Observation

Five tubers were randomly selected from each experimental unit through simple random sampling and data were observed regularly. Data were collected on the following parameters:

2.5.1 Days to first emergence of sprouts

The days required to induce the emergence of first sprouting were recorded as the days to first emergence. It was obtained by counting the days for the emergence of the first sprout after treating the tubers with respective treatments. It was observed regularly after the treatment and the days after treatment when the first sprout occurred were noted for each sample tuber.

2.5.2 Days for dormancy breakdown

Dormancy was considered to be broken when more than 80% of sample tubers showed about 2 mm long sprouts.

2.5.3 Number of sprouts per tuber

It is measured by counting the sprouts in each sprouted sample tuber. The observation was taken 10, 20, 30, 40, and 50 days after treatments.

2.5.4 Sprout length per tuber

The sprout length was measured with the help of a millimeter (mm) scale. The sample of five tubers was selected randomly, measured, and recorded every 10-day interval at 10, 25, 30, 40, and 50 days after treatments.

2.6 Statistical Analysis

All the collected data throughout the experimental period were tabulated in Ms-Excel and and significant mean differences were compared using statistical analysis using R studio software. The data were subjected to a one-way (treatments) analysis of variance (ANOVA), and significant mean differences were compared using Duncan's Multiple Range Test (DMRT) at a 0.05 percent level of significance.

3. RESULTS AND DISCUSSION

3.1 Days to the First Emergence of Sprouts

The experiment results demonstrated that treatment with different amounts of chemicals (BAP, ethanol, sugar, and gibberellic acid) had a substantial impact on the days when sprouts first appeared (Table 1). Gibberellic acid at 100 ppm (1.8) showed the minimum days to initial emergence, while Gibberellic acid at 50 ppm (2.06) showed the same results. The results obtained from 50 ppm GA3, 100 ppm BAP and 50% sugar solution were statistically similar. The experiment revealed that the control group had a maximum number of days until the first emergence (15.20). Based on the overall findings, it can be inferred that gibberellic acid was more effective than other treatments in promoting the initial signs of sprouting since GA3 promotes cell elongation in plants. The increased cell elongation facilitates the expansion of the shoot and root systems, allowing the sprouts to

push. The outcomes align with the discoveries made by other scholars. [20,21]. Rahman et al (2006) have demonstrated that reducing the number of days to 50% sprouting was the outcome of raising the GA3 concentration [20]. Similar to Zaghum et al (2021) conclusion that treatment with 300g sugar powder in 600ml water solution i.e. 50% sugar solution produced a 50% sprout percentage in dark conditions, 50% in semi-dark conditions, and 100% in open conditions over 12-16 days since sugar acts as a nutrient source to potato tuber and also creates create an osmotic potential that can draw moisture from the potato tuber's inner cells [18]. This initiates the sprouting process and encourages the growth of buds or eyes. The presence of ethanol in the media affects the rate at which micro-tubers sprout when placed in a medium containing either 1% or 8% sucrose. At both high and low sucrose levels in the medium. nearly 100% of the seeds germinate by day 5 or 6 [17].

3.2 Days to Breaking the Dormancy

Using various chemical concentrations (BAP, sugar, ethanol, and gibberellic acid) significantly affects how quickly potato tubers break dormancy. The results show that these treatments can greatly reduce tubers' days to break dormancy compared to the control group (Table 1). Gibberellic acid at 100 ppm (2.63) required the fewest days to break dormancy, trailed by Gibberellic acid at 50 ppm (3.06). The control group experienced a maximum of 27.26 days for the potato to emerge from dormancy. The results of using 50% sugar solution and 50

ppm GA3 were statistically equivalent, although BAP 100 ppm (3.63), BAP ppm (4.83), and 50% sugar solution (4.26) also assisted in ending potato dormancy earlier. Overall, GA3 is the most effective at breaking dormancy in potato tubers. BAP and sugar solutions are also useful for this purpose. All three treatments work by mobilizing stored nutrients and overcoming dormancy inhibitors. During the observation period, a high dose of gibberellic acid i.e. 100ppm was more effective than 10ppm GA3 treatments in breaking dormancy of potato tubers. This finding was aligned with the findings of Salimi et al. [22]. This result is also consistent with Turnip et al. [23], who reported that soaking the potato tuber seeds in a cytokinin solution shortened the dormancy release time by 12.58 days as compared to the period without soaking the potato tuber seeds in a cytokinin solution.

3.3 Number of Sprouts Per Tuber

Table 2 illustrates the results of the variance analysis, which indicated that the number of sprouts per tuber was significantly impacted by different treatments applied at successive data recordings. The 100 ppm Gibberellic acid (3.27) had the greatest sprouts at 10 DAT, while the control group had the weakest sprouts in the study (0.00). In Gibberellic acid 100 ppm (3.60) at 20 DAT, the highest sprouts per tuber was noted whereas the bare minimum of sprouts per tuber (1.20) was seen in control. When compared to Gibberellic acid 50 ppm (3.26) and 0.5% ethanol + 10% sucrose (3.26), which had

Treatments	Days to emergence	Days to dormancy break down
GA3 50 ppm	2.06 ^{cd}	3.06 ^d
GA3 100 ppm	1.80 ^d	2.63 ^d
BAP 50 ppm	2.86 ^c	4.83 ^{bcd}
BAP 100 ppm	2.26 ^{cd}	3.63 ^{cd}
0.5% ethanol + 1% sucrose	3.83 ^b	6.50 ^b
0.5% ethanol + 10% sucrose	2.80 ^b	5.56 ^{bc}
Sugar 50%	2.23 ^{cd}	4.26 ^{bcd}
Control	15.20 ^a	27.26 ^a
LSD	0.86	2.25
SEM(+-)	0.10	0.26
F-probability	<0.001	<0.001
CV%	12.14	18.04
Grand mean	4.13	7.22

CV= Coefficient of Variation, LSD= Least Significant Difference, SEM= Standard Error of Mean. The column with the same letter (s) in superscript indicates no significant difference between treatments. . '***' Significant at 0.001 Level of Significance; '*' Significant at 0.01 Level of significance; '*' Significant at 0.05 Level of Significance.

Treatments	10 DAT	20 DAT	30 DAT	40 DAT	50 DAT
GA3 50 ppm	3.00ª	3.26 ^{ab}	3.26 ^{ab}	3.26 ^{ab}	3.26 ^{ab}
GA3 100 ppm	3.27 ^{ab}	3.60ª	3.8ª	3.93ª	3.93 ^a
BAP 50 ppm	2.53 ^{bc}	2.86 ^{abc}	2.93 ^{ab}	2.93 ^{abc}	2.93 ^{abc}
BAP 100 ppm	2.13°	2.40 ^{bc}	2.60 ^{bc}	2.73 ^{bc}	2.73 ^{bc}
0.5% ethanol +	2.93 ^{ab}	3.00 ^{abc}	3.06 ^{ab}	3.20 ^{ab}	3.20 ^{ab}
1% sucrose					
0.5% ethanol +	2.46 ^{bc}	3.13 ^{abc}	3.26 ^{ab}	3.46 ^{ab}	3.46 ^{ab}
10% sucrose					
Sugar 50%	2.00 ^c	2.30°	2.53 ^{bc}	2.63 ^{bc}	2.63 ^{bc}
Control	0.00	1.20 ^d	1.80 ^c	1.86°	1.86 ^c
LSD	0.67	0.91	0.95	1.07	1.07
SEM(+-)	0.07	0.09	0.10	0.15	0.15
F-probability	<0.001	<0.01	<0.05	<0.05	<0.05
CV%	17.08	19.49	18.92	21.41	21.41
Grand mean	2.29	2.72	2.90	2.90	2.90

Table 2. The number of sprouts per tuber induced by different chemical doses on potato tuber

CV=Coefficient of variation, LSD= Least Significant Difference, SEM= Standard Error of Mean. The column with the same letter (s) in superscript indicates no significant difference between treatments. '***' Significant at 0.001 Level of Significance; '**' Significant at 0.05 Level of Significance.

Treatments	10 DAT	20 DAT	30 DAT	40 DAT	50 DAT
GA3 50 ppm	7.73	10.33ª	12.33ª	13.73ª	14.70 ^b
GA3 100 ppm	8.33	10.80 ^a	13.20 ^a	15.20ª	17.70 ^a
BAP 50 ppm	4.26	6.06 ^{bc}	8.20 ^b	9.26 ^{bc}	11.03°
BAP 100 ppm	5.73	8.00 ^b	8.93 ^b	11.06 ^b	13.06 ^{bc}
0.5% ethanol + 1% sucrose	3.06	4.93°	6.86 ^b	8.00 ^c	10.30°
0.5% ethanol + 10% sucrose	3.73	5.53 ^c	7.00b	8.46 ^{bc}	10.73°
Sugar 50%	5.06	6.86 ^{bc}	8.53 ^b	9.80 ^{bc}	11.36°
Control	0.00	1.26 ^d	2.20 ^c	3.33 ^d	4.53 ^d
LSD	1.79	2.10	1.90	2.63	14.53
SEM(+-)	0.18	0.22	0.20	0.27	0.30
F-probability	<0.001	<0.001	<0.001	<0.001	<0.001
CV%	21.83	18.08	13.08	15.44	14.53
Grand mean	4.74	6.72	8.40	9.85	11.67

Table 3. Sprout length per tuber (mm) in	nduced by different chemical doses
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CV=Coefficient of variation, LSD=Least Significant Difference, SEM=Standard Error of Mean. The column with the same letter (s) in superscript indicates no significant difference between treatments. '***' Significant at 0.001 Level of Significance; '**' Significant at 0.05 Level of Significance; '**' Significant at 0.01 Level of Significance; '**' Significant at 0.05 Level of Significance

the fewest sprouts in the control (1.80), Gibberellic acid 100 ppm (3.80) contained the most sprouts at 30 DAT. At 40 DAT, it was discovered that the number of sprouts per tuber was much larger in Gibberellic acid 100 ppm (3.93), while the lowest number of sprouts was obtained at the control (1.86). The results from 40 DAT and 50 DAT showed the same sprout density, with no consistent rise. Increased concentrations of GA3 were associated with similar outcomes to generating more sprouts [24,21]. These findings are similar to the results of Rossouw [25] where the result obtained by them indicates that a lower concentration of cytokinin resulted in more sprout growth than a higher concentration.

3.4 Length of Sprout Per Tuber

The impact of different chemical treatments on the length of sprouts (Table 3). The experiment demonstrated that sprout length was significantly impacted by varying treatment concentrations compared to the control. Gibberellic acid at 100 ppm (8.33 mm) produced the longest sprouts on 10 DAT, while the control group did not develop any sprouts at all. The length of sprouts per tuber in Gibberellic acid 100 ppm at 20 DAT is 10.80 mm, the longest among the other treatments. and 1.26 mm, the shortest in the control. The results from the 30-day assay also show that 100 ppm of gibberellic acid produces the longest sprouts, measuring 13.20 mm, whereas the control group shows the shortest sprouts, measuring 2.20 mm. The highest length of sprouts was seen at 40 DAT and 50 DAT in Gibberellic acid 100 ppm 15.20 mm & 17.70 mm, respectively, followed by Gibberellic acid 50 ppm 13.73 mm & 14.70 mm. In both observations, the control group produced the shortest sprout length. The control treatment yielded 3.33 mm at 40 DAT and 4.53 mm at 50 DAT. This result showed that the length of the sprout rises along with an increase in GA3 and BAP concentration [24]. The sprout length increased with an increase in GA3 concentration. Moreover, with longer storage times, sprout length rose in every treatment [21,11].

4. CONCLUSION

This study confirmed that gibberellic acid (GA3) is highly effective in breaking dormancy and promoting sprouting in freshly harvested potato tubers. Specifically, GA3 at 100 ppm reduced the time to sprout emergence and increased the number and length of sprouts more than other treatments. This suggests that GA3 can enhance seed potato quality and meet early market demands by accelerating sprouting and growth. The superior performance of GA3 compared to other chemicals like BAP, sugar solutions, and ethanol highlights its potential for improving potato production, particularly in areas with limited resources. However, the study was limited to one potato variety and location. Future research should include various potato types and conditions to validate these findings and optimize methods across different sprouting environments.

DISCLAIMER (ARTIFICIAL INTELLIGENCE)

Author(s) hereby declare that NO generative Al technologies such as Large Language Models (ChatGPT, COPILOT, etc) and text-to-image generators have been used during the writing or editing of manuscripts.

ACKNOWLEDGEMENT

This research was supported by PMAMP PIU and the Potato Crop Development Centre, Dolakha.

COMPETING INTERESTS

The authors have declared that no competing interests exist.

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