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Full Length Research Paper

Haulm application and dipping treatments of gibberellic acid on tuber dormancy breaking and sprout induction of potato (*Solanum tuberosum* L.) in Central Highlands of Ethiopia

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Potato, an important food and nutrition security can be produced two or more times in a year in Ethiopia. It plays an important role in human diet and a source of income for smallholder farmers. However, its productivity is low owing to limited availability of planting materials and poor tuber sprouting of improved varieties. Lack of quality seed is a major problem affecting expansion of potato production. The objective of the study is to determine effects of different methods and rates of Gibberellic acid (GA₃) application on dormancy of potato variety, Gera cultivar. The experiment was conducted at Holetta Agricultural Research Center during 2008 to 2009. It consisted of five levels of GA₃ as haulm application, a week prior to destruction and five levels of GA₃ as a dipping treatment immediately after harvest for 24 h. Randomized Complete Block Design with three replications was used. Result revealed GA₃ application affects dormancy and sprouting. Haulm application of GA₃ at 750 and 1000 ppm reduced dormancy period by 24 to 27 days, respectively. Dipping treatments of 40 and 50 ppm reduced dormancy period by 18 to 20 days, respectively. The study indicated that haulm application of GA₃ at 750 or 1000 ppm and dipping treatments of 40 or 50 ppm resulted in early dormancy termination, shoot emergence and increased sprout.

Key words: Gibberellic acid, dormancy breaking, haulm application, dipping, sprout induction.

INTRODUCTION

Potato (*Solanum tuberosum* L.) is the third most important food crop in the world after wheat and rice, with an annual global production exceeding 374 million tons

(CIP, 2016). It is perceived only as a source of carbohydrates, but is also an excellent source of essential amino acids (King and Slavin, 2013). Since

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2007, more potatoes are produced in developing than in industrialized countries (FAO, 2008). More than a billion people consume potato on a regular basis, and it is a vital source of income for millions of farmers (Devaux et al., 2014). In many African and low-income countries, such as Ethiopia, potato has emerged as an important crop, especially in the high and mid-altitude areas (Haverkort et al., 2012). Ethiopia has favorable climatic and edaphic conditions that favor the production of ware potato and high quality and virus-free seed potato that can be grown on 70% of the arable land in the country (Solomon, 1985; FAO, 2008). Despite the prevailing suitable conditions, potato productivity has remained low with the productivity of 13.9 t ha⁻¹ (CSA, 2017/18) which is still below world average of 19 t ha⁻¹ (FAOSTAT, 2018). Estimated potato cultivated land was 160,000 ha in 2001 (CSA, 2001) while it reached 0.3 million hectare in 2015 with production volume of 572,000 to 3.66 million tons, respectively (CSA, 2015/16).

Farmers produce between 19 and 38 t ha⁻¹ using improved seed and crop management practices (Gebremedhin et al., 2008). Paul et al. (2012) also stated that potato yields vary considerably across the world, with the lowest being in Sub-Saharan Africa which is less than 75% of the global average and less than 30% of the top producing regions. Many factors contribute to the low yield of potato such as narrow genetic basis of the varieties, lack of appropriate agronomic practices, use of poor quality planting material (seed tuber), low soil fertility, poor storage management for ware and seed potato, besides pest and diseases resistant high yield varieties which are stable (Haverkort et al., 2012; Gebru et al., 2017). In areas with a tradition of more than one production cycle per year and a bimodal rainfall pattern, there is little time between growing seasons to permit adequate tuber sprouting of improved potato varieties released in Ethiopia. In general, there is a limited supply of high quality seed tubers and it is one of the major constraints to potato production in many developing countries including Ethiopia (Gildemacher et al., 2009).

In central highlands of Ethiopia in particular and in the country in general, potato area coverage and production cycle is increasing owing to increased use of irrigation facilities and economic importance of the crop. However the Ethiopian, potato breeding program has so far failed to develop varieties with a shortest dormancy periods, which can fit the production cycle of most farmers. Most potato varieties released by research institutions in Ethiopia have suffered low rates of adoption by farmers, especially for irrigated agriculture and low moisture stress areas, due to long dormancy periods and other tuber quality attributes. Medium to long dormancy genotypes are thus not easy to incorporate into the predominant cropping system in which farmers retain seed from the previous harvest for replanting the next season (Abebe, 2010).

Productivity of good seed tubers in potatoes relies on factors like; day length, temperature, physiological age of

seed tubers, plant density, nitrogen, water supplying, and finally growth regulating materials or plant growth regulators (PGR) (Gregory, 1965). Plant growth regulators have considerable effects on tuber fertility and it is highly related to hormonal balance (Stuart and Cathey, 1961; Vreugdenhil and Struik, 2006). By treating the tubers using gibberellic acid, the tubers can sprout faster (Burton, 1989). Gibberellins are able to break dormancy of potato tubers by dipping (pre-soaking) the seed tubers or spraying the potato plants (Lorreta et al., 1995; Rappaport et al., 1957; Vreugdenhil and Sergeeva, 1999). More buds can be generated per unit area by using gibberellic acid (GA₃) in potatoes. It has been observed that sprouting management strategies are especially needed in production conditions where storage lasts several months and the growing season is short. This is especially the case of seed and ware potato production areas. Furthermore, farmers generally use poor tuber sprouting techniques, which also contribute to poor supply of quality seed for year-round potato production (Nigussie, 2011). The improvements of crop productivity in modern agricultural systems are increasingly dependent on manipulation of the physiological activities of the crop by chemical means (Subhadrabandhu et al., 1999). The management of potato tuber dormancy is of great importance for the ware, packing and processing markets and also for the seed industry. After harvesting, potato tuber is naturally dormant for 1 to 15 weeks depending on the cultivar and storage conditions (Wiltshire and Cobb, 1996).

Depending on the intended purpose, accelerated (that is, seed tubers) or delayed (that is ware potato, industrial processing) sprouting of the harvested tubers is favorable. As with many aspects of plant development, plant hormones have been playing a primary role in the regulation of potato tuber endo-dormancy (Rappaport and Wolf, 1969; Hemberg, 1985). Four of the five principal classes of plant hormones abscisic acid, cytokinins, gibberellic acid, and ethylene have been implicated in dormancy regulation (Hemberg, 1985; Suttle, 1996, 2004). It has been hypothesized that dormancy is regulated by the relative concentrations of growth promoters and inhibitors. Gibberellins and cytokines are generally considered to be growth promoters, whereas abscisic acid and ethylene are believed to inhibit sprout growth (Sonnewald, 2001). Endogenous hormones have been proposed to play a significant role in tuber dormancy regulation (Bruinsma et al., 1967). The level of endogenous GAs remains low during the middle period of storage (deep dormancy) and increase near the onset of dormancy (Rappaport et al., 1958). Thus, exogenous application of GA₃ is used to break potato tuber dormancy (Hemberg, 1985) and it is commercially used to break dormancy of potato tuber. Dipping or soaking of tuber in to GA₃ solution on wounded tuber break the dormancy of tuber (Lippert et al., 1958). According to Van Ittersum et al. (1993) and Alexopoulos et al. (2008), GA₃ is also applied on haulm to shorten the dormancy of

potato and stimulate sprout initiation in a short period of time. Moreover, haulm application of GA₃ two weeks prior to vine killing and dipping the tubers in GA₃ solution shorten the dormancy period (Lippert et al., 1958). The use of chemicals to regulate dormancy is a common practice in many countries. Being environmentally friendly and less toxic, GA₃ treatment is widely used in many countries for breaking tuber dormancy (Alexopoulos et al., 2008). Lack of good quality seed among growers is the major problem adversely affecting the expansion of potato production in many developing countries (Shibairo et al., 2006). One major problem facing production of quality potato seed is poor sprouting, due to dormancy, which leads to delayed planting and poor crop emergence and vigor (Wiersema, 1985).

Timely availability of well-sprouted seed tubers at the on-set of rain as well as for irrigated potato production is a prerequisite for attaining proper planting materials which leads to high yields. Due to unavailability of sprouted tubers for planting at desired time, small-scale farmers often promote potato sprouting by placing them in pits, sacks, teff straw and trenches and use genotypes with short dormancy. However, the availability of potato genotypes with short dormancy period is scarce while medium to long dormancy genotypes are thus not easy to incorporate in the predominant cropping system in which farmers retain seed from the previous harvest for replanting the next season. Farmers mostly prefer various traditional storage methods to enhance sprouting. Potato seeds sprouted in traditional ways are, however, of poor quality due to apical dominance, rotting and sprout etiolation caused by the dark conditions. Under Ethiopian condition, the utilization of chemicals to regulate potato dormancy is not common. This attributed to the lack of information regarding suitable chemicals, and their methods, rates, and time of application for efficient use. Hence, introduction of chemical that induced dormancy breaking is vital to have early seed planting materials. Therefore, this research aimed to determine the optimum method and rate of GA₃ application to shorten potato tuber dormancy using improved potato variety having long dormancy periods, under diffused light storage (DLS).

MATERIALS AND METHODS

Description of the study sites

The tuber dormancy breaking experiment was conducted during 2008 to 2009 cropping seasons at Holetta Agricultural Research Centre, which is located in the Oromia National Regional State and about 29 km far from Addis Ababa in west direction. The site lies at 9° 00' N latitude, 38° 30' E longitude and with an elevation of 2400 m in central Ethiopia. The daily average minimum and maximum temperatures of the area during the growing seasons (2005-2015) were 6.42 and 27.2°C, respectively, and the mean annual rainfall was 918.31 mm. The soil of the experimental site is Nitisols, which is characteristically reddish to brown in color. It has soil pH of 6.67

and clay in texture with contents of 62.5% clay, 30.0% silt and 7.5% sand. The soil has organic matter content of 2.18%, and total nitrogen, available phosphorus and exchangeable potassium contents of 0.18%, 30.58 ppm and 0.14 meq 100 g⁻¹ soils, respectively (Kidest et al., 2019).

Description of experimental materials and design

Potato cultivar, Gera which was nationally released in 2003 with a yielding potential of 25 tons/ha and having extended dormancy period of more than three months and having large, round and white tubers with deep eyes was used for the experiment. The experimental plots size of 3 m × 3 m was arranged in a Randomized Complete Block Design (RCBD) with three replications. Forty medium sized (35-45 mm) and well sprouted tubers were planted at a spacing of 75 cm and 30 cm between rows and between plants, respectively, in four rows having ten plants each. Phosphorus (P) was applied at the rate of 92 kg ha⁻¹ in the form of P₂O₅ and Nitrogen (N) at the rate of 110 kg ha⁻¹ as urea. The entire rate of P and the half rate of the N fertilizers were applied at the time of planting. The remaining half of the N was side dressed 45 days after planting. As a crop protection measure Ridomil® MZ 68% WP was sprayed twice at a rate of 2 kg ha⁻¹ before the occurrence of late blight to control the disease. Other cultural practices such as cultivation, weeding and earthing up were carried out according to the research recommendation (Lemaga et al., 1994).

Treatments

In this experiment, a week before harvesting (98 days after planting), from each plot, ten plants from the central rows were tagged and treated with the different rates (0, 250, 500, 750 and 1000 ppm) of GA₃ as a foliar application. The stock solution of GA₃ was prepared by dissolving a total of 3 g GA₃ (90% gibberellins A₃ Biochemical, BDH Limited Poole England) in 10 ml of ethanol (96%) and the final volume was made up to 1000 mL with double distilled water (DDW). The solution was applied as a fine spray by using a manual sprayer early in the morning to avoid rapid drying of the spray solution, due to transpiration until the solutions run off all plants.

Similarly, at harvest freshly harvested forty medium sized (35-45 mm) and healthy tubers were selected and dipped into the different concentrations (0, 10, 20, 30, 40 and 50 ppm) of GA₃ solution for 24 h. The stock solution of GA₃ was prepared by dissolving 3 g GA₃ (90% gibberellins A₃ Biochemical, BDH Limited Poole England) in 10 mL of ethanol (96%) and the final volume was made up to 2000 ml with double distilled water (DDW). The control tubers were treated with ethanol and double distilled water (DDW) only for the same duration (AL-Qesi, 1996).

Storage trial

To determine the effect of GA₃ on dormancy and sprout growth, ten uniform medium sized (35-45 mm) tubers for each treatment (foliar sprayed and dipped) were selected, labeled and stored in a naturally ventilated diffused light store being arranged in a randomized complete block design with three replications. Tubers were monitored every other day and continued until 95% of the tubers get sprouted. During the storage period, the internal temperature and relative humidity of the storage room was recorded every day using thermo hygrometer. The mean minimum and maximum temperatures in the store were 3.2 and 21.3°C, respectively and the average relative humidity was 62.4%.

Characteristics studied

Storage trial

Dormancy period: Dormancy period was counted as the number of days from dehauling (haulm cutting) to sprouting of 80% of the tubers with at least one sprouts longer than 2mm.

Average number of sprouts per tuber: Average number of sprout per individual tubers was recorded when 95% the tubers sprouted (110 days after harvesting).

Average sprout length (mm): The mean length of each sprouted tuber that emerged from individual tubers was measured when 95% of the tubers sprouted (110 days after harvesting).

Fresh mass of sprout (mg): When 95% of the tubers sprouted (110 days after harvest) tubers were de-sprouted and fresh weight of sprouts were recorded using sensitive balance.

Dry mass of sprout (mg): After measuring the fresh weight, the sprouts were dried at 70°C to constant mass in an oven and dry mass of sprout was recorded.

Weight loss of the tubers (%): Calculated from the difference in final weight of tubers selected, labeled and stored when 95% the tubers sprouted (110 days after harvesting) to initial weight.

Statistical analysis

The data was analyzed using analysis of variance (ANOVA) and treatment means were separated by Least Significant Differences (LSD) at 5% probability level by using SAS statistical software packages version 9.00 (SAS, 2010).

RESULT AND DISCUSSION

Dormancy break experiment

Dormancy period

Highly significant differences ($P < 0.01$) was found among the treatments with regard to tuber dormancy period (Table 1). The data showed that all haulm applications and dipping of GA₃ significantly reduced tuber dormancy period below the control with more reduction when the concentration of GA₃ increased. Haulm application of 750 and 1000 ppm GA₃ reduced the duration of tuber dormancy by about 24 and 27 days, respectively after harvesting as compared to control and other treatments. Similarly, tubers treated with 40 and 50 ppm GA₃ solutions reduced the duration of tuber dormancy by about 18 and 20 days, respectively. Moreover, low GA₃ concentration (10 ppm) used as dipping resulted in 8 days of reduction as compared to untreated tubers. In accordance with the current results (Dogonadze et al., 2002) reported that GA₃ treatment immediately after harvesting reduced the duration of tuber dormancy by 38 to 42 days. These authors indicated that GA₃ is involved in breakage of dormancy and growth stimulation. Alexopoulos et al. (2008) showed that a haulm application

of GA₃ just before haulm destruction, shortened the dormancy period of the harvested seed tubers up to three months. In line with this study, a foliar spray of gibberellic acid, 3 to 6 days before haulm killing shorten potato tuber dormancy period and induced sprouting (Van Ittersum et al., 1993). In addition, Shibairo et al. (2006) reported that postharvest application of gibberellins to tubers grown from seed potatoes promotes the breakage of dormancy. Coleman (1987) also reported that exogenous gibberellins (GA) generally terminate dormancy in potatoes and may play important roles as endogenous regulators of bud dormancy and development. This is also in agreement with several other reports (Kim et al., 1996; Claassens and Vreugdenhil, 2000).

Average number of sprout per tuber

Highly significant differences ($P < 0.01$) was found among treatments with regard to the number of sprout per tuber as presented in Table 1. Regardless of the concentration, all haulm application treatments and dipping of tubers in 40 and 50 ppm GA₃ gave significantly higher sprout number compared to the control. In line with the current investigation, Alexopoulos et al. (2008) reported that, sprout number of tubers from plants sprayed with high GA₃ concentration was significantly higher than that of tubers from control plants. Similarly, Shibairo et al. (2006) found that irrespective of the concentration, GA₃ treatments (1, 5, 10 and 50 mg/L) significantly increased the number of sprouting buds per tuber compared to the control. GA₃ treatments increase the number of sprouts (Van Hiele, 1961; Ezekiel and Singh, 2005; Alexopoulos et al., 2007a; Otroshy and Struik, 2008) the length of the sprouts (Bruinsma and Swart, 1970) and proportion of sprouts by about 10% (Holmes et al., 1970). Similarly, Demo (2002) showed that increase in GA₃ concentration led to increase in sprouting percentage, number of sprouts per tuber, sprout length and sprout vigor. Results of the current experiment indicated that both haulm application and dipping treatments of GA₃ increased number of sprouts and sprouting capacity and a good alternative to improve potato seed quality to be utilized as for next planting.

Average sprout length per tuber

Average sprout length was significantly ($P < 0.01$) influenced by GA₃ treatments (Table 1). Applications of GA₃ at a rate of 500, 750 or 1000 ppm increased sprout length by about 91% as compared to the control. Similarly, in reference to the control (48 mm long) about 83% sprout length increment was obtained in response to dipping of the tubers in 40 or 50 ppm GA₃ solution. The data revealed that both haulm application and dipping treatment with GA₃ showed increasing trend of sprout length with increasing the rate of GA₃. In agreement with

Table 1. Effects of treating seed tubers with gibberellic acid on length of dormancy period, average sprout number and length of potato tuber.

Treatment	Dormancy period (days)	Average sprout number per tuber	Average sprout length (mm)
Control (Ethanol and DDW)	106.00 ^a	2.00 ^d	48 ^d
Haulm application of 250 ppm GA ₃	94.67 ^b	4.00 ^{abc}	80 ^b
Haulm application of 500 ppm GA ₃	85.33 ^{cd}	4.00 ^{abc}	90 ^a
Haulm application of 750 ppm GA ₃	82.33 ^{de}	4.33 ^{ab}	92 ^a
Haulm application of 1000 ppm GA ₃	79.00 ^e	5.33 ^a	93 ^a
Dipping tubers in 10 ppm of GA ₃	98.33 ^b	2.67 ^{cd}	55 ^d
Dipping tubers in 20 ppm of GA ₃	97.67 ^b	3.00 ^{bcd}	70 ^c
Dipping tubers in 30 ppm of GA ₃	95.67 ^b	3.33 ^{bcd}	80 ^b
Dipping tubers in 40 ppm of GA ₃	87.67 ^c	3.67 ^{bc}	86 ^{ab}
Dipping tubers in 50 ppm of GA ₃	86.00 ^{cd}	4.00 ^{abc}	88 ^a
Mean	91.27	3.63	78
CV (%)	2.34	20.58	5.31
Level of significance at p ≤ 0.05	0.0001	0.0001	0.0001

Means within a column followed by the same letters are not significantly different at the prescribed level of significance. ** = significant at 1% probability level, LSD (0.05)-Least significant difference at 0.05 probability level, CV (%) -Coefficient of variation in percent.

current results, Alexopoulos et al. (2007b) observed that the mean sprout length per tuber following treatment with GA₃ or GA₃ + BA was significantly higher than that of the controls. Similarly, Bruinsma et al. (1967), Alexopoulos et al. (2007b) showed that sprout length on seed tubers increases by exogenous application of GA₃. In addition, Bruinsma and Swart (1970) reported that mini-tubers treated with gibberellic acid were effective in increasing the length of sprouts. Lim et al. (2004) also noted that tubers of GA₃ treated showed fast sprout growth and potato tubers treated with higher dose of GA₃ (150 mg/L) sprouted earlier than other treatments. Thus, GA₃ treatments resulted in high sprout growth rates possibly due to an increase in assimilate flow towards the growing sprouts.

Fresh and dry mass of sprouts

Highly significant difference (P<0.01) was found among treatments with regard to fresh and dry mass of sprout per tuber (Table 2). Haulm application of GA₃ at 1000 ppm resulted in the highest fresh mass (1040 mg) of sprout which was 103% more as compared to the control. In the same way, foliar spray of GA₃ at a rates of 500, 750 or 1000 ppm and dipping in 50 ppm of GA₃ brought about 78 and 52% dry sprout mass increment compared to the control (112 mg). In agreement to this study, Lim et al. (2004) found that the fresh weight of sprouts per tuber following treatment with GA₃ or GA₃+BA was significantly higher than that of the controls. Therefore, increased concentration of GA₃ treatment increased growth of sprouts per tuber and also the rate of transfer of dry

matter from tuber to sprout and final improved sprout dry matter. In the current study, fresh and dry mass of sprouts were positively correlated with average sprout number ($r = 0.97^{**}$; $r = 0.81^{**}$ at (P<0.01) and sprout length ($r = 0.98^{**}$; $r = 0.86^{**}$ at (P<0.01) indicating that GA₃ treatment increased sprout mass by increasing both sprout number and length.

Percentage weight loss of tubers

Highly significant differences (P<0.01) was found among treatments with regard to percentage weight loss of the tubers (Table 2). The percent weight loss of tubers were significantly higher in the tubers treated with 1000 ppm (92.8%) followed by 750 ppm (78.7%) of GA₃ as compared to the control (58.2%). In the present study, tubers weight loss may be due to water loss, utilization of reserve carbohydrates by newly emerging sprouts and respiration of mother tubers. In agreement with this study, Alexopoulos et al. (2007a) reported that GA₃ treated tubers showed significantly higher weight loss as compared to untreated tubers. The authors described that, the higher weight loss in GA₃ treated tubers in the first 7 days after treatment could be attributed to the incisions made to facilitate the entry of GA₃. Similarly, Burton (1989) and Shibairo et al. (2006) indicated that sprouting is accompanied by many physiological changes including increases in reducing sugar content, respiration, water loss, and glycoalkaloid content and also mentioned that GA₃ increased the rate of weight loss as compared to untreated tubers. The higher weight loss in GA₃ treated

Table 2. Effects of seed tubers treatment with gibberellic acid on sprout fresh and dry mass, and weight loss percentage of tubers.

Treatment	Fresh mass of sprout per tuber (mg)	Dry mass of sprout per tuber (mg)	Percentage weight loss per tuber (%)
Control (Ethanol and DDW)	510 ^e	112 ^c	58.2 ^f
Haulm application of 250 ppm GA ₃	750 ^{bcd}	174 ^b	71.5 ^{cd}
Haulm application of 500 ppm GA ₃	780 ^{bc}	182 ^{ab}	75.2 ^{bc}
Haulm application of 750 ppm GA ₃	820 ^b	191 ^{ab}	78.7 ^b
Haulm application of 1000 ppm GA ₃	1040 ^a	224 ^a	92.8 ^a
Dipping tubers in 10 ppm of GA ₃	600 ^{de}	145 ^{bc}	62.5 ^{ef}
Dipping tubers in 20 ppm of GA ₃	630 ^{cde}	149 ^{bc}	66.5 ^{de}
Dipping tubers in 30 ppm of GA ₃	640 ^{cde}	150 ^{bc}	70.0 ^{cd}
Dipping tubers in 40 ppm of GA ₃	730 ^{bcd}	170 ^b	70.5 ^{cd}
Dipping tubers in 50 ppm of GA ₃	730 ^{bcd}	171 ^b	71.3 ^{cd}
Mean	720	170	71.7
CV (%)	12.09	14.77	4.81
Level of significance at (p ≤ 0.05)	0.001	0.001	0.001

Means within a column followed by the same letters are not significantly different at the prescribed level of significance. ** = significant at 1% probability level, LSD (0.05)-Least significant difference at 0.05 probability level, CV (%)=Coefficient of variation in percent

tubers may be due to the higher rate of metabolism which is associated with sprout initiation and growth (Reust, 1986). Burton (1989) stated that treating potato tubers with GA₃ or GA₃ + BA caused an increase in weight loss and respiratory activity. The author further indicated that, the higher rates of weight loss and respiration in tubers from GA₃ treated plants probably resulted from the presence of sprouts on these tubers.

Conclusion and Recommendation

In Ethiopia, potato is the fastest growing major crop in the developing world with important economic impact on many resource-poor farming families. However, in Ethiopia, the yield per unit area of potato is very low compared to those of other countries. It is one of the most widely used root and tuber crop in human diet. It is also an important cash crop for farmers in the mid and highlands of the country, where it is grown abundantly. However, lack of quality planting materials among growers is a limiting factor adversely affecting production and productivity in these areas. Most potato producer farmers generally use poor tuber sprouting techniques, which also contribute to poor supply of quality seed for year-round potato production. Thus, bringing potatoes to a warm place was used by more than 20% of the farmers in Ethiopia, while also putting the seed potatoes in bags was frequently mentioned. Conversely, identifying appropriate management practices to improve the quality of planting materials is a priority to introduce plant growth regulator for potato producers. Thus, both haulm

application and dipping methods of treatments have effect on breaking dormancy, early emergence of shoots. Haulm applications of GA₃ at 750 and 1000 ppm reduced dormancy period by 24 and 27 days, respectively. It also hastened early physiological maturity, increased average sprout number and sprout length of tubers, respectively. Similarly, dipping treatment of 40 and 50 ppm reduced dormancy period by 18 days and 20 days, respectively, and had more effect over the control than lower concentrations. Haulm application of 750 or 1000 ppm reduced days to emergence by 11 days while dipping of seed tubers in 40 and 50 ppm reduced days to emergence by 6 and 8 days, respectively. Therefore, GA₃ affected the physiological age of the tubers by inducing the breakage of bud dormancy at all stages of tuber growth. Although the experiment was conducted in one location and season using a single cultivar it is reasonable to point out that foliar application of gibberellic acid one week before harvest and dipping tubers resulted in shortened dormancy period, increased sprout mass and improved both yield and quality of the subsequent potato generation.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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Full Length Research Paper

Comparative qualitative phytochemical analysis of oil, juice and dry forms of garlic (*Allium sativum*) and different varieties of onions (*Allium cepa*) consumed in Makurdi metropolis

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The qualitative phytochemical analysis of oil, juice and dry forms of garlic and three species of onions (red, brown and white onions) was investigated. Samples of fresh and dried garlic, red, brown and white onions were prepared and analyzed. All the samples were analyzed for the presence of phytochemicals which include saponins, flavonoids, phenols, tannins, triterpenoids, cardiac glycosides, steroids and alkaloids. Results of the juice samples showed that garlic has the lowest phytochemical content while red and brown onions recorded the highest and similar phytochemical contents. All the samples gave positive results for saponins and cardiac glycosides. For the dry samples, garlic had the lowest phytochemicals while the red and brown onions had the highest. The oil extracts of garlic and white onions had lower phyto-nutrients while red and brown species still possessed higher phyto-nutrients with steroids and saponins present in all samples. This study concludes that brown onions has the highest phytonutrients in all forms and are recommended as the most preferred option in health and disease.

Key words: Flavonoid, tannins, saponin, phytochemical, onions, garlic, Makurdi.

INTRODUCTION

Plants have over time served mankind as sources of useful drugs, food, additives, flavouring agents, colourants, binders and lubricants (Falodun et al., 2006). Medicinal plants are sources of important drugs for

the treatment of diseases either alone or in combination with other plants (Awonubi, 1988). Chemical compounds found in plants include alkaloids, glycosides, essential oils, saponins, tannins, steroids, terpenoids, resins,

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flavonoids, proteins and others. These chemical compounds are potent bioactive compounds found in parts of medicinal plants which are useful for therapeutic purposes (Soforowa, 1993; Nwadiaro and Nwachukwu, 2007). These inherent bioactive principles differ among plants owing to their biodiversity and as such they produce a definite physiological effect on the human body. Banso and Olutimayin (2001) have shown that plants comprise of a wide variety of active principles. Some of the bioactive principles of plant origin possess antimicrobial properties. Thus, the knowledge of medicinal plants is important in pharmaceutical industry and this supports their use as base for the development of new drugs.

The increased reliance on the use of medicinal plants in industrialized countries had been traced to the extraction and development of several drugs and chemotherapeutics from these plants as well as from traditionally used rural herbal medicine (UNESCO, 1998).

Allium sativum L., is commonly called garlic. It is a perennial plant widely cultivated and belongs to the family Alliaceae and genus *Allium*. Garlic has long, flat grass like leaves and a papery hood around the flowers (Milner, 2005). The greenish white or pink flowers are grouped together at the end of a long stalk. The stalk rises directly from the flower bulb, which is the part of the plant used as food and medicine (Awonubi, 1988). The most active components of fresh garlic are alliin and an enzyme called allinase. When garlic is chewed, chopped, bruised or cut, these compounds mix to form allicin, which is responsible for garlic's strong smell. Its medicinal claims have included cures for coughs, cold, toothaches, some viral infections and open wounds (Fluck, 1973).

A. sativum Linn has been known to possess both dietary and medicinal properties (Ross et al., 2001), and it is proven to have antimicrobial effects (Reuter et al., 1996). The plant also possesses phytochemical constituents (Cavallito and Bailey, 1944). The antimicrobial properties of garlic were first described by Louis Pasteur in 1958, and since then, research had demonstrated its effectiveness against bacteria, protozoa, fungi and some viruses (Jaber and Al-Mossawi, 2007). In its pure form, it has been found to exhibit antibacterial activity against a wide range of Gram-negative and Gram-positive bacteria, including multidrug-resistant enterotoxigenic strains of *Escherichia coli* and also possesses antifungal activity, antiparasitic, and antiviral activity (Ross et al., 2001; Pai and Platt, 1992). The antimicrobial activity of garlic has been attributed to the presence of allicin (a thiosulphinates) whose removal completely renders garlic ineffective against microorganisms (Hughes and Lawson, 1991). Allicin, the main active principle related to *A. sativum* chemistry is obtained by crushing or cutting garlic cloves. The odourless amino acid, alliin present in the garlic cloves, is metabolized by the enzyme allinase (a cysteine sulphoxidelyase) to allicin and other thiosulphinates,

which besides their antimicrobial effect; produce the characteristic odour of garlic (Block, 1985). Allicin is considered to be responsible for the bacteriostatic properties of garlic. *A. sativum* extracts obtained with ethanol (ethanolic garlic extract, EGE) and acetone (acetonic garlic extract, AGE) extracted by drying at 60°C (Eja et al., 2007), and by Soxhlet apparatus (EL-Mahmood, 2009) was claimed to have direct implication in the inhibition of the *in-vitro* growth of gram positive, gram negative and diarrhoeagenic bacteria responsible for serious gastric diseases such as ulcers and even gastric cancer.

Allium cepa commonly known as onions have been valued for their medicinal qualities by many cultures around the globe. Numerous health benefits have been attributed to the vegetable, including prevention of cancer and cardiovascular disorders. Sequel to this, studies on specific compounds found in onion bulbs have been carried out (Block, 1985). Onions have a unique combination of three families of compounds that are believed to have effects on human health. They include fructans, flavonoids and organosulphur compounds. Fructans are small carbohydrate molecules that help maintain gastrointestinal health by sustaining beneficial bacteria (Eja et al., 2007). Block(1985) and Eja et al.(2007) have focused on a flavonoid- quercetin, which is found at particularly high levels in onions. It functions as an antioxidant, deactivating molecules that are injurious to cells in the body.

The organosulphur compounds are largely responsible for the taste and smell of onions. These compounds reduce symptoms associated with diabetes mellitus, inhibit platelet aggregation (involved in thrombosis) and prevent inflammatory processes associated with asthma (Hughes and Lawson, 1991).

Plant essential oils are generally of quite complex composition containing volatile components more or less modified during preparation. The volatile fractions of essential oils constitute 90–95% of the oil in weight, contains the monoterpene and sesquiterpene hydrocarbons as well as their oxygenated derivatives along with aliphatic aldehydes, alcohols, and esters (Eja et al., 2007).

Garlic and onions contain oils which are essential oils, consisting of chemical compounds which have hydrogen, carbon and oxygen as their building blocks. Essential garlic oil contains variety of sulphides such as diallyldisulphide and dillytrisulphide. During the process of distillation, allicin is completely eliminated from the oil. Commercially available garlic oil capsules generally contain vegetable oil and a small amount of garlic essential oil because of the pungent odors (Jaber and Al-Mossawi, 2007).

Regular consumption of garlic oil can reduce blood pressure; prevent heart disease including atherosclerosis, high cholesterol and cancer. Garlic oil is an effective antibiotic, anti-viral and anti-fungal agent, which could be

used to prevent nausea, diarrhea, ease coughs and could be used even in treatment of conditions such as malaria and cholera probably an immune system enhancement (Eja et al., 2007).

Despite the nutritional and vast health benefits of the allium species, these plants are not being used by some people due to their pungent odour and irritation to the eyes. Therefore, the research was aimed at ascertaining what form (oil, juice or dried) of onion or garlic which show the presence of most phytochemical constituent which could be subsequently processed, packaged and recommended as supplement in health and disease.

MATERIALS AND METHODS

Collection of samples

A. sativum and *A. cepa* samples of the three available types— red, brown and white onions were collected from Wurukum market and Wadata markets both in Makurdi metropolis, Benue State, Nigeria.

Processing of plant materials

The collected *A. sativum* bulbs collected were cleaned thoroughly. Part of it was oven dried. The dried bulbs were blended into fine powder and stored in airtight container at room temperature. Some of the bulbs were left for oil extraction.

The collected *A. cepa* bulbs were also cleaned thoroughly, part of it was made into juice for analysis and another part was left for oil analysis.

Preparation of extracts

The organic solvents such as petroleum ether, n-hexane, chloroform, methanol and distilled water were used for the extraction of active compounds from the *A. sativum* and *A. cepa* bulbs for six hours. The extraction of oil was done using soxhlet apparatus. For every 200 mL of the each solvent, 25 g of the crushed plant leaves powder was used for Soxhlet extraction. Onions and garlic were chopped into small pieces and mashed with a domestic blender. It was then loaded into a thimble and extracted using n-hexane.

Oil extraction

The extraction of garlic oil and onion oil was conducted with a soxhlet extractor using n-hexane (boiling point of 40 – 60°C) for 6 h. The oils were obtained after the solvent was removed under reduced temperature and pressure and refluxing at 70°C so as to remove any excess solvent used for the oil extracted. The extracted oils were stored in a refrigerator freezer at 2°C for subsequent analyses (Ameh et al., 2012). This method is called soxhlet extraction or hot continuous extraction.

Juice extract

Fresh garlic and onions were chopped and blended. The pulp was diluted with distilled water and then filtered using a muslin cloth and the filtrate was analyzed for the presence of phytochemicals according to the method described by El-Mahmood (2009).

Phytochemical analyses

The oil and the dried forms of garlic extracts were analyzed for the presence of different phytochemicals. In a similar way, the oil and juice from the different species of onions (red, brown and white) were analyzed for the presence of phytochemical compounds. The analysis of these phytochemical compounds was done based on the methods described by Gazuwa et al. (2013)

Phytochemical analysis on the oil, dry and juice of garlic and red, brown and white onion species was done using standard qualitative procedures; Dragendorf test for alkaloids, sodium hydroxide test for flavonoids, ferric chloride test for tannins, Salkowski test for cardiac glycosides, Liebermann-Burchard test for terpenes, general test for saponins and phenols etc.

Test for saponins

Three milliliters (3ml) of aqueous extract was diluted with 20 ml of distilled water and shaken in a graduated cylinder for 15 min. The formation of a layer of foam measuring about 1 cm indicates the presence of saponins (Singh and Chauhan, 2014).

Test for flavonoids

Two milliliters (2 ml) of dilute sodium hydroxide was added to 3 ml of extract in a test tube. A yellow solution that turns colourless on addition of concentrated HCl indicates the presence of flavonoids (Prashanth and Krishnaiah, 2014).

Test for phenolic compounds and tannins

Three millimeters (3 ml) of oil, juice and dried extracts were tested with 3% ferric chloride; the presence of a dark green colour indicates the presence of phenolic compounds. Also, about 2 ml of extract was tested with few drops of 10 % ferric chloride added gradually. A blue-black or blue-green precipitate shows the presence of tannins (Itelima et al., 2016).

Test for triterpenoids and steroids

Few drops of concentrated H₂SO₄ were added to a small portion of aqueous extract in a test tube and shaken. Formation of a yellow colored layer indicates the presence of triterpenoids while a red colour indicates the presence of steroids (Prashanth, Krishnaiah, 2014).

Test for cardiac glycosides

A small portion of aqueous extract was poured in a test tube, 2 ml of glacial acetic acid, a drop of ferric chloride solution and 2 ml of conc. H₂SO₄ were added. A brown ring formation at the interphase indicates the presence of cardiac glycosides (Singh and Chauhan, 2014).

Test for alkaloids

A small portion of aqueous extract was poured in a test tube, few drops of 1% HCl were added followed by the addition of Mayer's reagent. Formation of a white or creamy precipitate indicates the presence of alkaloids (Prashanth and Krishnaiah, 2014).

Table 1. Phytochemical analysis of juice forms of garlic and three onion species.

Phytochemical	Garlic	Red onions	Brown onions	White onions
Flavonoids	-	+	+	+
Phenols	-	+	+	+
Tannins	-	+	+	+
Triterpenoids	-	+	+	-
Steroids	-	-	-	-
Cardiac glycosides	+	+	+	+
Alkaloids	+	-	-	-
Saponins	+	+	+	+

+, Present; -, Absent.

Table 2 Phytochemical analysis of the dry forms of garlic and the three onion species.

Phytochemical	Garlic	Red onions	Brown onions	White onions
Flavonoids	-	+	+	-
Phenols	-	+	+	+
Tannins	-	+	+	+
Triterpenoids	-	-	-	+
Steroids	+	-	+	-
Cardiac glycosides	+	+	+	-
Alkaloids	-	-	-	-
Saponins	+	+	+	+

+, Present; -, Absent.

Table 3. Phytochemical analysis of oils from the samples.

Phytochemical	Garlic	Red onions	Brown onions	White onions
Flavonoids	-	+	+	-
Phenols	-	+	+	-
Tannins	-	+	+	-
Triterpenoids	-	+	+	-
Steroids	+	+	+	+
Cardiac glycosides	+	+	+	+
Alkaloids	+	-	-	+
Saponins	+	+	+	+

+, Present; -, Absent.

RESULTS

The results of the qualitative phytochemical analysis for the juice forms of garlic and three onion species (red, brown and white onions) were analyzed.

Result from Table 1 shows that red and brown onion juice had a greater distribution of phytochemicals when compared with other samples. Garlic juice had the lowest distribution of phytochemicals. Saponin and cardiac glycosides was present in all samples while steroids was absent in all juice samples.

Results from Table 2 shows that dry brown onions had a better distribution of phytochemicals and was followed closely by dry red and white onion samples respectively. Dry garlic recorded the lowest phytochemical distribution. Saponin was present in all samples while alkaloid was absent in all samples.

From Table 3, oil samples of red and brown onion had the same phytochemical composition and distribution when compared with white onions and garlic. Steroids, cardiac glycosides and saponins were present in all oil samples.

Table 4. Comparison of the different forms of garlic samples.

Phytochemical	Juice	Dry	Oil
Flavonoids	-	-	-
Phenols	-	-	-
Tannins	-	-	-
Triterpenoids	-	-	-
Steroids	-	+	+
Cardiac glycosides	+	+	+
Alkaloids	+	-	+
Saponins	+	+	+

+, Present; -, Absent.

Table 5. Comparison of the different forms of red onions.

Phytochemical	Juice	Dry	Oil
Flavonoids	+	+	+
Phenols	+	+	+
Tannins	+	+	+
Triterpenoids	+	-	+
Steroids	-	-	+
Cardiac glycosides	+	+	+
Alkaloids	-	-	-
Saponins	+	+	+

+, Present; -, Absent.

Table 6. Comparison of the different forms of brown onions.

Phytochemical	Juice	Dry	Oil
Flavonoids	+	+	+
Phenols	+	+	+
Tannins	+	+	+
Triterpenoids	+	-	+
Steroids	-	+	+
Cardiac glycosides	+	+	+
Alkaloids	-	-	-
Saponins	+	+	+

+, Present; -, Absent.

Table 4 shows the comparative phytochemical distribution of all forms (juice, dried and oil) of garlic samples. Flavonoids, phenols, tannins and triterpenoids were absent in all forms. Cardiac glycosides and saponins were present in all forms.

From Table 5, flavonoids, phenols, tannins, Cardiac glycosides and saponins were present in all forms (juice, dried and oil) of red onion samples while alkaloids were absent in all red onion samples.

Results from Table 6 reveals the presence of flavonoids, phenols, tannins, cardiac glycosides and saponins in juice, dry and oil samples of brown onions. Alkaloids was absent in all samples.

From Table 7, juice, dry and oil samples of white onions showed varying presence of phytochemicals. Phytochemicals were present more in the juice than in the oil and dry forms. Saponin was present in all samples.

Table 7. Comparison of the different forms of white onions.

Phytochemicals	Juice	Dry	Oil
Flavonoids	+	-	-
Phenols	+	+	-
Tannins	+	+	-
Triterpenoids	-	+	-
Steroids	-	-	+
Cardiac glycosides	+	-	+
Alkaloids	-	-	+
Saponins	+	+	+

+, Present; -, Absent.

DISCUSSION

Garlic, red, brown and white onions showed the presence of varying number of phytochemicals. This confirmed a similar qualitative work on garlic and onions by Gazuwa et al. (2013). Comparing the juice from the four samples, as seen in Table 1, garlic had the lowest number of phytochemicals and was closely followed by white onions. Red and brown onions showed similar phytochemical contents and both recorded the highest of the four samples. Steroids were absent in all samples while cardiac glycosides and saponins were present in all samples. These results are in tandem with studies carried out by Ameh et al. (2012). This result suggests a rich presence of phytonutrients with cardioprotective and antihypertensive potentials. Cardioprotective and antihypertensive properties of garlic were reported by Asdaq and Inamdar (2011) and Bhandari (2012). Cardioprotective and antihypertensive properties of onions was confirmed by Lanzotti (2006).

From Table 2, garlic also had the lowest number of phytonutrients followed by red and white onions with brown onions having the highest number of phytonutrients. Here, triterpenoids and alkaloids were absent in all samples while saponins were present in all samples. This is in consonance with studies done by Eja et al. (2007). This suggests the presence of phytochemicals with antimicrobial, antifungal, hypolipidemic and anticancer potentials. These phytochemicals have been reported by Sengupta et al. (2004) to have anticancer properties. Dried brown onions possessed the highest number of phytochemicals and may be considered as an antibiotic treatment option above others. The antimicrobial activity of allium species have been reported (Reuter et al., 1996; Ross et al., 2001; Bhandari, 2012).

The results of the oil samples from Table 3 shows garlic and white onions had lower number of phytonutrients when compared with red and brown onions which recorded the highest number of phytonutrients. Research has proven that Allium oils have high antioxidant and antibacterial activity (Mnayer et al.,

2014). Steroids, cardiac glycosides and saponins are present in all the samples. This conforms to the work done by Dronet al. (1997), thus, suggesting a very rich amount of phytonutrients with heart health benefits, anti-cancer potentials and hypolipidemic potentials. Red and brown onions oils can therefore be recommended as preferred sources of nutrients for remedy of these diseases since they have the highest phytochemicals.

The results of the individual samples in their different forms are shown in Tables 4 to 7. Comparison was done for the three garlic samples. The result from Table 4 shows that garlic oil had the highest phytochemical content when compared with the juice and dry forms which recorded similar findings. Cardiac glycosides and saponin were present in all samples while flavonoids, phenols, tannins and triterpenoids were absent in all samples. Although, garlic in its different forms can be used as remedy for cardiovascular disease, hypertension and cancer due to their phytochemical content, garlic oil can be used as preferred source over the other forms. Orengo et al. (2016) also confirmed the presence of these active components in onion and garlic.

The results of Table 5 show that the oil sample of red onions had the highest number of phytochemicals followed by the juice, while the dry form had the least number of phytochemicals. Here flavonoids, phenols, tannins, cardiac glycosides and saponins are present in all forms while alkaloids are absent in all forms. This conforms to a study conducted by Dini et al. (2008).

Table 6 also shows the oil form of brown onions had the highest number of phytochemical nutrients while the juice and dry forms followed closely. Here also, flavonoids, phenols, tannins, cardiac glycosides and saponins are present in all forms while alkaloids is absent in all forms. Ogbonna et al. (2016) confirmed the presence of these phytochemicals in onion samples. Brown onion samples can therefore be used in similar manner as the red onions. Here also the oil sample had slightly more number of phytonutrients than the juice and dry samples.

Results from Table 7 show white onions juice had the highest number of phytochemical content when compared

with the dry and oil samples. These results support the findings of Dini et al. (2008). Saponin was present in all samples.

In comparison, the oil extracted from garlic contains slightly more number of phytochemicals than the juice and dry forms, likewise the red onions and the brown onions. However, the juice form of the white onions had slightly more number of phytochemicals than the oil and dry forms. Different authors have linked the antimicrobial analysis of plants to the presence of phytochemicals. Nwadiaro and Nwachukwu (2007) linked the antimicrobial activity of plants to the presence of tannins, alkaloids, flavonoids and saponins. Also, qualitative phytochemical analysis of garlic and onions has been done by different researchers with positive results for different phytonutrients.

Conclusion

The results of these analyses show that both garlic and onions possess varying number of phytochemicals. The juice and dry forms of brown onions had greater number of phytochemicals followed by red, white onions and garlic respectively. The oil of brown and red onions had the highest number of phytochemicals. The oil of garlic, red and brown onions had the highest number of phytochemicals while the juice form of white onions had the highest number of phytonutrients. This study thus concludes that brown and red onions had the highest number of phytonutrient.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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Full Length Research Paper

Impact of pesticides on the growth of *Coriandrum sativum*

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Pest management is a severe constraint in agriculture as pests decrease the plant yield, productivity and also act as vectors causing various plant diseases. Pesticides are used to devour pests; however, irrational usage results in adverse impacts on the ecosystem. The present investigation was aimed to study the effects of fenvalerate, cypermethrin and chlorpyrifos on the growth of *Coriandrum sativum* for 35 days until the plants attained the flowering stage. The study was conducted in triplicates and various growth parameters such as germination rate, shoot height, biomass and moisture content of pesticide-induced plants were calculated and compared with control. On the 35th day, the plants displayed enhanced shoot heights under recommended pesticide dosage of 2.5 mL L⁻¹ for cypermethrin (11.41 ± 0.08 cm) and fenvalerate (12.43 ± 0.2 cm) rather than the control plants (9.12 ± 0.06 cm). However, detrimental effects on plant growth and early mortality were observed in chlorpyrifos treated plantlets. Student's t-test revealed a marked difference in plant growth under different pesticide concentration gradients at a 5% level of significance. Analysis of shoot heights and analysis of variance (ANOVA) concluded that there was no significant mean difference of plants grown under cypermethrin and fenvalerate stress at 5.0 and 7.5 mL L⁻¹ concentration with plants in control. Hence, the present study establishes an optimal pesticide range of 5.0 ± 2.5 mL L⁻¹, which did not prove injurious to the growth and productivity of *C. sativum*.

Key words: Chlorpyrifos, *Coriandrum sativum*, cypermethrin, fenvalerate, germination.

INTRODUCTION

In agriculture, various disease-causing organisms including insects, larvae, pathogenic fungi, viruses and weeds, severely affect the growth and productivity of crops. Organic or inorganic pesticides are xenobiotic

compounds that are routinely administered to debar pests from crops. Pesticides comprise fungicide, herbicide, nematicide, molluscicide, germicide, antimicrobial agents and insect or animal repellents (Randall et al., 2013). In

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Abbreviations: ANOVA, Analysis of variance; CNS, central nervous system; E.C., emulsifiable concentrate; EC, electrical conductivity; KVK, Krishi Vigyan Kendra; SPSS, Statistical Package for the Social Sciences; TA, Tatafen; TR, Tricel; US, Ustaad.

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most agricultural practices, pesticides are used to increase the yield, productivity and vigour of crops. Pimentel (1995) reported that only 0.1% of the applied pesticide acts against the pest, and 99.9% of the remaining affects the environment. De Oliveira et al. (2012) estimated that annually millions of tons of pesticides are applied to crop fields out of which less than 5% acts against the target organisms while the excess fraction (> 95%) of pesticides target the beneficial soil microflora and leads to the pollution of soil and water bodies (Alavanja, 2012; Carvalho, 2006; Freire et al., 2015; López-Pérez et al., 2006; Meyer et al., 2010; Power, 2010; Turra et al., 2010; Oliveira et al., 2016). India accounts for the maximum usage of insecticides (76%), fungicides (13%), herbicides (10%) and others (1%), as compared to 44% of insecticides usage globally. Most commonly used pesticides in Barak Valley, Assam are organophosphates (dimethoate, chlorpyrifos, monocrotophos, dichlorophos, profenofos); synthetic pyrethroid (fenvalerate, cypermethrin); organochlorides (endosulfan, DDT); and carbamates (carbofuran) (Dey et al., 2013). These pesticides are reported to enter the food chain by the process of biomagnifications affecting the consumers and farmers. González et al. (2011) found traces of pesticides in fruits, vegetable crops and even in other processed food products which signifies that the pesticides are non-recalcitrant compounds, and retain their toxicity even after detoxification and degradation processes. Pesticide sprays can directly hit non-target vegetation or can drift or volatilise from the treated area, and contaminate air, soil and non-target agents (Aktar et al., 2009). Due to extensive agricultural practises in the same cultivable land, pesticides which were applied to the previously grown crops gets accumulated in the soil by the process of leaching which proves detrimental for plant health and human consumption as they are hard to convert into less toxic forms by chemical and biological processes. As a result, high concentrations of pesticides hinder various biological processes of the plants causing chlorosis, nutrient imbalances, oxidative stress and decline in enzyme activity. It also affects seedling germination, reproductive health or flowering and yield.

The present study aims to understand the effect of the pesticides on the growth patterns of an essential annual herb Coriander (*Coriandrum sativum*), a plant from Apiaceae family, which is often regarded as the "spice of happiness" by Egyptians (Hameed et al., 2017). *C. sativum* is cultivated during the winter months, and maturation or flowering can be achieved within a month, it proved to be a perfect candidate for our study. Being rich in antioxidants, the plant has tremendous health benefits that include antispasmodic, aphrodisiac, hypolipidemic, hypoglycaemic, and antimutagenic properties. The leaves and seeds are commonly consumed which contains lots of mineral oils, and flavouring compounds like limonene, linalool, pinen, terpinen and geraniol (Mahendra and Bisht, 2011; Maroufi et al., 2010; Sahib et al., 2013). The

growth patterns of *C. sativum* were studied under three different pesticide application which includes TATAfen (fenvalerate 20% emulsifiable concentrate (E.C.) [a TATA product]; Tricel (Chloropyrifos 20% E.C.) [Excel Crop Care]; and Ustaad (cypermethrin 10% E.C.) [UPL]); which are extensively applied in the crop fields in this region. Fenvalerate falls under the category of synthetic pyrethroids which is applied against a wide range of pests and is of low toxicity to mammals. Short-time exposure to humans causes itching of skin and irritation of eyes, whereas prolonged exposure causes lead to neurological disorders and endocrine dysfunction (Umeda et al., 2016). Chloropyrifos is amongst the widely used organophosphate pesticide which is useful against all biting and chewing pests (Bozdogan et al., 2015). It is effective for longer durations and does not wash off in rainwater due to its higher persistence value and is very toxic to mammals, including humans (Mitran et al., 2017). Cypermethrin is a synthetic pyrethroid pesticide and insect repellent which is effective in killing many pests like aphids, thrips, mites, moths and is extensively used to protect vegetable crops. It is also reported to cause germ cell mutations and potential genotoxic effects in mice models (Aktar et al., 2009; Emam and Abdelhameed, 2017; Dey et al., 2013). Hence, the present study was aimed to understand the effects of three pesticides on the growth parameters of *C. sativum* and finally to establish the pesticide concentration range to which the plants show minimal distortion in growth patterns.

MATERIALS AND METHODS

Study site, sampling and soil analysis

The study was conducted during December to mid-February in the year 2016-2017 in Silchar, Assam (24° 48' N, 92° 47' E). The average temperature that prevailed during this session was 11 to 23°C, with no rainfall recorded during the span of the study. The soil sample was collected by random sampling method from the banks of Barak River, Silchar (24° 5' N and 92° 46' E) from a depth of 10 to 20 cm with no history of pesticide application ever. The soil samples were grinded, shade dried and sieved through 2 mm pore sieve. The soil sample was analysed for soil pH, electrical conductivity (mS cm^{-1}), texture, consistency, moisture content (%), bulk density (g mL^{-1}), particle density (g mL^{-1}), and porosity (%). Estimation of macronutrients (C, N, P, K) and micronutrients (S, Zn, B, Fe) contents were performed at Soil Testing Laboratory, Krishi Vigyan Kendra (KVK), Arunachal, Cachar, Assam (Iwara et al., 2011). The soil used for the present study was dark-brown coloured silt-clay-loamy soil with high moisture content ($23.41 \pm 4.36\%$), indicating that the soil was fertile with high organic content (Wall, 2005). The soil pH was recorded as 7.38 ± 0.07 (mildly alkaline), and electrical conductivity (E.C.) was $0.37 \pm 0.03 \text{ mS cm}^{-1}$, which means that the soil is critical for salt-sensitive crops (Mitran et al., 2017). Due to high organic content in the soil, the bulk density was low ($0.78 \pm 0.06 \text{ g mL}^{-1}$) and particle density was medium ($1.81 \pm 0.37 \text{ g mL}^{-1}$); however, the soil was highly porous ($56.21 \pm 8.64\%$), and also showed high drainage capacity and hydraulic conductivity. The presence of macronutrients (C, N, P, K) and micronutrients

Table 1. Physico-chemical properties of various soil samples of pot culture under concentration gradient of three different pesticides.

Sample	pH	E.C. (mS cm ⁻¹)	Carbon (%)	Nitrogen (kg ha ⁻¹)	Phosphorus (kg ha ⁻¹)	Potassium (kg ha ⁻¹)	Sulphur (mg kg ⁻¹)	Zinc (mg kg ⁻¹)	Boron (mg kg ⁻¹)	Iron (mg kg ⁻¹)
Control	7.40	0.42	2.02	692.90	120.80	343.70	9.62	5.04	0.81	12.30
US= Ustaad (Cypermethrin, 10% E.C.)										
US (2.5 mL L ⁻¹)	7.33	0.40	2.00	686.50	99.55	317.70	2.24	4.32	1.65	17.00
US (5.0 mL L ⁻¹)	7.35	0.37	1.97	680.10	96.44	270.20	5.11	1.44	1.01	8.40
US (7.5 mL L ⁻¹)	7.38	0.39	1.91	660.90	95.11	314.30	3.47	2.16	1.42	10.40
US (10.0 mL L ⁻¹)	7.45	0.31	1.97	680.10	92.01	317.70	3.88	3.96	0.73	6.94
US (12.5 mL L ⁻¹)	7.47	0.37	2.04	699.30	97.77	299.60	4.70	2.16	0.13	8.17
TA= Tatafen (Fenvalerate, 20% E.C.)										
TA (1.0 mL L ⁻¹)	7.32	0.33	1.95	673.70	75.15	334.60	5.93	1.80	0.18	7.28
TA (2.5 mL L ⁻¹)	7.27	0.35	1.91	660.90	79.59	315.40	5.11	2.88	0.23	5.82
TA (5.0 mL L ⁻¹)	7.32	0.32	1.88	654.40	85.35	327.80	3.47	2.52	0.04	8.96
TA (7.5 mL L ⁻¹)	7.37	0.37	1.95	673.70	90.23	323.30	2.65	3.60	0.27	15.60
TA (10.0 mL L ⁻¹)	7.42	0.41	1.93	667.30	89.79	339.10	0.00	1.80	0.36	7.28
TA (12.5 mL L ⁻¹)	7.46	0.42	2.00	686.50	94.22	331.20	0.00	0.72	0.82	8.96
TR= Tricel (Chloropyriphos, 20% E.C.)										
TR (1.0 mL L ⁻¹)	7.30	0.34	2.02	692.90	77.81	309.70	0.60	5.40	0.82	8.40
TR (2.5 mL L ⁻¹)	7.21	0.41	1.91	660.90	94.67	338.00	0.19	3.60	0.41	7.72
TR (5.0 mL L ⁻¹)	7.38	0.36	1.97	680.10	87.13	330.10	1.01	1.08	0.38	9.96
TR (7.5 mL L ⁻¹)	7.41	0.40	1.88	654.40	98.21	322.20	1.83	3.60	0.36	9.07
TR (10.0 mL L ⁻¹)	7.43	0.36	1.95	673.70	96.44	309.70	0.00	1.08	0.23	8.28
TR (12.5 mL L ⁻¹)	7.45	0.40	1.93	667.30	92.89	333.50	0.00	0.36	0.13	6.04

*Soil test conducted after 35 days of incubation. pH: Strongly acidic (<4.5 - 5.5), Mild acidic (5.6 - 6.5), Neutral (6.6 - 7.3), Mildly alkaline (7.4 - 8), Strongly alkaline (8.1 - > 9); EC: Normal (< 0.08), Critical for salt-sensitive crops (0.08 - 1.6), Critical for salt-tolerant crops (1.6 - 2.5), Injurious to all crops (> 2.5); Carbon: Very low (< 0.25), Low (0.25 - 0.49), Medium (0.5 - 0.75), High (0.76 - 1), Very high (> 1); Nitrogen: Very low (< 136), Low (136 - 271.9), Medium (272 - 544), High (544.1 - 816), Very high (> 816); Phosphorus: Very low (< 11), Low (11 - 22.4), Medium (22.4 - 56), High (56.1 - 84), Very high (> 84); Potassium: Very low (< 68), Low (68 - 135.9), Medium (136 - 336.9), High (337 - 506), Very high (> 506); Sulphur: Very low (< 5), Low (5 - 9.9), Medium (10 - 15), High (15.1 - 22.5), Very high (> 22.5); Zinc: Deficient (< 0.06); Iron: Deficient (< 4.5); Boron: Deficient (< 0.05).

(S, Zn, B, Fe) was also estimated which are summarised in Table 1. Organic carbon ($1.95 \pm 0.04\%$), nitrogen ($675.03 \pm 12.91 \text{ kg ha}^{-1}$), phosphorus ($92.86 \pm 9.75 \text{ kg ha}^{-1}$), zinc ($2.7 \pm 1.44 \text{ mg kg}^{-1}$) and iron ($9.37 \pm 2.88 \text{ mg kg}^{-1}$) contents were found to be very high amounts; potassium ($321.82 \pm 16.77 \text{ kg ha}^{-1}$) and boron ($0.53 \pm 0.44 \text{ mg kg}^{-1}$) were present in moderate ranges while sulphur ($3.32 \pm 3.44 \text{ mg kg}^{-1}$) content was extremely low.

Pot experimental studies

Pot preparation

Pre-autoclaved soil samples were filled equally in earthen pots (14 cm x 14 cm x 11 cm), labelled and grouped for three different pesticide treatments. Six different concentrations (1.0, 2.5, 5.0, 7.5, 10.0, and 12.5 mL L⁻¹) for each pesticide

were chosen, and three replicates for each concentration were allotted for the study. Pots without the presence of pesticide served as control setup (Nath et al., 2013).

Seedling inoculation

Seeds of *C. sativum* were collected under the supervision

Table 2. Germination rate of *Coriandrum sativum* seedlings, inoculated with pesticides at different concentrations.

Concentration (mL L ⁻¹)	Germination rate (%)		
	Fenvalerate	Cypermethrin	Chloropyrifos
1.0	100	100	100
2.5	100	100	90
5.0	85	90	75
7.5	65	70	55
10.0	50	50	35
12.5	35	40	30

and direction of the personnel of Plant Protection Department, KVK. Seeds were washed and soaked in double-distilled water for 24 h. The seeds that settled at the bottom were considered viable and were selected for the study. Seeds were wrapped in a moist cloth for another 48 h until the seeds swelled, and approximately twenty seeds were sown in each pot and watered daily (Nath et al., 2018).

Pesticide administration

Fenvalerate, cypermethrin and chlorpyrifos were sprayed on the plant leaves using a foliar spray of 1 L capacity in the early morning at an interval of ten days as suggested by the Plant Protection and Soil Science Department, KVK Cachar. Due to pesticides retention in the cultivable lands from previous farming practises, the pesticide concentration remains higher in the crop fields than the recommended dosage of 2.5 mL L⁻¹. Therefore, to mimic the crop field conditions, six different concentrations (1.0, 2.5, 5.0, 7.5, 10.0, and 12.5 mL L⁻¹) were considered for each pesticide ranging below and above the recommended dosage.

Study of growth parameters

The effect of three different pesticides and their effects at different concentrations were studied till maturation, and the flowering stage was attained. Germination rate, shoot height, biomass and moisture content with increasing pesticide concentration were also calculated. Germination rate was estimated using the following formula (Zahoranová et al., 2016):

$$\text{Germination (\%)} = \frac{\text{Germinated seeds}}{\text{Total planted seeds}} \times 100$$

Shoot heights were calculated at an interval of 5 days from the day seeds were sown till 35th day after which the plants were harvested, and the shoots were weighed to estimate the biomass of the plant whereas dry weight was estimated by exposing the plants in a hot air oven at 100°C for 2 h. The moisture content of the plants was calculated by the following formula (Medrano et al., 2015):

$$\text{Moisture content} = 1 - \frac{\text{Dry weight}}{\text{Fresh weight}} \times 100$$

Statistical analysis

Student's t-test was performed in Microsoft Excel to study the independent nature of plant growth, that is, growths of the plants

subjected to each pesticide concentration are independent, and they do not have any influence on each other. Statistical analysis was performed with shoot heights expressed as mean \pm standard deviation (SD). Software SPSS 16.0 was used to calculate the analysis of variance (ANOVA) and multiple comparison tests, considering the level of significance at $P < 0.05$. The comparison between groups of the plant heights with increasing pesticide concentration was performed assuming equal variances (Tukey's-b test) and unequal variances (Games-Howell test). Therefore, the null hypothesis, H_0 set up for the experiment was, 'with an increase in the pesticide concentration, there is no significant change in the growth parameters of *C. sativum* plants' (Namiki et al., 2018; Nath et al., 2018).

RESULTS AND DISCUSSION

Effect of pesticides on plant growth parameters

Germination rate and plant maturity

The germination rate was observed to be affected with an increase in pesticide concentration, while maximum germination was observed in control pots, showing 100% seedling viability. Germination of plants in control and pesticide concentrations of 1.0 and 2.5 mL L⁻¹ was observed within 48 h of seedling inoculation, whereas plants with 5.0 and 7.5 mL L⁻¹ concentration took three to five days to germinate. However, plants in extreme pesticide concentrations (10.0 and 12.5 mL L⁻¹) germinated after one week of inoculation, which is because plants with an increase in stressed conditions need more time to acclimatise to the condition and its metabolism also slows down; therefore, the rate of germination also varied with concentration (Table 2) (Chaudhry et al., 2002; Rennenberg, 1987). This study is in agreement with Kilic et al. (2015), who demonstrated the toxic effects of chlorantraniliprole pesticide in maize plant, rendering growth reduction in coleoptile and radicle length. Reduced coleoptile growth was also observed by Moore and Kroger (2010) in *Oryza sativa* on exposure to insecticides and herbicides like diazinon, metolachlor, atrazine, lambda-cyhalothrin and fipronil (Moore and Kroger, 2010). The damaging effect of pesticides can be reduced by proper crop management and application of

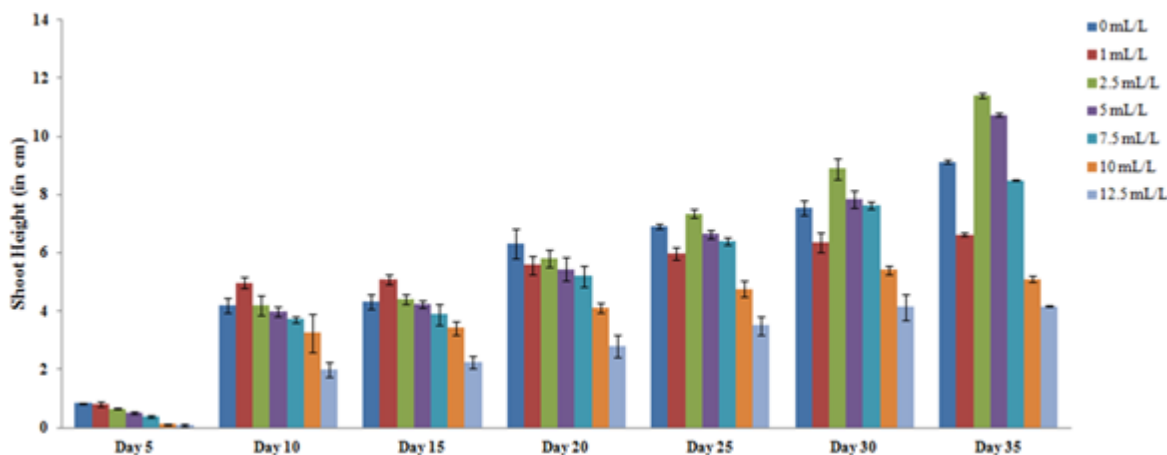


Figure 1. Effect of cypermethrin on the shoot lengths (Mean \pm S.D.) of *Coriandrum sativum*.

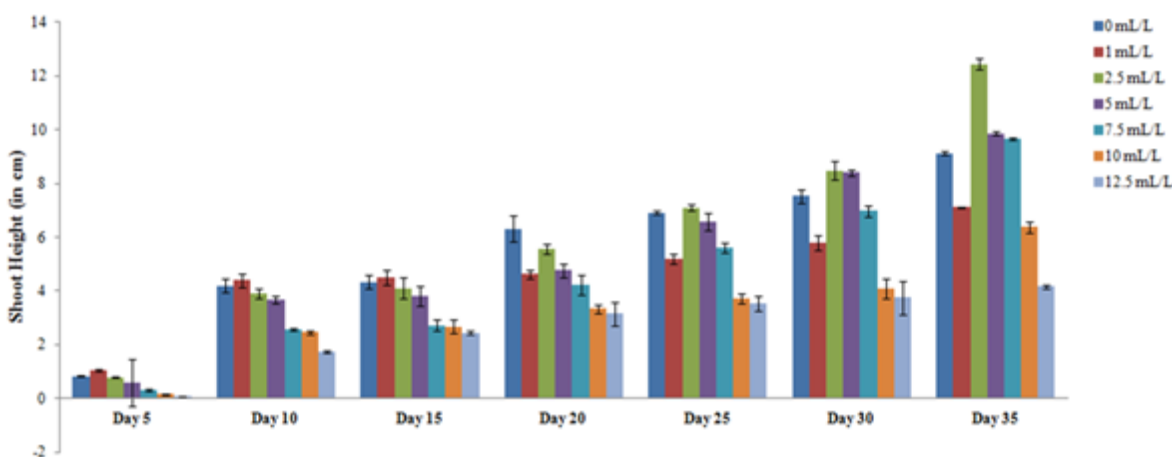


Figure 2. Effect of fenvalerate on the shoot lengths (Mean \pm S.D.) of *Coriandrum sativum*.

organic manure and bio-fertiliser in the crop field.

Estimation of shoot height

Plants which were grown under different concentrations of fenvalerate (Figure 1) and cypermethrin (Figure 2) showed a steady declining growth in the initial 15 to 20 days of the experiment whereas plantlets with pesticide dosage of 1.0 mL L⁻¹ were observed to have enhanced growth than the control plantlets. It may be because, at low concentration, the plants used the pesticides as a source of nutrition, and below the recommended dosage (2.5 mL L⁻¹), pesticides cannot prove detrimental to plant growth (Ait Barka et al., 2004). On 20th day onwards, when the plants reached the flowering stage, the plants at 2.5 mL L⁻¹ showed better growth than plants in control and 1.0 mL L⁻¹ of pesticide. This proves that the recommended dosage of pesticides has minimal effect in

the retardation of plant growth. On the other hand, the plants subjected to chlorpyrifos treatment showed a steady declining growth curve for the entire period of the experiment, and early mortality was observed in the plants (Figure 3). The average shoot lengths of plants on 15th day (before flowering) in control were 4.33 \pm 0.25 cm, and plants under 2.5 mL L⁻¹ concentration of pesticides were 4.40 \pm 0.17 cm (cypermethrin), 4.1 \pm 0.39 cm (fenvalerate) and 2.4 \pm 0.08 cm (chlorpyrifos). However, a considerable decline in the shoot length was observed at higher pesticide amendments (12.5 mL L⁻¹), which was noted as 2.25 \pm 0.21, 2.45 \pm 0.07 and 1.4 \pm 0.45 cm for cypermethrin, fenvalerate and chlorpyrifos induced plants, respectively. After attaining the flowering stage on 35th day, maximum growth was noted in the recommended dosage of 2.5 mL L⁻¹ concentration of cypermethrin and fenvalerate, exhibiting shoot length of 11.41 \pm 0.08 and 12.43 \pm 0.2 cm, respectively. Plants in control treatment also showed moderate shoot length of

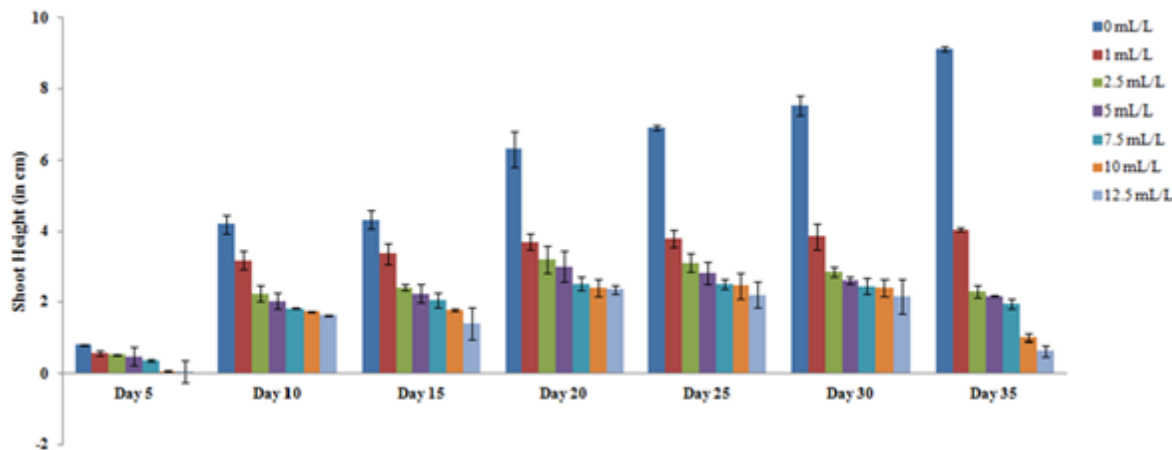


Figure 3. Effect of chlorpyrifos on the shoot lengths (Mean \pm S.D.) of *Coriandrum sativum*.

9.12 \pm 0.06 cm. However, steep retardation in shoot length under 12.5 mL L⁻¹ concentration of pesticides was observed, measuring 4.17 \pm 0.02 cm, 4.16 \pm 0.05 cm in cypermethrin and fenvalerate induced plants. Interestingly, the plants under chlorpyrifos treatment produced harmful effects on plant growth parameters and even failed to attain maturity. The detrimental effects were also visually observed, which includes reduced vigour, retarded shoot growth, drooping leaves, less branching and chlorosis. The resulting order of decreased shoot length of plants supplemented with pesticide at day 35 was: fenvalerate > cypermethrin > chlorpyrifos, which revealed that fenvalerate showed least retarding effect on the growth of plants.

Statistical inference

The t-test analysis of *C. sativum* shoots heights supplemented with cypermethrin, fenvalerate and chlorpyrifos showed a marked difference, at a 5% level of significance on both the 15th and 35th days. The tabulated value at 5% level of significance for 6 and 40 degrees of freedom was 2.336 whereas the calculated value was higher in case of cypermethrin ($t(46) = 18.573$, $p < 0.05$), fenvalerate ($t(46) = 16.123$, $p < 0.05$) and chlorpyrifos ($t(46) = 39.127$, $p < 0.05$). A similar trend was observed on the 35th day, where the tabulated values for cypermethrin, fenvalerate and chlorpyrifos were $t(46) = 8.014$, $p < 0.05$; $t(46) = 9.490$, $p < 0.05$; and $t(46) = 20.195$, $p < 0.05$, respectively. The calculated value was found to be greater than the tabulated value on both 15th and 35th days; we, therefore, reject the null hypothesis and conclude that there is a significant effect in the plant's growth with the change in pesticide concentration.

Multiple comparison studies of the shoot heights of *C. sativum* between control and other concentrations of

pesticides were calculated after 15th and 35th days of inoculation at $P < 0.05$ which revealed that on the 35th day; almost all the plants subjected to cypermethrin, fenvalerate and chlorpyrifos displayed a significant mean difference among each other at $P < 0.05$. On 35th day of inoculation, cypermethrin and fenvalerate at 2.5 mL L⁻¹ concentration marked a significant mean difference with plants in 1.0, 10.0 and 12.5 mL L⁻¹ while chlorpyrifos displayed significant mean difference with plants in control (6.57 \pm 1.15 cm), 1.0 mL L⁻¹ (1.52 \pm 0.1 cm), 10.0 mL L⁻¹ (1.46 \pm 0.11 cm) and 12.5 mL L⁻¹ (1.7 \pm 0.11 cm). The result signifies that the plants can resist the pesticide stress at 5.0 and 7.5 mL L⁻¹ as no significant mean difference was observed at these concentrations (Tables 3 to 5). The shoot heights of plants grown under cypermethrin at 5.0 and 7.5 mL L⁻¹ were recorded as 10.74 \pm 0.05 and 8.48 \pm 0.02 cm, respectively while for fenvalerate at 5.0 and 7.5 mL L⁻¹ shoot heights were 9.86 \pm 0.05 and 9.66 \pm 0.05 cm, respectively. This result clearly illustrates that plants under 5.0 and 7.5 mL L⁻¹ of pesticides showed no much difference as compared to plants in control with shoot heights of 9.12 \pm 0.06 cm.

Biomass and moisture content

Biomass reduction of shoots after 35 days of seedling inoculation displayed similar trends with shoot heights. Biomass of plants in control was calculated to be 7.403 g, and the moisture content was recorded as 84.141%. Maximum biomass was recorded at 2.5 mL L⁻¹ concentration of cypermethrin (5.324 g) and fenvalerate (5.517 g). However, with a further increase in pesticide concentration, there was a declining trend in biomass for both cypermethrin and fenvalerate. However, there is a gradual declining trend in the moisture contents of plants under the treatment of cypermethrin and fenvalerate. The moisture content recorded at the recommended dosage

Table 3. Multiple comparison tests and mean difference between the shoot lengths of *C. sativum* under cypermethrin stress after 15 days and 35 days of seedling inoculation.

Parameter (mL L ⁻¹)	After 15 days of seedling inoculation						
	Control	1.0 mL L ⁻¹	2.5 mL L ⁻¹	5.0 mL L ⁻¹	7.5 mL L ⁻¹	10.0 mL L ⁻¹	12.5 mL L ⁻¹
Control	0	0.75 ± 0.31	0.07 ± 0.26	0.09 ± 0.24	0.43 ± 0.3	0.91 ± 0.2*	2.08 ± 0.25
1.0	2.51 ± 1.43	0	0.68 ± 0.32	0.84 ± 0.31	1.18 ± 0.36	1.66 ± 0.28*	2.83 ± 0.32*
2.5	2.31 ± 1.31	4.82 ± 1.06*	0	0.16 ± 0.26	0.5 ± 0.32	0.98 ± 0.23*	2.15 ± 0.27*
5.0	1.67 ± 1.21	4.18 ± 0.94*	0.64 ± 0.73	0	0.34 ± 0.3	0.82 ± 0.2*	1.99 ± 0.25*
7.5	0.61 ± 1.52	1.9 ± 1.31	2.92 ± 1.17	2.28 ± 1.06	0	0.48 ± 0.28	1.65 ± 0.31*
10.0	3.87 ± 1.23	1.36 ± 0.96	6.18 ± 0.76*	5.54 ± 0.58*	3.26 ± 1.08	0	1.17 ± 0.22*
12.5	4.89 ± 1.26*	2.38 ± 0.1	7.2 ± 0.81*	6.56 ± 0.63*	4.28 ± 1.11	1.02 ± 0.66	0

After 35 days of seedling inoculation

Shoot heights of *C. sativum* at *P < 0.05 (ANOVA). The results are expressed as the means ± standard deviations of three independent replicates.

Table 4. Multiple comparison tests and mean difference between the shoot lengths of *C. sativum* under fenvalerate stress after 15 days and 35 days of seedling inoculation.

Parameter (mL L ⁻¹)	After 15 days of seedling inoculation						
	Control	1.0 mL L ⁻¹	2.5 mL L ⁻¹	5.0 mL L ⁻¹	7.5 mL L ⁻¹	10.0 mL L ⁻¹	12.5 mL L ⁻¹
Control	0	0.19 ± 0.23	0.23 ± 0.18	0.53 ± 0.26	1.61 ± 0.2*	1.65 ± 0.23*	1.88 ± 0.48
1.0	1.95 ± 1.21	0	0.42 ± 0.17	0.72 ± 0.25	1.8 ± 0.19*	1.84 ± 0.22*	2.07 ± 0.48*
2.5	3.53 ± 1.41	5.48 ± 0.89*	0	0.3 ± 0.2	1.38 ± 0.12*	1.42 ± 0.16*	1.65 ± 0.45
5.0	0.81 ± 1.31	2.76 ± 0.74	2.72 ± 1.03	0	1.08 ± 0.22*	1.12 ± 0.25*	1.35 ± 0.49
7.5	0.65 ± 1.17	2.60 ± 0.44*	2.88 ± 0.84	0.16 ± 0.67	0	0.04 ± 0.19	0.27 ± 0.46
10.0	2.87 ± 1.3	0.92 ± 0.71	6.40 ± 1*	3.68 ± 0.87*	3.52 ± 0.64*	0	0.23 ± 0.47
12.5	4.87 ± 1.16*	2.92 ± 0.41*	8.40 ± 0.82*	5.68 ± 0.65*	5.52 ± 0.27*	2 ± 0.61	0

After 35 days of seedling inoculation

Shoot heights of *C. sativum* at *P < 0.05 (ANOVA). The results are expressed as the means ± standard deviations of three independent replicates.

Table 5. Multiple comparison tests and mean difference between the shoot lengths of *C. sativum* under Chlorpyrifos stress after 15 days and 35 days of seedling inoculation.

Parameter (mL L ⁻¹)	After 15 days of seedling inoculation						
	Control	1.0 mL L ⁻¹	2.5 mL L ⁻¹	5.0 mL L ⁻¹	7.5 mL L ⁻¹	10.0 mL L ⁻¹	12.5 mL L ⁻¹
Control	0	0.97 ± 0.24*	1.93 ± 0.28*	2.09 ± 0.21*	2.27 ± 0.24*	2.55 ± 0.26*	2.93 ± 0.2*
1.0	5.05 ± 1.15*	0	0.96 ± 0.29	1.12 ± 0.22*	1.30 ± 0.25*	1.58 ± 0.26*	1.96 ± 0.2*
2.5	6.57 ± 1.15*	1.52 ± 0.1*	0	0.16 ± 0.26	0.34 ± 0.28	0.62 ± 0.3	1 ± 0.25*
5.0	6.89 ± 1.15*	1.84 ± 0.12*	0.32 ± 0.14	0	0.18 ± 0.22	0.46 ± 0.23	0.84 ± 0.17*
7.5	6.97 ± 1.16*	1.92 ± 0.14*	0.4 ± 0.15	0.8 ± 0.17	0	0.28 ± 0.26	0.66 ± 0.2
10.0	8.03 ± 1.15*	2.98 ± 0.09*	1.46 ± 0.11*	1.14 ± 0.13*	1.06 ± 0.14*	0	0.38 ± 0.22
12.5	8.27 ± 1.15*	3.22 ± 0.09*	1.7 ± 0.11*	1.38 ± 0.13*	1.3 ± 0.14*	0.24 ± 0.1	0

After 35 days of seedling inoculation

Shoot heights of *C. sativum* at *P < 0.05 (ANOVA). The results are expressed as the means ± standard deviations of three independent replicates.

of 2.5 mL L⁻¹ concentration of cypermethrin and fenvalerate was calculated to be 75.8 and 73.68%, respectively. Since early mortality was observed for plants under chlorpyrifos stress, its harvest was negligible at the end of the study period. Therefore, biomass and moisture content for these plants could not

be estimated (Table 6). The present study is in agreement with Carrascosa et al. (2015) who also reported biomass reduction of plants treated with fenamiphos and oxamyl nematicides; however, with the application of organic or bio-pesticides with nematophagous fungi augmented with neem paste displayed a significant

Table 6. Biomass and moisture content of *Coriandrum sativum* under cypermethrin and fenvalerate stress.

Pesticide concentration (mL L ⁻¹)	Biomass content of shoot (in g)		Moisture content of plant (in %)	
	Cypermethrin	Fenvalerate	Cypermethrin	Fenvalerate
Control (0)		7.403		84.141
1.0	0.835	0.721	77.48	81.27
2.5	5.324	5.517	75.80	73.68
5.0	3.006	4.329	68.82	70.24
7.5	1.825	1.677	60.27	61.24
10.0	1.388	0.907	48.55	37.81
12.5	0.116	0.651	43.96	28.57

increase in the biomass content. Therefore, pesticide-contaminated fields should be substituted by bio-fertilisers and bio-pesticides to reduce the harmful effects of the pesticides and restore the soil nutritive value as well as soil microbiota which play a pivotal role in plant growth, nutrient uptake and other functioning.

Conclusion

The study revealed that among the three different pesticides used, chlorpyrifos proved lethal to *C. sativum* at the recommended dosage of 2.5 mL L⁻¹, thereby causing substantial mortality of plants before the maturation stage. However, cypermethrin and fenvalerate enhanced the growth of plants at 2.5 mL L⁻¹, but with an increase in the pesticide concentration, there was a significant gradual decline in the plant growth parameters. The most striking results of the study revealed that on increasing the pesticide concentration to 5.0 and 7.5 mL L⁻¹, no marked difference in plant productivity was observed as compared to plants in control. The study establishes an optimal pesticide window range of 5.0 ± 2.5 mL L⁻¹, which did not cause any adverse effects in the growth parameters of *C. sativum*. As pesticides concentration elevates in the soil due to its longer retention capacity, subsequent cropping on the same cultivable land causes retardation in plant growth even if the soil is highly fertile and conducive for farming. Abiding by the recommended pesticide dosage is, therefore, the key for enhancing and maintaining good productivity and vigour of crops.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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