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Black pepper vine establishment as affected by growing media and disinfectants

Tajudeen B. Akinrinola*¹; Olamijuwon E. Oguntade¹; Kafayat O Oyelami¹; Folasade O. Adeyemi² **Address:**

- ¹ Department of Crop and Horticultural Sciences, Faculty of Agriculture, University of Ibadan, Ibadan, Nigeria
- ² Department of Soil Resources and Environmental Management, Faculty of Agricultural Sciences, Ekiti State University, Nigeria

*Corresponding author: Tajudeen B. Akinrinola. email: tb.akinrinola@gmail.com

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Black pepper is economically the most widely used spice crop around the world. Its cultivation is limited by the nursery's low success rate of vine cuttings. Appropriate growing media and uninfected cuttings could improve its establishment. Hence, this study investigates black pepper vine-cutting establishments using different growing media and disinfectants. A 2×5 factorial experiment involving two growing media [100% Soil and Sawdust+Soil (1:1)] and five disinfectants (Aloe vera, 10% HgCl₂, 20% Hypochlorite, 70% Ethanol, 100% Ethanol) were evaluated in a randomized complete design with five replicates. Data on days to sprouting, percentage of mortality, and days to dieback were subjected to analysis of variance and significantly different means were separated (p<0.05). Black pepper vines established in 100% Soil (26.91) had significantly earlier days of sprouting than Soil+Sawdust (38.31). The HgCl₂-treated vines sprouted significantly earlier than Hypochlorite, while others were similar. Combining 100% Soil and HgCl2 promoted the earliest days of sprouting. The 100% Soil and HgCl2 treatments significantly lowered mortality compared to Soil+Sawdust and other disinfectants. Vine mortality ranged from 0.00% (Soil × HgCl₂) to 60.00% Sawdust+Soil × Hypochlorite and Sawdust+Soil × 70% Ethanol. Vines in Sawdust+Soil (17.00) had shorter days to dieback than 100% Soil (21.67). Aloe vera treatment had no dieback. Days to dieback range from 0.00 (Soil × Aloe vera, Sawdust+Soil × 100% Ethanol and Soil × Hypochlorite) to 55.67 (Soil × 70% Ethanol) for the interaction effect. The 100% Soil with 10% mercuric chloride was most suitable for establishing black pepper vines.

Keywords: Black pepper, growing media, disinfectants, sprouting, vine mortality

INTRODUCTION

Black pepper (*Piper nigrum* L.) is considered the "King of spices" or "Black gold" due to its durability and value (Hammouti *et al.*, 2019). It is called "Iyere" in Yoruba and "Uziza" in Igbo. The other common names of black pepper are Benin pepper, Guinea pepper and false cubeb (Attah *et al.*, 2012). The fruit is a single-seeded berry comprising 1.02% volatile oil and 5-9% alkaloids, such as piperine, chavicine, piperidine, piperetine and resin (Zheng *et al.*, 2016). It is a widely used spice that adds a flavour of its own to dishes and enhances the taste of other ingredients (Spence, 2024). The plant is mainly grown for its fruits. The fruits are used as a spice in culinary preparation, food flavouring, table source, seasoning, and perfumery worldwide. Black pepper is one of the peppers with diversified products (Wang *et al.*, 2021). It is mainly grown in tropical countries like Brazil, China, India, Indonesia, Malaysia and Sri Lanka (Takooree *et al.*, 2019). It is semi-cultivated in Southern Nigeria (Olife and Onwualu, 2013). Currently, the production of black pepper in the global market has not been able to match the growing demand for the crop and its products (Korah and Mohankumar, 2021).

Black pepper grows well in a warm and humid climate and requires a well-draining and nutrient-rich growth medium for optimal growth and development (Ilahi and Ahmad, 2017). Black pepper can be propagated vegetatively or through seeds, but seedlings raised from seeds will not breed true to type due to its heterozygous nature (Ghosh and Haque, 2019). The preferred method for propagation is vine cutting, even for commercial cultivation. However, this method can be challenging due to high mortality rates and establishment failures (Ee and Shang, 2017). Factors such as aeration, mineral nutrients, and moisture content of the growing medium affect the establishment and development of the seedlings (Kaushal and Kumari, 2020). Many proposed media are too expensive for most farmers. Black pepper establishments with cheap and locally available media reportedly increased survival and early growth (Akinrinola and Onadeko, 2023; Akinrinola and Anyanumeh, 2025). However, vine-cutting infection could also be a serious factor in the survival of cuttings.



Contaminations of cuttings proffer a major challenge to cutting establishment in the nursery. Different disinfectants have been used to establish cuttings in the tissue culture (Okoroafor, 2022). These disinfectants are essential for maintaining a clean and sterile environment to prevent infection of plants during the cutting of vines that can negatively affect plant propagation success (Hassan and Hendi, 2021). However, their effects on stem cuttings can vary based on the specific sterilisation method (Lykogianni *et al.*, 2023). Different sterilisation techniques can affect the efficiency and success of propagation through stem cuttings. In addition, the choice of sterilisation method can influence the success of growth and survival rates of plantlets (Baysal-Gure *et al.*, 2016). Therefore, selecting the appropriate sterilisation method is essential to ensure the desired outcomes when working with stem cuttings.

Disinfectants formulated for horticultural or agricultural purposes vary (Baysal-Gure *et al.*, 2016). Ethanol and sodium Hypochlorite reportedly eliminate exogenous bacteria on plant surfaces (Hodkinson *et al.*, 2019). Other disinfectants used for stem cuttings include sodium chloride, mercuric chloride, Hypochlorite and Ethanol (Nwite *et al.*, 2022). *Aloe vera* can also be a cheap, natural and effective disinfectant, providing a safe and non-toxic alternative to chemical pesticides and fungicides (Shahbaz *et al.*, 2022). These disinfectants should be used in proper dilution to ensure the health and vigour of propagated plants, thus ensuring successful growth and development (Hodkinson *et al.*, 2019). Most disinfectants reported are for tissue culture multiplication (Okoroafor, 2022). However, there is a dearth of information on locally available media and the use of disinfectants on black pepper vines cutting establishment in the open nursery. Therefore, the performance of black pepper vine cuttings affected by different growing media and disinfectants was determined.

MATERIALS AND METHODS

The experimental location:

The experiment was carried out between July and September 2023 under the shade of a citrus tree at the back of the Department of Agronomy, Faculty of Agriculture, University of Ibadan, Oyo State, Nigeria. The coordinates are 7.34°N and 3.89°E, with an altitude of 227 m above sea level. The fourth night's wet and dry thermometer readings during the observation period are presented in Figure 1. The wet and dry temperatures in a greenhouse (measured with a dry and wet bulb thermometer) provide insight into the differences in vapour pressure deficit between the two growing periods, which affect the variation in plant transpiration and growth rates. The first planting started during the short break in rainfall, while the second planting commenced during the second season. The time difference may lead to variation in relative humidity and temperature, which affects cutting survival and rate of growth.

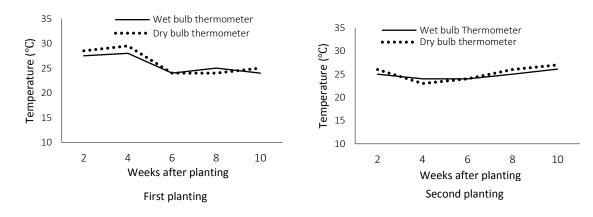


Fig. 1. The wet and dry bulb thermometer (°C) readings during the first and plantings at the experimental site.

Experiment design and treatments:

The experiment was a 2×5 factorial experiments involving two growing media, M1: (50% Sawdust+50% Soil) and M2: (100% Soil), as well as five disinfectants in a randomised complete block design with five replicates. The growth media were mixed properly to get a perfect combination, constituting the ten treatments as follows:

T1: M1 × Aloe vera, T2: M1 × HgCl₂, T3: M1 × Hypochlorite, T4: M1 × 70% Ethanol, T5: M1 × 100% Ethanol, T6: M2 × Aloe vera, T7: M2 × HgCl₂, T8: M2 × Hypochlorite, T9: M2 × 70% Ethanol, T10: M2 × 100% Ethanol.

Preparing the disinfectants:

To prepare the correct mixture, 10 g of *Aloe vera* was combined with 100 mL of water to form an *A. vera* solution. For HgCl₂, 10% Mercuric chloride was mixed with distilled water to make 100 mL, and for Hypochlorite, 20% Hypochlorite with 2 drops of tween 20 was combined with distilled water to make it 100 mL. The 70% Ethanol and 100% Ethanol were diluted with distilled water to make 100 mL using a 100 mL measuring cylinder for both the 70% and 100% Ethanol solutions, respectively.

Pre-planting operations:

The growth media (Sawdust and topsoil) were mixed and bagged in standard 2 kg perforated nursery planting polythene bags of 27 cm × 25 cm with a thickness of 1 mm. Five replications were employed. The planting bags were filled with the growth media of different percentages, such that 25 bags contained 50% Sawdust and 50% Soil (M1), and the remaining 25 bags contained 100% Soil (M2). This was achieved using a bowl as a means of measurement for accuracy.

Collection of planting materials:

The black pepper vines were collected in the evening from the established black pepper plot in the Department of Agronomy, Faculty of Agriculture, University of Ibadan. The vines were planted the same day under the shade of a citrus tree at the back of the Department of Agronomy.

Collection of growing media:

The growing media (Sawdust and topsoil) were collected from different locations. Sawdust was collected at the Bodija sawmill in Ibadan, while the topsoil was collected from an organic farm at the Teaching and Research farm, University of Ibadan, Ibadan, Nigeria. The pH of the Sawdust+Soil and Soil media used in the study were 6.56 and 6.43, respectively.

Planting:

The black pepper vine cuttings with three nodes each were planted into polythene bags containing the media, one vine per bag. At planting, a groove wider than the size of the cutting was made at the center so as not to rub off the solution when inserting the vine. Before planting the vines, part of the vine was immersed in a disinfectant. The poly bags received daily irrigation until sprouting and afterwards, at intervals of two days.

Data collection:

Data collected were media pH, days to sprouting, number of leaves, leaf area, and days to dieback of plants. The leaf area was measured from the length and the breadth multiplied by a constant value of 0.61 (Partelli *et al.*, 2007). Data was collected and recorded at two-week intervals. The number of leaves was counted manually, and a metre rule was used to measure the leaf length and breadth. The days of sprouting were considered the days of the appearance of young leaves at the nodes of the vines. Cutting mortality was estimated by counting the number of unsprouted cuttings as a percentage. The days to dieback of the plant were taken on the day the sprouted cuttings showed visible browning symptoms.

Statistical analysis:

The collected data were subjected to two-way analysis of variance (Gomez and Gomez, 1984) and significantly different means between the growing media and disinfectants and were separated using Least Significant Difference (LSD) at (p<0.05). Statistical Analysis System (SAS) software version 9.0 was used for the statistical analysis.

RESULTS

Influence of growing media and disinfectants on days to sprouting in black pepper:

Different growing media and disinfectants significantly affected the rates at which black pepper vines sprouted during the first and second plantings (Table 1). The use of 100% Soil significantly encouraged earlier sprouting of black pepper vines than the Soil+Sawdust medium during both plantings. The $HgCl_2$ -treated cuttings had significantly reduced days to sprouting than the Hypochlorite-treated cuttings, while the other disinfectants were similar during the first planting. However, the 100% Ethanol treatment reduced the days to sprouting by 2 and 6 days compared to 70% Ethanol and *A. vera*, respectively (with no significant differences). During the second planting, the Hypochlorite-treated cuttings had significantly delayed sprouting compared to the other treatments. The $HgCl_2$ treatment had significantly earlier sprouting than other treatments, except for 100% Ethanol-treated cuttings. During both plantings, the interaction between the growing media and disinfectants revealed that Sawdust+Soil × Hypochlorite had a significantly higher number of days to sprouting than Sawdust+Soil × $HgCl_2$, Soil × $HgCl_2$

Table 1. Effects of growing media and disinfectants on days to sprouting and mortality of the black pepper vines.

	VIIIes.	Days to	sprouting	Mortality (%)			
Treatments		First planting	Second planting	First planting	Second planting		
	Media						
1	Sawdust+Soil	38.31	37.33	46.15	48.00		
2	Soil	26.91	24.47	24.44	30.67		
	LSD (0.05)	8.35	2.10	15.09	10.05		
	Disinfectants						
1	Aloe vera	36.67	32.17	40.00	43.33		
2	HgCl ₂	24.63	23.00	15.00	26.67		
3	Hypochlorite	42.40	40.33	48.00	53.33		
4	70% Ethanol	30.60	33.00	48.00	40.00		
5	100% Ethanol	28.71	26.00	28.57	33.33		
	LSD _(0.05)	14.60	7.21	7.46	15.90		
	Interactions						
1	Sawdust+Soil × Aloe vera	44.33	35.33	40.00	46.67		
2	Sawdust+Soil × HgCl₂	26.00	24.67	40.00	40.00		
3	Sawdust+Soil × Hypochlorite	53.00	53.00	60.00	60.00		
4	Sawdust+Soil × 70% Ethanol	38.50	42.67	60.00	53.33		
5	Sawdust+Soil × 100% Ethanol	34.67	31.00	40.00	40.00		
6	Soil × Aloe vera	29.00	29.00	40.00	40.00		
7	Soil × HgCl ₂	23.80	21.33	0.00	13.33		
8	Soil × Hypochlorite	35.33	27.67	40.00	46.67		
9	Soil × 70% Ethanol	25.33	23.33	40.00	26.67		
10	Soil × 100% Ethanol	24.25	21.00	20.00	26.67		
	LSD (0.05)	10.20	10.20	16.93	22.48		
	P-Values						
	Media	0.05*	<.001**	<.001**	<.001**		
	Disinfectants	<.001**	<.001**	<.001**	<.001**		
	Interactions	<.001**	<.001**	<.001**	<.001**		

WAP = Weeks After Planting; LSD = Least Significant Difference; P values = < 0.05, ns = Not Significant; *, and ** = Significance at 0.05 and 0.001 levels of probability, respectively

Influence of growing media and disinfectants on percentage of mortality:

A significant increase in vine mortality was observed for the Sawdust+Soil medium compared to soil during both plantings (Table 1). The Hypochlorite and 70% Ethanol treatments had significantly higher vine mortality than the other treatments during the first planting. However, the mortality of cuttings for Hypochlorite, 70% Ethanol and *A. vera* treatments were similar during the second planting. The HgCl₂ treatments during both plantings had the lowest mortality values. The interactions between the growing media and disinfectant showed significant variations among the treatments in both plantings. The Sawdust+Soil × Hypochlorite and Sawdust+Soil × 70% Ethanol (60.00) were similar for vine mortality but had significantly higher values than the other treatment interactions during the first planting. The Soil × 100% Ethanol treatment had a significantly lower vine mortality than the other treatment interactions. The trend observed during the second planting was similar to the first, except that the lowest values in Soil × HgCl₂, Soil × 100% Ethanol, and Soil × 70% Ethanol were comparable.

Influence of growing media and disinfectants on the number of leaves of black pepper:

The influences of growing media on the number of leaves of black pepper were consistent in both plantings (Table 2). The cuttings raised in both media had a decrease in the number of leaves at 10 WAP. The cuttings raised in the Soil medium had more leaves than those raised in the Sawdust+Soil medium throughout the observation periods, except at four and six WAP during the first and second plantings, respectively. The influence of disinfectants on the number of leaves was insignificant at two and four WAP during the first planting. However, 70% Ethanol-treated cuttings had a significantly higher number of leaves than HgCl₂-treated cuttings at six WAP. The number of leaves ranged from 0.8 (70% Ethanol) to 1.50 (*A. vera*) at 8 WAP, but were similar. At 10 WAP, no significant difference in the number of leaves was observed among disinfectants, but A. vera and 70% Ethanol had the highest values. During the second planting, a significantly higher number of

leaves was observed in the 70% Ethanol-treated cuttings compared to the 70% Ethanol-treated cuttings at 6 WAP. The responses of black pepper cutting to disinfectants were similar for the number of leaves at eight WAP but differed significantly at 10 WAP. The interactions between media and disinfectants were comparable throughout the observation periods during the first planting. However, Sawdust+Soil × Hypochlorite had the lowest number of leaves at 2, 4, 6 and 8 WAP, while Sawdust+Soil × HgCl₂ had the lowest value at 10 WAP. During the second planting, the influences of growing media by disinfectant interactions were similar for the number of leaves throughout observation periods, except at 6 WAP, in which Sawdust+Soil × 70% Ethanol-treated cuttings had significantly higher values than Sawdust+Soil × HgCl₂ and Soil × HgCl₂ treated HgCl₂-treated cuttings.

Influence of growing media and disinfectants on black pepper leaf area (cm²):

The influence of growing media on the leaf area observed was similar during the first and second plantings (Table 3). However, the cuttings raised in Soil had more leaves than those raised in the Sawdust+Soil medium throughout the observation periods, except at 4 and 6 WAP during the first and second plantings, respectively. The highest leaf area was observed in the vines treated with HgCl₂, while the lowest was observed under Hypochlorite treatment at 4 WAP during the first planting. At 8 and 10 WAP, there was a decline in the leaf area of the 70% Ethanol-treated cuttings, with significantly lower values than the other treatments. The *A. vera* treated cuttings had 0.44, 41.82 and 6.06% higher leaf area than HgCl₂, Hypochlorite and 100% Ethanol treatments. During the second planting, the *A. vera* treated plants

Table 2. Effects of growing media and disinfectants on the number of leaves of black pepper vines during the first and second plantings

Treatments		First planting (WAP)				Second planting (WAP)			
		4	6	8	10	4	6	8	10
	MEDIA								
1	Sawdust+Soil	0.38	0.69	1.08	0.85	0.67	1.00	1.20	0.87
2	Soil	0.67	0.83	1.00	0.89	0.73	0.87	1.00	0.93
	LSD (0.05)	ns	ns	ns	ns	ns	ns	ns	ns
	DISINFECTANTS								
1	Aloe vera	0.33	0.67	1.00	1.00	0.67	1.00	1.17	1.83
2	HgCl₂	0.63	0.75	0.75	0.75	0.67	0.83	0.67	1.67
3	Hypochlorite	0.40	0.60	1.00	0.80	0.33	0.67	1.17	0.83
4	70% Ethanol	0.60	1.00	1.40	1.00	1.00	1.33	1.33	0.83
5	100% Ethanol	0.71	0.86	1.14	0.86	0.83	0.83	1.17	1.67
	LSD _(0.05)	ns	ns	0.56	ns	ns	0.66	ns	0.93
	INTERACTIONS								
1	Sawdust+Soil × Aloe vera	0.33	0.67	1.00	1.00	0.67	1.33	1.33	2.00
2	Sawdust+Soil × HgCl ₂	0.33	0.67	0.67	0.67	0.67	0.67	0.67	1.33
3	Sawdust+Soil × Hypochlorite	0.00	0.50	1.00	0.50	0.00	1.00	0.33	0.33
4	Sawdust+Soil × 70% Ethanol	0.50	1.00	1.50	1.00	1.00	1.67	1.00	1.00
5	Sawdust+Soil × 100%	0.67	0.67	1.33	1.00	1.00	1.33	1.00	1.00
	Ethanol								
6	Soil × Aloe vera	0.33	0.67	1.00	1.00	0.67	1.00	1.00	1.67
7	Soil × HgCl₂	0.80	0.80	0.80	0.80	0.67	0.67	0.67	1.00
8	Soil × Hypochlorite	0.67	0.67	1.00	1.00	0.67	1.33	1.33	1.33
9	Soil × 70% Ethanol	0.67	1.00	1.33	1.00	1.00	1.00	0.67	0.67
10	Soil × 100% Ethanol	0.75	1.00	1.00	0.75	0.67	1.00	1.00	1.33
	LSD (0.05)	ns	ns	0.75	ns	ns	0.93	ns	1.32
			•	P-Values	<u> </u>				
	Media	>.05	>.05	>.05	>.05	>.05	>.05	>.05	>.05
	Disinfectants	>.05	>.05	<.05*	>.05	>.05	<.05*	>.05	<.05*
	Interactions	>.05	>.05	<.0001***	>.05	>.05	<.0001***	>.05	<.0001***

WAP = Weeks After Planting; LSD = Least Significant Difference; P values = < 0.05, ns = Not Significant; *, and *** = Significance at 0.05 and 0.0001 levels of probability, respectively

Table 3. Effects of growing media and disinfectants on the leaf areas (cm²) of black pepper vines during the first and second plantings

	Treatments		First plant	ting (WAP)		Second planting (WAP)			
		4	6	8	10	4	6	8	10
	MEDIA								
1	Sawdust+Soil	3.66	14.66	13.59	13.83	6.87	14.77	12.18	12.56
2	Soil	1.27	16.78	15.05	15.58	11.44	14.06	15.04	15.68
	LSD (0.05)	0.67	ns	ns	ns	ns	ns	ns	ns
	DISINFECTANTS								
1	Aloe vera	9.53	16.34	17.98	18.50	12.37	15.99	17.46	17.95
2	HgCl ₂	13.28	20.93	17.90	18.04	10.59	11.71	12.80	13.03
3	Hypochlorite	4.06	10.12	10.46	11.49	3.33	11.11	8.58	8.99
4	70% Ethanol	5.31	10.79	5.20	5.46	8.30	13.23	8.15	8.96
5	100% Ethanol	9.44	17.51	16.89	17.16	11.19	20.04	21.05	21.68
	LSD (0.05)	4.53	ns	7.40	7.50	9.02	ns	11.44	11.81
	INTERACTIONS								
1	Sawdust+Soil × <i>Aloe vera</i>	3.23	11.35	13.02	13.76	4.63	6.12	8.21	9.02
2	Sawdust+Soil × HgCl₂	4.78	6.68	17.64	17.59	12.78	14.44	14.64	14.79
3	Sawdust+Soil × Hypochlorite	0.99	8.52	6.73	6.74	2.02	13.39	4.49	4.49
4	Sawdust+Soil × 70% Ethanol	4.14	14.06	5.65	5.76	7.22	18.81	12.58	12.66
5	Sawdust+Soil × 100% Ethanol	4.46	20.46	19.99	20.24	7.70	21.08	20.97	21.82
6	Soil × <i>Aloe vera</i>	15.84	21.32	22.94	23.24	20.12	25.86	26.70	26.87
7	Soil × HgCl₂	18.38	23.49	18.05	18.30	8.39	8.97	10.97	11.26
8	Soil × Hypochlorite	6.10c	11.19	12.95	14.66	4.64	8.83	12.68	13.48
9	Soil × 70% Ethanol	6.09c	8.61	4.90	5.26	9.39	7.65	3.72	5.26
10	Soil × 100% Ethanol	13.17	15.31	14.56	14.84	14.68	19.00	21.12	21.54
	LSD (0.05)	9.22	12.37	ns	ns	12.77	16.36	16.17	16.71
	P-Values								
	Media	<.001**	>.05	>.05	>.05	>.05	>.05	>.05	>.05
	Disinfectants	<.001**	>.05	<.001**	<.001**	<.001**	>.05	<.001**	<.001**
	Interactions	<.0001***	<.0001	>.05	>.05	<.0001***	<.0001***	<.0001***	<.0001***

WAP = Weeks after Planting; LSD = Least Significant Difference; P values = < 0.05, ns = Not Significant; *, and ** = Significance at 0.05, and 0.001 levels of probability, respectively

had significantly higher leaf area than Hypochlorite-treated cuttings at 4 WAP. The leaf area ranged from 11.11 (Hypochlorite) to 20.04 (100% Ethanol) and was similar for the treatments at six WAP. The 100% Ethanol-treated cuttings had significantly higher leaf areas than Hypochlorite and 70% Ethanol-treated cuttings at 8 and 10 WAP. The interactions of growing media and disinfectants had no significant influence on leaf area. The Sawdust+Soil × Hypochlorite treatment had the lowest value at 2, 4 and 6 WAP, while Soil × 70% Ethanol had the lowest values at 8 and 10 WAP. The highest leaf area values at 2, 4 and 6 WAS were observed under Soil × A. vera, Soil × HgCl₂ and Soil × HgCl₂, respectively during the first planting. The Soil × A. vera had the highest leaf area values at 8 and 10 WAP. Aloe vera × Soil and Sawdust+Soil × Hypochlorite had consistent leaf area increase throughout the observation periods. The cuttings raised from the other treatment interactions experienced a decrease in leaf area values after 6 WAP. During the second planting, the leaf area of black pepper varied significantly among treatments with cuttings raised in Soil × A. vera having the highest values throughout the observation periods. The Sawdust+Soil × Hypochlorite-treated cuttings had the lowest values at 4, 8 and 10 WAP, while the Sawdust+Soil × A. vera treatment had the lowest value at 6 WAP.

Effects of growing media and disinfectants on days to Dieback:

Raising black pepper cutting in either Soil or Sawdust+Soil media had no significant effect on the days to dieback during the first and second plantings (Table 4). However, the Sawdust+Soil medium had 4.67 earlier days to dieback than Soil as the growing medium. The 70% Ethanol as a disinfectant in black pepper cutting had a significantly higher number of days to vine-cutting dieback, while no dieback was recorded for *A. vera*. There was no significant difference between the 70% Ethanol and the other treatments. The interaction effects of growing media and disinfectants indicated that the Sawdust+Soil \times *Aloe vera*, Sawdust+Soil \times 100% Ethanol, Soil \times *Aloe vera* and Soil \times Hypochlorite treatments had no dieback. However, Soil \times 70% Ethanol had the highest days to dieback.

The media pH after the study indicated no significant variation between the two media during the first planting. The pH for the different disinfectants ranges from 6.38 (HgCl₂) to 6.74 ($A.\ vera$). The Soil × HgCl₂ interaction had the lowest pH, while the Sawdust+Soil × $Aloe\ vera$ treatment had the highest pH value.

During the second planting, the pH was significantly higher in Sawdust+Soil than in Soil. *Aloe vera* use resulted in a considerably lower media pH than the other treatments. The Sawdust+Soil × *Aloe vera* interaction had a significantly higher pH than other treatment combinations.

Table 4. Effects of growing media and disinfectants on days to dieback during the first and second plantings

			OTDD '	pH				
	Treatments	First planting	Second planting	First planting	Second planting			
	MEDIA							
1	Sawdust+Soil	17.00	20.00	6.56	6.55			
2	Soil	21.67	13.80	6.43	6.42			
	LSD at 0.05	ns	7.32	0.12	0.10			
	DISINFECTANTS							
1	Aloe vera	0.00	0.00	6.74	6.69			
2	HgCl ₂	26.13	21.67	6.38	6.43			
3	Hypochlorite	15.4	26.00	6.47	6.44			
4	70% Ethanol	49.20	36.83	6.41	6.43			
5	100% Ethanol	11.29	0.00	6.46	6.42			
	LSD at 0.05	18.24	34.39	0.09	0.15			
	INTERACTIONS							
1	Sawdust+Soil × Aloe vera	0.00	0.00	6.82	6.83			
2	Sawdust+Soil × HgCl₂	21.67	21.67	6.41	6.47			
3	Sawdust+Soil × Hypochlorite	38.50	52.00	6.49	6.47			
4	Sawdust+Soil × 70% Ethanol	39.50	26.33	6.48	6.48			
5	Sawdust+Soil × 100% Ethanol	0.00	0.00	6.57	6.49			
6	Soil × Aloe vera	0.00	0.00	6.67	6.55			
7	Soil × HgCl₂	28.80	21.67	6.36	6.39			
8	Soil × Hypochlorite	0.00	0.00	6.46	6.41			
9	Soil × 70% Ethanol	55.67	47.33	6.37	6.37			
10	Soil × 100% Ethanol	19.75	0.00b	6.37	6.36			
	LSD at 0.05	48.64	48.64	0.22	0.22			
	P-Values							
	Media	>.05*	<.0001***	<.001**	<.001**			
	Disinfectants	<.001**	<.001**	<.001**	<.001**			
	Interactions	<.0001***	<.0001***	<.0001***	<.0001***			

DTDD= Days to Dieback; WAP = Weeks After Planting; LSD = Least Significant Difference; P values = < 0.05, ns = Not Significant; *, and *** = Significance at 0.05 and 0.0001 levels of probability, respectively

DISCUSSION

Sprouting in vegetative propagation is initiated by metabolic activities and the multiplication of cells in the bud located at the leaf axil of the cutting under appropriate environmental conditions (Shu et al., 2024). Early sprouting of the vine cuttings was affected by the growing media used in the black pepper vine establishment. The plants established with 100% Soil sprouted earlier than 50% Soil + 50% Sawdust. This agrees with a study conducted by (Agboola et al., 2018), who investigated the effect of Sawdust as a growth medium on the establishment of tomatoes. They reported that the unavailability of soil microorganisms that can make the Sawdust medium nutrient-rich through decomposition resulted in poor tomato establishment. The result showed that HgCl2 was significantly different from Hypochlorite but similar to the other disinfectants for days to sprouting. This indicated that HgCl₂, besides being more efficient than Hypochlorite in cleaning the cuttings of harmful fungi and bacteria, also promotes sprouting. Although reports have indicated Hypochlorite as a good disinfectant in tomatoes (Lykogianni et al., 2023), This variation could be due to the composition of Na in the Hypochlorite that causes mild moisture stress that delayed sprouting in vine cuttings. The interactions of Soil with disinfectants reduced sprouting substantially compared to Soil+Sawdust with the earliest sprouting observed in the Soil × HgCl2 interaction. The early sprouting observed with the HgCl2 suggests that this treatment offers more favourable conditions for bud growth than the other media by disinfectant interactions. The lower sprouting from the Sawdust with disinfectant interactions could be due to the reduced chemical effectiveness of Sawdust. Organic matter has been proven to lower disinfectant efficacy (Allingham et al., 2024).

The mortality of cuttings was significantly lower in the cuttings raised in Soil compared to the Sawdust+Soil treatment. The trend observed for the sprouted cuttings was inversely related: the higher the number of sprouted cuttings, the lower the observed mortality. Cutting mortality after planting is generally due to the inability of the cuttings to cope with the environmental factors to establish roots and commence growth (Nyiramvuyekure et al., 2023). The higher survival of cuttings raised with Soil could not have had

better conditions than those raised in Soil+Sawdust, other than the variation in the medium for growth. Thus, the Soil was better than the Sawdust+Soil by promoting a higher percentage of cutting survival. The result corroborated (Akinrinola and Onadeko's, 2023) report that black pepper cuttings established with Soil had lower cutting mortality than Sawdust and Soil mixtures. Applying HgCl2 in disinfecting black pepper cutting before planting significantly increased cutting survival compared to the other treatments. The variations among the disinfectants could be attributed to the differences in their efficacy in suppressing harmful microbes or not being mild on the cuttings. The A. vera treatment also had high mortality; however, the highest vine mortality was observed for Hypochlorite and 70% Ethanol treatments. The result indicated that the HgCl2 had a better influence on the cuttings. The effects could be achieved through the reduction of harmful microbes and enhanced rooting that supports the growth of the cuttings compared to the other treatments. The influence of media x disinfectant interactions indicated significant differences among treatments. Soil x HgCl2 treatment had the lowest vine mortality, while the highest value was observed in Sawdust+Soil × Hypochlorite and Sawdust+Soil × 70% Ethanol. These interactions mainly involving Sawdust indicated non-supportive options for establishing black pepper cuttings. This could be related to the presence of organic matter in the Sawdust that reduced the efficacy of disinfectant in suppressing harmful microbes. This result conforms to (Allingham et al., 2024) findings that the potency of disinfectants is reduced by organic matter. The comparatively lower cutting mortality substantiates this claim observed for 100% Soil to Sawdust+Soil treatments.

There were noticeable variations in the number of leaves throughout the observation periods between the growing media. This study affirmed the report that Soil supported prolonged growth in the nursery than Sawdust+Soil medium (Akinrinola and Onadeko's, 2023). Under disinfectant treatments, the number of leaves was similar but had significant variations between the 70% Ethanol and HgCl2 treatments at 6 WAP and a subsequent reduction in the number of leaves for most treatments. This could imply depletion in the soil's nutrient status, consequently, the nutrient acquisition for further growth decreased. The trends observed for the media x disinfectant interactions were similar to the plant's responses to disinfectants, except that the values for the Sawdust+Soil were higher than 100% Soil. Similar trends for the influences of media, disinfectants, and their interactions were observed for the leaf area. The number of leaves and leaf area indicates the plant's ability to capture sunlight and commence photosynthesis. The variations in the results observed may be accounted for by the improved environment that triggered the production of photosynthetically active leaves, which accelerates cell division and expansion of leaves (Koch et al., 2019). They found that increased leaf area directly correlated with increased leaf number because of the leaves' contribution to improved net photosynthesis rate, which determines growth. This implied that the transplanting of the seedlings under these conditions should be targeted towards 6 and 7 WAP. However, the higher number of leaves observed for the 70% Ethanol did not translate to higher leaf area.

Seedling dieback results from unfavourable conditions that limit the ability of the plant's rooting system to acquire nutrients and moisture from the soil for active growth. The factors responsible for dieback could be biotic or edaphic (Lemaire *et al.*, 2022). The prolonged sprouting could also have produced weak seedlings. The cuttings established through Sawdust+Soil experienced earlier days of dieback than the Soil medium. The possibility of infection through Sawdust contained in the medium could be responsible. The days to dieback for the established cuttings were highest for 70% Ethanol and lowest for *A. vera*. The disinfectants that promoted early sprouting resulted in the early days of the seedlings' dieback. With the exclusion of other factors, the availability of water and nutrients in the media ensures that the plants can accumulate photosynthates required for growth (Kochetova *et al.*, 2022). However, the depletion of these resources leads to established seedling dieback. The *A. vera* treatments, with Sawdust+Soil or Soil, had no dieback. This result confirms (Shahbaz *et al.*, 2022; Akinrinola and Anyanumeh, 2025) reports that *A. vera* soothes and heals any wounds or damage caused to the plant and reduces pest infestation, thus promoting plant health and growth. There were consistencies in the observed results for the interactions, except that 100% Ethanol had no dieback recorded during the second planting. The reduction in dieback value for the treatment could explain the lower mean value observed in the Soil treatment.

The media pH values were similar. Consequently, no variation in the response of the seedlings to pH is expected. The pH of the media ranged from 6.36 (Soil × HgCl₂) to 6.82 (Sawdust+ Soil × A. vera), indicating that the media was slightly acidic to neutral. The pH range of 5.5 to 6.5 is considered optimum for black pepper (Zu *et al.*, 2014). Media acidification resulting in an Al3+ increase can impair root growth, cause nutritional imbalance and disrupt water uptake of acid-sensitive species (Rahman *et al.*, 2023). Microbial activities are affected by media pH, thus the bioavailability of macro- and micro-nutrients. Soil microbes reportedly thrive better under slightly acidic conditions with a pH range of 6-7 due to the high bioavailability of most nutrients (Rahman *et al.*, 2021). Consequently, there would be a need to minimise the effects of

harmful bacteria and fungi to enhance crop establishment. The range of the media pH suggests no considerable variation in the seedling response. However, the variation observed must have been caused by disinfectants' influence in suppressing cuttings' infection and improving the media condition for root development.

CONCLUSION

The results revealed that the black pepper vine cuttings grown in 100% Soil sprouted earlier, had reduced mortality and increased days before dieback compared to the vines established with 50% Sawdust + 50% Soil. Dipping black cuttings in $HgCl_2$ encouraged early sprouting and low mortality. The 100% Soil combination with mercuric chloride ($HgCl_2$) encouraged earlier sprouting compared to other treatments. The vine mortality was in order Sawdust+Soil × Hypochlorite, Sawdust+Soil × 70% Ethanol > Sawdust+Soil × Aloe vera, Sawdust+Soil × $HgCl_2$, Sawdust+Soil × $HgCl_2$, Sawdust+Soil × $HgCl_2$, Black pepper growth was enhanced when 100% Soil was used, as seen by improvements in leaf area, number of leaves, and days to sprout.

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