

Asian Research Journal of Agriculture

Volume 16, Issue 1, Page 6-16, 2023; Article no.ARJA.97101 ISSN: 2456-561X

# Influence of 2,4-dichloro Phenoxy Acetic Acid on Seeds Germination and Early Growth of Mesquite (Prosopis juliflora Swarz) DC Seedlings

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#### Authors' contributions

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

#### Article Information

DOI: 10.9734/ARJA/2023/v16i1378

#### **Open Peer Review History:**

This journal follows the Advanced Open Peer Review policy. Identity of the Reviewers, Editor(s) and additional Reviewers, peer review comments, different versions of the manuscript, comments of the editors, etc are available here: https://www.sdiarticle5.com/review-history/97101

**Original Research Article** 

Received: 18/10/2022 Accepted: 29/12/2022 Published: 27/02/2023

# ABSTRACT

This study was conducted to evaluate the effect of 2,4-dichloro phenoxy acidic acid (2,4-D) on seed germination and the early growth of mesquite (*Prosopis juliflora*) seedlings. The three sizes of mesquite at the flowering stage (small, medium, and large) were treated with 2,4-D at different rates  $(0, 6\times10^3, 12\times10^3, 18\times10^3 \text{ and } 24\times10^3 \text{ mg a.i.} / \text{L})$  dissolved in two solvents (diesel or water), the application method basal bark treatment was used in the two different seasons. The results showed that: the three factors (2,4-D rates, tree sizes, and solvent types) significantly decreased germination percentage with the increase of 2,4-D rate dissolved in diesel with all sizes of trees, while the emergence percentage, radical length, and shoot height were significantly decreased as 2,4-D rate increase dissolved in diesel or water. In the winter season, the root length of seedlings

Asian Res. J. Agric., vol. 16, no. 1, pp. 6-16, 2023

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raised from seeds collected from the three tree sizes treated with 2,4-D dissolved in water was significantly decreased as a 2,4-D rate increased, while in the rainy season, the root length was affected with 2,4-D dissolved in diesel. In addition, the effect of 2,4-D rates and solvent types was evaluated for all parameters, the results found that: 2,4-D dissolved in diesel significantly decreased germination percent than the corresponding rates dissolved in water. Emergence percentage and shoot height and radical length were significantly decreased with 2,4-D dissolved in diesel or water compared to their control. In the winter season, the root length is affected by 2,4-D dissolved in water, while in the rainy season affected by 2,4-D dissolved in diesel.

Keywords: Mesquite; seedlings; emergence; 2,4-D; shoot height and roots length germination; radical length.

# **1. INTRODUCTION**

The fruits of mesquite (P. juliflora) known as pods are green, becoming commonly whitevellow when fully mature after 60 days of flowering [1]. The pods of mesquite do not open at maturity, both domestic and wild animals play a main role in breaking the dormancy of mesquite seeds by grazing upon the pods, the un-chewed seeds passing through the guts of animals that, enhances seeds germination [2]. Mesquite trees commonly propagated by producing a large number of small-sized seeds: about 25 per pod, germinated, seeds expended most of their energy in developing a root system and locating a water source as soon as possible [3]. The primary product of photosynthesis in higher plants is starch, which is stored as a carbohydrate that supports metabolism and growth during the dark when photosynthesis is not possible [4]. The mobilization of starch by the  $\alpha$ -amylase enzyme initiated seed germination [5]. In addition, nitrogen compounds convert stored starch to soluble sugar, which is needed for germination and adjusting the potassium and sodium ratio; and increasing the production of ATP and seed respiration [6].

2,4-dichloro phenoxy acetic acid (2,4-D) mimics the effects of the natural auxin (Indole- 3-acetic acid) (IAA) in plants [7]. There are two types of formulations of 2,4-D: amine salts and esters [8]. Natural auxins IAA usually inactivated very quickly through conjugation and degradation in the plant, while synthetic auxins such as 2,4-D persist for long periods of time within the plant, this phenomenon is described as an auxin overdose which leads to an imbalance in auxin and interactions with other hormones at the tissue level [9].

Chloroplasts have been considered the initial target of 2,4-D- since the chemical is mainly accumulated and subsequently metabolized in

these organelles [10]. Plants usually metabolize herbicides via processes that convert the parent molecule to more polar products and insoluble residues The selectivity of auxinic [11]. herbicides may be due to the vascular tissue structure of dicot [12], which possesses a cambium, and the synthetic auxin is not metabolized as quickly as the endogenous auxin [13]. Application of 2,4-D herbicide inhibited the seed germination percentage by penetrating the seed coat and is effective as a pre-emergence treatment. Auxins like 2.4 -D are metabolically broken down by the isozyme oxidase with the release of phenols, which accumulated in the tissues and inhibit the germination of seeds [14].

This work aims to evaluate the effect of 2,4-D on the germination percentage and the radical length, emergence percentage, and shoot height, root length of seeds collected from 2,4-D-treated mesquite trees at the flowering stage.

# 2. MATERIALS AND METHODS

# 2.1 Field Experiments

#### 2.1.1 Sites of the field experiments

Field experiments were conducted during the winter season of 2017/ 2018 and the rainy season of 2018 in the demonstration farm of the Faculty of Agriculture University of Khartoum-Sudan. The site lies at lat.15, 40 N, long 32, 32E, in a semi-arid zone, characterized by a great variation in temperature, which ranges from 18° C to 40° C, and relative humidity of 34% to 75% [15].

#### 2.1.2 Planting materials

Naturally stand mesquite trees were carefully selected in the two locations, selected trees were districted to three spots, and for each spot, thirty mesquite trees were carefully selected and classified for three sizes (small, medium, and large), ten in each based on the number of stems per tree and diameter of the canopy (in meters). Small trees, 1 to 2 stems and less than 2 meters for canopy, medium trees, 3 to 5 stems and 2-3 meters for canopy and large trees, more than 5 stems and more than 3 meters for canopy.

#### 2.1.3 Pre-treatment

At the flowering stage, ten inflorescences in any tree (small, medium, and large) were randomly selected, and marked before treatment application.

# 2.1.4 Chemical solutions and application method

2,4 -D was applied at four rates  $6 \times 10^3$ ,  $12 \times 10^3$ ,  $18 \times 10^3$  and  $24 \times 10^3$  mg a.i. / L in tank mixture with water or diesel. In addition, water and diesel are used individually as control. The application was conducted in two seasons (Winter and rainy season), method of basal bark treatment was used in which 2,4-D mixtures were sprayed around the lower part of the tree stem at about 30 cm above the soil level [16].

#### 2.1.5 Collection of Pods and seed preparation

Ripen pods produced from marked inflorescences were collected in paper bags from any group of treated trees (small, medium, and large) from any block, seeds were removed from pods using a Sharpe knife and scissors and used immediately after extraction. Dormancy is broken by soaking the seeds in sulfuric acid (abt. 98%) for a few minutes and washing them thoroughly with distilled water until the acid is removed; the seeds are soaked for 24 hours in distilled water.

# 2.2 Experimental Design

Field experiments were arranged in factorial in Randomized Complete Block Design (RCBD) with three replicates. The experimental unit consisted of three mesquite trees of different sizes (small, medium, and large) for any treatment in the block. Treated with five rates of 2,4-D, dissolved in two solvents (Table 1).

#### 2.3 Data Collection

#### 2.3.1 Germination percentage

Ten seeds from any treatment were placed in a Petri – dish (9 cm. id) lined with two Whatman

filter papers moistened with 2.5 ml of distilled water. Petri - dishes wrapped in black polyethylene under laboratory conditions. The laboratory experiment was arranged in factorial in a Completely Randomized Design (CRD) with three replicates, after five days. The percentage of germination was counted and the radical length was measured after one week.

### 2.3.2 Radical length

One week later the mean of radical length (cm) was measured (five germinated seeds were taken)

#### 2.3.3 Emergence percentage

Ten seeds from any treatment were transferred to the greenhouse and planted in polyethylene bags, (ten seeds in each), filled with 5 kg of soil mixed (1:1 v/v) loam and sand (40 cm. in diameter) with drainage holes in the bottom. The experiments were conducted during the period from May to June 2017 and from January to February 2018 for seeds collected from trees treated in the winter and rainy seasons, respectively. The polyethylene were bags kept in the greenhouse under natural conditions of temperature and day length and irrigated every two days. The experiment was arranged in factorial in Randomized Complete Block Design (RCBD) with three replicates. The seedling emergence percent was watched over a period of 15 days and the emergence percent was calculated.

#### 2.3.4 Shoot height and root length

The shoot height and root length were measured one month after emergence from the ten seedlings in the bag, seedlings were taken out of the plastic bags for any treatments and cleaning, and seedling height: and root length were measured in cm.

# 2.4 Data Analyses

Data subjected to analysis of variance test (ANOVA) and means statistically separated by least significant difference test (LSD) for comparisons using a computer statistical software, (Statistix 8), and differences between means at (0.05) level of significance.

Treatment	2,4-D rate.in (10 <sup>3</sup> ) mg
S/D/(2,4-D)/R0	2,4-D at rate zero dissolved in diesel
S/D/(2,4-D)/R1	2,4-D at rate 6 dissolved in diesel
S/D/(2,4-D)/R2	2,4-D at rate 12 dissolved in diesel
S/D/(2,4-D)/R3	2,4-D at rate 18 dissolved in diesel
S/D/(2,4-D)/R4	2,4-D at rate 24 dissolved in diesel
S/W/(2,4-D)/R0	2,4-D at rate zero dissolved in water
S/W(2,4-D)/R1	2,4-D at rate 6 dissolved in water
S/W(2,4-D)/R2	2,4-D at rate 12 dissolved in water
S/W/(2,4-D)/R3	2,4-D at rate 18 dissolved in water
S/W/(2,4-D)/R4	2,4-D at rate 24 dissolved in water
M/D/(2,4-D)/R0	2,4-D at rate zero dissolved in diesel
M/D/(2,4-D)/R1	2,4-D at rate 6 dissolved in diesel
M/D/(2,4-D)/R2	2,4-D at rate 12 dissolved in diesel
M/D/(2,4-D)/R3	2,4-D at rate 18 dissolved in diesel
M/D/(2,4-D)//R4	2,4-D at rate 24 dissolved in diesel
M/W/(2,4-D)/R0	2,4-D at rate zero dissolved in water
M/W/(2,4-D)/R1	2,4-D at rate 6 dissolved in water
M/W/(2,4-D)/R2	2,4-D at rate 12 dissolved in water
M/W/(2,4-D)/R3	2,4-D at rate 18 dissolved in water
M/W/(2,4-D)/R4	2,4-D at rate 24 dissolved in water
L/D/(2,4-D)/R0	2,4-D at rate zero dissolved in diesel
L/D/(2,4-D)/R1	2,4-D at rate 6 dissolved in diesel
L/D/(2,4-D)/R2	2,4-D at rate 12 dissolved in diesel
L/D/(2,4-D)/R3	2,4-D at rate 18 dissolved in diesel
L/D/(2,4-D)/R4	2,4-D at rate 24 dissolved in diesel
L/W/(2,4-D)/R0	2,4-D at rate zero dissolved in water
L/W/(2,4-D)/R1	2,4-D at rate 6 dissolved in water
L/W/(2,4-D)/R2	2,4-D at rate 12 dissolved in water
L/W/(2,4-D)//R3	2,4-D at rate 18 dissolved in water
L/W/(2,4-D)/R4	2,4-D at rate 24 dissolved in water

Table 1. The different treatments and 2,4-D rates

S=Small, M=Medium and L=Large R=Rate, D=Diesel, W=Water

#### 3. RESULTS

# 3.1 Effect of 2,4-D, Solvent Types, and Tree Sizes on Seeds Germination Percentage

Table 2 shows the effect of 2,4-D on the germination percentage of seeds collected from the three mesquite tree sizes treated different rates of 2,4-D dissolved with in diesel, showing a significant decrease in germination percent with an increase of 2,4-D rate. The results showed that the two high rates of 2,4-D  $(18 \times 10^3 \text{ and } 24 \times 10^3 \text{ mg a.i.})$  dissolved in diesel, decreased the germination percentage of seeds collected from three tree sizes compared to the control. The overall mean of the effect of 2,4-D dissolved in diesel on germination percentage was decreased by 26.52%. 26.4%, 32.57%, and 47.37%, 25.51%, 47.44% than, the water solvent for small, medium, and large trees in the winter and rainy seasons, respectively.

### 3.2 Effect of 2,4-D, Solvent Types, and Tree Sizes on Seedlings Emergence Percentage

The statistical analysis of variance indicated significant differences (P= 0.05) in response to the effect of the two high rates of 2,4-D dissolved in diesel or water in seed emergence percent of mesquite seeds in the two seasons. The decrease of emergence percentage in response to 2,4-D was increased in the rainy season than in the winter season for seeds collected from the three tree sizes. The overall mean of the emergence percentage decreased with 2,4-D dissolved in diesel than dissolved in water.

### 3.3 Effect of 2,4-D, Solvent Types, and Tree Sizes on Radical Length, Shoot Height, and Root Length

A greater mean value of shoot height and root length were observed in seedlings raised from seeds collected from mesquite trees treated in the winter season compared to that raised from

Treatment	Germination Percentage								
		Winter seaso	on		Rainy season				
	S	М	L	S	Μ	L			
D2,4-DR0	93.3 ab	93.3 ab	93.3 ab	70.0 defg	86.7 abcd	76.7 cdef			
D2,4-DR1	83.3 bcd	90.0 abc	83.3 bcd	83.3 abcde	76.7 cdef	76.7 cdef			
D2,4-DR2	70.0 def	60.0 fg	63.3 efg	80.0 bcdef	63.3 fg	56.7gh			
D2,4-DR3	40.0 h	63.3 efg	63.3 efg	40.0 h	70.0 efg	63.3 fg			
D2,4-DR4	53.3 gh	60.0 fg	53.3 gh	43.3 h	63.3 fg	56.7 gh			
Mean	67.98	73.32	71.3	63.32	72.00	66.02			
W2,4-DR0	86.7 abc	96.7 ab	100.0 a	100.0 a	100.0 a	100.0 a			
W2,4-DR1	93.3 ab	96.7 ab	96. ab	96.7 ab	96.7 ab	100.0 a			
W2,4-DR2	76.7 cde	90.0 abc	93.3 ab	100.0 a	93.3 abc	90.0 bc			
W2,4-DR3	83.3 bcd	96.7 ab	90.0 abc	66.7 efg	93.3 abc	96.7 ab			
W2,4-DR4	90.0 abc	83.3 bcd	93.3 ab	100.0 a	100.0 a	100.0 a			
Mean	86.00	92.68	94.52	92.68	96.66	97.34			
SE±	8.07			9.24					

# Table 2. Effect of 2,4-D on germination percentage of seeds collected from mesquite trees in different sizes

# Table 3. Effect of 2,4-D on the emergence percentage of seeds collected from mesquite trees in different sizes

Treatment	Emergence Percentage								
		Winter season		Rainy season					
	S	Μ	L	S	M	L			
D2,4-DR0	53.33 fghijk	70 abcdefg	70.0abcdefg	63.33abc	56.67 cde	46.67 cdef			
D2,4-DR1	56.67 efghij	56.67 efghij	66.7bcdefgh	46.67cdef	16.67 ghi	20.00 ghi			
D2,4-DR2	43.33 im	63.33cdefghi	50.0ghijk	30.00 ghi	13.33 hi	20.00 ghi			
D2,4-DR3	26.67nop	23.33mnop	46.67ki	10.00 i	10.00 i	16.67 ghi			
D2,4-DR4	13.33op	10.00p	23.33 mnop	10.00 i	10.00 i	10.00 i			
Mean	38.67	44.67	50.14	32.00	21.33	22.67			
W2,4-DR0	83.33 ab	86.67 ab	90.00 a	80.00 ab	73.33 ab	86.67a			
W2,4-DR1	73.33 abcdef	60.00 defghi	76.67 abcde	73.33 ab	63.33 abc	76.67ab			
W2,4-DR2	80.00cd	60.00 defghi	63.33 cdefghi	46.67 c def	60.00bcd	40.00 cdefg			
W2,4-DR3	63.33cdefghi	46.67 hijkl	70.00 bcdefg	26.67 fghi	46.67 cdef	36.67defgh			
W2,4-DR4	33.33klmno	36.67 jklmn	66.7 bcdefgh	33.33 efghi	56.67 cde	30.00 fghi			
Mean	66.66	58.00	73.34	52.00	60.00	54.00			
SE±	10.60			13.67					

rainy season seeds (Table 4). It found that all treatments in the winter season gave higher values of shoot height and root length compared to their respective treatments in the rainy seasons. In the rainy season, there were nonsignificant differences in shoot height between all 2,4-D rates dissolved in diesel, while all 2,4-D rates dissolved in water significantly decreased shoot height compared to the control. In the winter season, the three high rates of 2,4-D dissolved in diesel or water: significantly decreased the shoot height of seedlings. Seedlings raised from seeds collected from treated trees with 2.4-D dissolved in diesel in the winter season showed non-significant differences in root length, while the three high rates of 2,4-D dissolved in water significantly decreased root length compared to the control. In the rainy season, all 2,4-D rates dissolved in diesel significantly decreased root length, while only the

high three rates dissolved in water significantly decreased root length compared to the control. In addition, the two high rates of 2,4-D dissolved in diesel or water significantly decreased the radical length in both seasons (Table 4).

# 3.4 Effect of 2,4-D, and Solvents Types on Germination %, Emergence %, Radical Length Shoot Height, and Root Length

All rates of 2,4-D dissolved in diesel solvent had significantly less germination percent and radical length compared to the corresponding rates dissolved in water. The two high rates of 2,4-D  $(18 \times 10^3 \text{ and } 24 \times 10^3 \text{ mg a.i.})$  gave less germination percent, which gave (55.6%, 55.6%) and (57.8%, 54.4%) and less radical length (4.96 cm, 4.04 cm) and (4.61 cm, 3.71 cm) in winter

Treatment			Radic	al length					Shoot he	ight						Root length		
		Winter			Rainy			Winter			Rainy			Winter			Rainy	
	S	М	L	S	М	L	S	М	L	S	Μ	L	S	М	L	S	M	L
D2,4-DR0	9.03	6.87	9.33 a	7.77	6.00	5.67	32.73	32.83 abc	30.83	11.90	8.60	8.93	19.60	22.17	26.27	16.0a	14.77ab	12.23
	ab	abcd		abcde	efgh	efghi	abc		abcde	В	bcdef	bcdefg	cdef	abcdef	abc			cdefghi
D2,4-DR1	8.03	7.00	9.03	7.87	4.80	5.33	28.97	28.67	31.30	10.67	9.60	9.66	22.97	21.77	26.43	10.67ghijklmn	10.27ijklmn	9.93 klmn
	abcd	abcd	ab	abcd	ijkl	efghi	abcdefg	abcdefg	abcd	Bcd	bcdef	bcdef	abcdef	bcdef	abc			
D2,4-DR2	6.50	7.83	5.70	6.47	5.17	5.20	24.20 efg	24.43 efg	23.83 fg	10.10	9.60	6.47 fg	20.00	18.07	20.03	11.10	9.00n	9.00 n
	cd	abcd	efgh	efgh	ijk	fghi				bcdef	bcdef		bcdef	def	bcdef	Fghijklm		
D2,4-DR3	4.47	5.50	4.90	3.50 n	4.67	3.67	25.33 defg	23.83 fg	23.57 g	8.50	6.83	4.00 g	19.27	20.57	19.63	10.50	9.67lmn	9.63 mn
	efg	efgh	efgh		lm	m				bcdef	efg		cdef	bcdef	cdef	Hijklmn		
D2,4-DR4	4.60	3.27 i	4.27	3.83 n	3.80 n	3.50	24.30 efg	23.77 fg	23.33 g	8.50	6.67	4.00 g	17.73 ef	17.97	18.33	6.63 o	10.1jklmn	9.67 lmn
	efgh		gh			m				Bcdef	efg			def	cdef			
Mean	6.54	6.09	6.66	7.09	6.09	4.67	27.11	26.71	26.57	9.93	8.26	6.61	19.91	20.11	22.14	10.98	10.76	10.09
W2,4-DR0	9.13 a	9.33 a	8.00	8.03	8.57 a	8.33	30.67	33.27 ab	34.87 a	30.10 a	29.67 a	32.20 a	27.93 ab	30.10 a	26.00	14.0abc	12.43 cdefgh	12.83
			abcd	abc		abc	abcde								abcd			bcdef
W2,4-DR1	6.57	8.77	8.57	8.43 ab	8.10	8.00	30.23	29.50	27.27	11.20	11.33	10.77	24.77	21.70	20.86	13.87bc	12.07 cdefghij	12.30
	bcd	abc	abc		abc	abc	abcdef	abcdefg	bcdefg	bc	bc	bcd	abcdef	bcdef	bcdef			cdefghi
W2,4-DR2	8.23	7.40	5.80	7.57	7.47	7.20	24.90 defg	25.63 defg	24.70	11.00	11.00	10.43	17.30 ef	19.10	16.73 f	13.4bcde	12.67 cdefg	12.40
	abcd	abcd	efg	abcd	abcd	bcd			defg	bc	bc	bcde		cdef				cdefgh
W2,4-DR3	5.10	7.10	6.00	6.60	7.33	6.50 ij	29.90	25.17 defg	26.57	11.83 b	8.60	9.43	23.43	19.73	16.56f	12.77bcdef	11.33 fghijklm	12.23
	efgh	bcd	efg	defg	bcd		abcdefg		bcdefg		bcdef	bcdef	abcdef	cdef				cdefghi
W2,4-DR4	5.40	7.07	5.80	6.33 fg	7.10	6.23	26.40	24.07 efg	25.80	9.43	10.00	8.00	19.73	20.33	16.67 f	11.87defghijk	11.73 efghijkl	8.67 no
	efgh	bcd	efg		cd	ijk	cdefg		defg	bcdef	bcdef	cdef	cdef	bcdef				
Mean	6.89	7.93	6.83	7.39	7.71	7.25	28.42	27.53	27.82	14.71	14.12	14.17	22.63	22.19	20.07	11.20	12.07	11.69
SE±	1.26			0.66			3.40			1.88			4.02			1.05		

Table 4. Effect of 2,4-D on radical length, shoot height and root length of seedlings raised from mesquite trees in different sizes

Treatment	Germination %(Petri dishes)		Emergence % (Greenhouse)		Radio	al length	Root	t length	Shoot height		
	W	R	W	R	W	R	W	R	W	R	
D2,4-DR0	93.3ab	77.8c	57.78bcd	50.0b	8.41ab	6.48cd	22.68bcd	13.57a	29.65ab	9.81bc	
D2,4-DR1	85.6b	78.9c	60.0bcd	33.33cd	8.02abc	6.00de	23.72ab	11.06cd	24.15cde	9.98bc	
D2,4-DR2	64.4c	66.7d	58.89bcd	21.11de	6.68 cd	5.61e	19.37bcde	9.93de	24.15cde	8.72c	
D2,4-DR3	55.6c	57.8de	23.33e	13.33e	4.96ef	4.61f	19.82bcde	9.7e	24.24de	6.44d	
D2,4-DR4	55.6c	54.4e	23.33e	10.0e	4.04f	3.71g	18.01de	8.8e	23.8e	6.39d	
Mean	70.9	67.12	44.67	25.55	6.42	5.28	20.73	10.61	25.20	8.27	
W2,4-DR0	94.4ab	100.0a	86.67a	74.44a	8.82 a	8.31a	28.01a	12.38ab	32.93a	30.66a	
W2,4-DR1	95.6a	97.8a	70.0b	48.89b	7.97bc	8.18a	22.44bcd	12.38ab	29.02bcd	11.1b	
W2,4-DR2	86.7ab	94.4ab	52.22d	35.0b	7.14bcd	7.41b	17.71e	12.82a	25.08e	10.81bc	
W2,4-DR3	88.9ab	85.6bc	65.56bc	36.67bc	6.07de	6.81bc	19.37bcde	12.45ab	27.21bcd	9.95bc	
W2,4-DR4	88.9ab	100.0a	55.56cd	40.0bc	6.09de	6.56cd	27.74bc	11.42bc	25.42de	9.14bc	
Mean	90.0	95.56	66.00	47.00	7.22	7.44	23.05	12.29	27.93	14.33	
SE±	4.69	5.29	6.12	7.66	0.73	0.38	2.35	0.60	1.96	1.09	

Table 5. Effect of 2,4-D on germination %, emergence %, radical length, Shoot height, and root length

and rainy seasons, respectively. In the rainy season, all 2,4-D rates dissolved in diesel significantly decreased emergence percentage, while in the winter season, only the two high rates of 2,4-D dissolved in diesel solvent showed significant differences in emergence percentage compared to the control. All 2,4-D rates dissolved in diesel had significantly less shoot height compared to the corresponding rates in water. In the winter season, all 2,4-D rates dissolved in water significantly decreased root length, while in the rainy season, only the two high rates of 2,4-D ( $18 \times 10^3$  and  $24 \times 10^3$  ppm a.i.) dissolved in diesel showed a significant difference in root length compared to the control (Table 5).

### 4. DISCUSSION

In this study, 2,4-D (auxin-like herbicide) dissolved in diesel was applied at the flowering stage of a mesquite tree, and the germination percent of seeds collected from the treated tree significantly decreased as the rate of 2,4-D increased. The present study is in line with many researchers, [17], who found that; Dicamba (auxinic herbicide) applied at the reproductive stage of soya beans, reduced seeds germination percentage. Application of 2.4-D at the beginning of seed formation of soybean caused a reduction of 8% in seed germination compared to the application at the beginning of flowering [18]. 2,4-D was also, found to be efficient in inhibiting seed germination and radical length of Hibiscus cannabinus at a rate of 5000 ppm compared to the control which gave 98% and 15.7 cm for germination and radical length, respectively [19]. Liu [20] found that The application of 2.4-D in wheat at the soft dough stage of seeds, resulted in a higher level of residue of the active ingredient of 2,4-D in seeds, compared to the application of herbicide at ripening stages.. This may possibly result from the increased translocation of photoassimilates from the various plant structures to the seeds [21]. This period considers a shorter time for the metabolism of the 2,4-D herbicide which penetrates the seed coat and is effective as a pre-emergence herbicide. Synthetic auxins, like 2,4-D, are metabolically broken down by the isozyme oxidase with the release of phenols, the accumulation of these phenols in the tissues might inhibit the germination of seeds [14].

Henner et al. [22], reported that small molecules of petroleum hydrocarbons are phytotoxic for germination. The toxic hydrocarbon molecules in diesel solvents inhibit the activities of amylase and starch phosphorylase [23].

The emergence and seedling establishment constitute the most critical periods in the life cycle of the plants. The emergence percent of seeds collected from treated trees in both rainy and winter seasons showed a significant decrease. 2.4-D at low concentration acts as auxin and stimulates the growth of plant cells and interacts with endogenous hormones like ethylene, gibberellic acid, abscisic acid, and auxins [10]. 2,4-D at a high concentration which acts as herbicide and persists for a long time within the plant and is described as an auxin overdose [9]. The application of 2.4-D in wheat at the soft dough stage of seeds, resulted in a higher level of residue of the active ingredient of 2.4-D in seeds, compared to the application of herbicide at ripening stages [20]. The largest level of residue of herbicide 2.4-D found at the soft dough stage, compared to the ripening stages, may possibly result from increased translocation of photoassimilates from the various plant structures to the seeds [21]. In monocot stems, the vascular tissues (the phloem and xylem) are scattered in bundles and lack a vascular cambium and synthetic auxin can quickly inactivate by conjugation, while in dicot stems, the vascular tissues are formed in rings and possess a cambium and the synthetic auxin is not metabolized as quickly as the endogenous auxin [13]. Application 2,4-D herbicide at the flowering stage of mesquite trees (dicot stem), increased the herbicide residue in seeds, this is due to that, mesquite pods ripen 60 days after flowering [1]. This period considered is a shorter time for the metabolism of the 2,4-D herbicide and penetrating the seed coat and is effective as a pre-emergence herbicide and decreased emergence percent. In addition, the toxic hydrocarbon molecules in diesel solvents inhibit the activities of amylase and starch phosphorylase, [23]. The primary product of photosynthesis in higher plants is starch, which is stored as a carbohydrate that supports metabolism and growth during the dark when photosynthesis is not possible [4]. The mobilization of starch by the  $\alpha$ -amylase enzyme initiated seed germination [5]. The importance of amylase activities in germination, which convert stored starch to soluble sugar, which adjusts the K<sup>+</sup>/Na<sup>+</sup> ratio and increases ATP production and seed respiration [6].

The decrease in root length and shoot height observed in the present study is in line with that observed by [24], who found that the root length and shoot height of Coriandrum sativum treated with 2.4-D decreased as 2.4-D concentration increased. Dicamba (auxinic herbicide) applied at the reproductive stage of soybean reduced plant height [17]. Khan et al. [25] Reported that the shoot and root length of Triticum aestivum and Phalaris minor raised from seeds treated with different concentrations of 2,4 -D, decreased with the increase of 2,4-D concentration. Also, the application of 2.4-D near sovbean sowing caused a decrease in shoots and root length [26]. Abelmoschus esculentus treated with 2,4-D at five rates (1-5 ppm) showed a reduction in shoot height and root length decreased after five days of treatment [27]. 2,4-D lasts for a long time resulting in the overproduction of ethylene [28]. Ethylene production in response to 2.4-D is the stimulation of  $H_2O_2$  which is considered the second messenger in abscisic acid synthesis (ABA) [29]. Overproduction of H<sub>2</sub>O<sub>2</sub>, which mediates stomata closure to reduce water loss and promotes oxidative damage, which reduces the production of plant biomass [30]. In addition, alkanes and polycyclic aromatic hydrocarbons (PAHs) in the diesel solvent increased the toxicity, which reduced the growth of the stem and the root [31]. The performance of 2,4-D in the rainy season decreased the shoot height more than in the winter season, this may have been due to high humidity in the rainy season which increased the uptake and translocation of 2,4-D [8].

# 5. CONCLUSION

Mesquite tree commonly propagated bv producing a large number of viable seeds. The result of this study showed that 2,4-D decreased the viability of seeds which reflected in germination decreased and emergence percentage and also decreased the shoot height and root length, which affected seedlings' establishment. The results found that the sizes of the trees have no effect on all parameters. The results of this study contribute to reducing spread of this tree. Therefore, the we recommend more studies to find out whether this effect was continuing or for a single production season.

# **COMPETING INTERESTS**

Authors have declared that no competing interests exist.

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Peer-review history: The peer review history for this paper can be accessed here: https://www.sdiarticle5.com/review-history/97101