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Aloe vera Leaf Gel Extract as a Natural Tool for Sustainable Weed Management: Effects on Germination and Early Growth

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ABSTRACT

This research was carried out to assess the allelopathic activity (Aloe vera) aqueous leaf gel extracts on seed germination and some seedling growth parameters of wheat (Triticum aestivum L.), wild barley (Hordeum spontaneum) and black mustard (Brassica nigra). The aqueous extract was added by five concentration levels (0%, 5%, 10%, 15% and 20%) to 20 crops and weed seeds species in sterilized petri dish for 10 days at 22°C. The experiment as arranged out completely randomized design. High performance liquid chromatography was used to quantify and determine bioactive compound in the leaf gel. The findings showed that increasing concentrations from 5% to 20 % of aqueous extracts of Aloe vera leaf gel significantly reduced seed germination, germination speed, (radical, plumule, and seedling length) and (radicle, plumule and seedling dry weight) of all plant species. Whereas, according to the results increasing concentration levels from 5% to 20 % significantly increased inhibition of seed germination percentage, radicle, plumule and seedling growth inhibition. The outcomes revealed that the effects were concentration dependent. Some phenolic compounds such as Palmitic acid, Folic acid, linoleic, Stearic acid, Lauric acid and Myristic acid were profiled in Aloe vera leaf gel's aqueous extract by using high performance liquid chromatography (HPLC). The study's outcomes suggest that Aloe vera leaf gel's aqueous extract could perform as bioherbicide for managing some weeds and supporting sustainable agriculture practices thereby reducing reliance on chemical herbicide.

مستخلص هلام الالوفيرا كأداة طبيعية لمكافحة الأدغال بشكل مستدام: التأثيرات على الإنبات والنمو المبكر

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الخلاصة

أجريت هذه الدراسة لتقييم الإمكانات الأليلوباثية لمستخلص المائي لهلام أوراق الألوفيرا على إنبات البذور وبعض الصفات بادرات الحنطة ***Triticum aestivum* L.)** والشعير البري **(*Hordeum spontaneum*)** والخردل البري **(*Brassica nigra*).** تمت إضافة المستخلص المائي بخمسة تراكيز (٠٪، ٥٪، ١٠٪، ١٥٪ و ٢٠٪) إلى ٢٠ بذرة من كل نوع نباتي في طبق بتريديش

معقم لمدة ١٠ أيام عند ٢٢ درجة مئوية. تم استخدام التصميم العشوائي الكامل.كما و تم استخدام كروماتوغرافيا السائل عالية الأداء لقياس وتحديد المركب النشط بيولوجيًا في هلام الأوراق. أظهرت النتائج أن زيادة التركيز من ٥% إلى ٢٠% من المستخلصات المائية ثبطت معنويا إنبات البذور وسرعة الإنبات وطول الجذير و الرويشة و البادرات والوزن الجاف للجذير والرويشة والبادرات لجميع أنواع النباتات. أدت زيادة مستويات التركيز من ٥% إلى ٢٠% إلى زيادة معنوية في تثبيط نسبة إنبات البذور وتثبيط نمو الجذير والريشة والبادرات. تم تحديد بعض المركبات الفينولية مثل حامض البالمتيك وحامض الفوليك واللينوليك وحامض الستياريك وحامض اللوريك وحامض الميريستيك في المستخلص المائي لهلام الأوراق الألوفيرا باستخدام كروماتوغرافيا السائل عالية الأداء **.(HPLC)** تشير نتائج الدراسة إلى امكانية استخدام المستخلص المائي كمبيد حيوي لإدارة بعض الأدغال الضارة ودعم ممارسات الزراعة المستدامة والذكية وبالتالي تقليل الاعتماد على مبيدات الأدغال الكيميائية.

الكلمات المفتاحية: الألوفيرا، الأليلوباثي، مبيدات الحيوية للأدغال، الإدارة المستدامة للأدغال الضارة، المركبات النشطة بيولوجيا.

1. INTRODUCTION

Allelopathy is a natural phenomenon of one-plant releases inhibitory chemical substances that inhibit the growth of neighboring plants (Molisch, 1937). Allelopathic plants produce allelochemicals that release into the environment and transfer to the target plant (Olofsdotter et al., 2002).The essential role of phenolic and alkaloids is through their effect on the natural activities of the plant such as germination, growth, photosynthesis, transpiration, biochemical metabolism, nucleic acid and protein synthesis (Duke, 2015). Previous records demonstrated that various secondary plant metabolites are intended to exhibit bioherbicide actions connected to the formation of allelochemicals (Bogatek, 2006). There are allelopathic experiment studied on weeds and crops that used different plant parts component such extracts, exudates, or residues to show the allelopathic potential between weeds and crop (Ali and Sakri, 2010). Allelopathic substances enter the environment through a variety of pathways, including plant leaf volatiles, leaching, root exudation, and plant the decomposition (Wetson and Duke, 2003). Several plant species and groups have been shown to exhibit allelopathic activity; these plants may be used in ecological and agricultural systems (rice, 1995). The main factors responsible for low production of cereal crops are biotic and abiotic factors, weeds are considered as biotic factor that adversely affect growth, yield, and quality of grain through competition and allelopathy (Jauhar and Al-Mafrajy, 2023). Recently, results of researches illustrated that using herbicides to manage weeds may cause some environmental risks, health, water contamination and altering the growth of soil microorganisms (Nikneshan et al., 2011). Moreover, 290 weed species have developed herbicide resistance (Heap, 2005, Mennan et al., 2012). As a result, plants that have allelopathic properties can be used as an alternative bio or synthetic herbicide for weed controlling. Therefore, allelopathy may regard as a possible tool for weed management to increase crop production (Cheema and Khaliq, 2000). According to Batish, et al. (2001), weed growth negatively affected by the allelopathic potential of several plant species. There are more 360 species of Aloe plant. Aloe vera is a plant that have medicinal properties of the family Liliaceae family. It is a succulent herb, which cultivated in many location of the world (Mahor and Ali 2016). Since ancient times, Aloe vera's exudates have been used for medical and cosmetic uses (Tanaka et al. 2006). The gel of Aloe vera has different biological and physiological properties. The cell of leaf is parenchymatic have liquid yellowish latex and pure gel (Ni et al. 2004). The latex of Aloe vera contain essential amino acids, mono-and polysaccharide, lignin, macronutrient, vitamins, gibberellins and salicylic acid (Bouderau and Beland 2006). Generally, a vast number of secondary metabolite contain in medicinal plants (Li et al. 2022). Germination of black nightshade (Solanum nigrum L.) 19.6% were reduced by applying Aloe vera leaf water extracts at 10%, also reduced barley and lettuce seed germination , root length and seedling dry weight (Baličević et al. 2018). The aim of this study was to determine bio herbicidal effect of Aloe vera leaf gel's aqueous extract on seed germination and seeding growth of

wheat (Triticum aestivum L.), wild barley (Hordeum spontaneum) and black mustard (Brassica nigra) as an initial step toward developing eco-friendly bioherbicide.

2. MATERIALS AND METHODS

2.1: Extract preparation: thoroughly

Aloe vera was obtained from the garden of the college of Agricultural Engineering Sciences/ Salahaddin University-Erbil. Purified water was used to washing leaves carefully, then leaves were cut to portions to extraction the gelatinous ingredient inside it. The electronic blender was used to mix 200 milliliter of the gel extraction, next that, the distilled water was drenched into the gel to prepare crude extract (20%), the shaker (LAB SK-180-Pro) with 120 rpm was used to shaking the extract for 48 hours at 25 °C, then, four layers of cheesecloth and No.1.Whatman filter paper were used to filtration. Finally, the crude extract was diluted with distilled water to prepare (5%, 10%, 15%) concentration levels.

2.2: Bioassay:

The bioassay experiment was carried out in the laboratories Agricultural Engineering sciences on 15 October 2024. Arrange of *Aloe vera* aqueous extract concentrations (0%, 5%, 10%, 15% 20%) were prepared using distilled water. Twenty seeds of wheat (*Triticum aestivum* L.) Var. Hawler 2, wild barley (*Hordeum spontaneum*) and black mustard (*Brassica nigra*) placed in petri dished (9cm) contain (filter paper #1), then 10 ml of extract used to treat each of them. Finally they were covered by Para-film and located in growth chamber at (20º- 25 º C) darkly. In the days (2^{th}, 4^{th}, 7^{th}, 10^{th}) germination percentage were recorded. Finally, the length of radicle, plumule and seedling in (cm) were calculated, after that the seedling placed in electronic oven at (40º C) for 72 hours or until stabile weight dry weight of radicle, plumule and seedling were recorded in (mg).

2.3: Recorded data:

The data that recorded were:

1. Germination percentage = germinated seeds/ total tested seeds * 100

2. Germination speed (seed/day) = (N1/1 + N2/2 + N3/3 + N4/4), which N= number of germinated seed on 1^{st}, 2^{nd}, 3^{rd}, 4^{th} counting which they are after two days, four days, seven days and ten days from the beginning of the experiment (Norsworthy, 2003).

3. Inhibition Percentage = {(control germination percentage - treatment germination percentage) / control germination percentage} * 100

4. Radicle Length (cm). 5. Plumule Length (cm). 6. Seedling Length (cm). 7. Radicle Dry weight (mg). 8. Plumule Dry weight (mg). 9. Seedling Dry weight (mg).

10. Radicle Growth Inhibition Percentage= {1- (treated radicle dry weigh/ control radicle dry weight)}*100

11. Plumule Growth Inhibition Percentage = {1- (treated plumule dry weigh/ control plumule dry weight)}*100

12. Seedling Growth Inhibition Percentage= {1- (treated seedling dry weigh/ control seedling dry weight)}*100

2.4: Chromatography Analysis:

The chromatography analysis were conducted in the GAGRO veterinary center from Erbil city. Ten ml of *Aloe vera* leaf gel soaked in 100 ml of distilled water to prepare for 72 hours at 25°C, then, the extract solved with petroleum and diethyl ether to separate lipids and glycoside by filtering. The samples stored in dark tube and at (-4°C) until running by (HPLC) (Numan, 2018).

High performance liquid chromatography (UV Detector L-7000), (Pump L- 7100), (225nm wavelength), (column c18) and (0.5 mish) was used to identify phenolic compounds. (Mobile phase of acetonitrile 30%), (deionized water 70%) and (10 ml of solution inject) were used to identify. Lauric, myristic, palmitic, stearic, folic, linoleic, gallic, P-hydroxi banzoic, protocatechuic, syringic, vanillic acids were used as a library standard for poly phenolic compounds.

2.5: Statistical Analysis:

ANOA linear model were used to analyze the recorded data (SPSS). The design is completely randomized (CRD) each treatment replicate three times. Duncan's multiple range test at ($P \leq 0.01$) was selected for significance differences, determination, for each plant separately.

3. Results and Discussion:

3.1: Phenolic compound contents in leaf gel of *Aloe vera*.

The examination of was observed by high performance liquid chromatography (HPL).There are six bioactive compound were identified as shown the results in Table (1). It was noted that several compounds showed up in good amounts, such as compound Palmitic acid 22.61%, Folic acid 17.81%, linoleic acid 9.21% and Stearic acid 4.11%. While some of the other compounds showed up at very low amount such as the compounds Lauric acid 0.2% and Myristic acid 0.81. The studies revealed that phenolic substances significantly inhibit the growth, water relationship chlorophyll content and photosynthetic products of radish (Rasul and Ali, 2020). According to studies, allelopathic properties of gallic acid, chlorogenic acid, acid, caffeic acid, ferulic, and vanlic acid have been observed to significantly affect the prevention of germination and growth of some crops. Studies have shown that gallic acid's allelopathic characteristics have significant effects on certain plant growth and the inhibition of seed germination (Naby and Ali, 2020).

Table 1. Analysis HPL of an extract of leaf gel of Aloe vera

No.	Retention Time	Component Name	μg ml^{-1}
1	2.040	Lauric acid	0.26
2	2.732	Myristic acid	0.81
3	3.407	Palmitic acid	22.61
4	4.307	Stearic acid	4.11
5	5.073	Folic acid	17.81
6	5.923	linoleic acid	9.21

3.1: Influence of *Aloe vera* leaf gel's aqueous extract on germination and seedling growth characteristics of wheat (*Triticum aestivum* L.).

As shown the results in Table 2 the impact of different concentration levels of aqueous extract of leaf gel of *Aloe vera* on germination speed and some seedling parameters. The results show that some studied traits for (*Triticum aestivum* L.) were recorded to have a reduction significantly by the application higher concentration (20%). Where the data were (18.56 cm/day, 16.44 cm, 12.14 cm, 28.58 cm, 113.57 mg, 152.17 mg and 265.73 mg) for germination speed, (length of radicle, plumule and seedling) and (dry weight of radicle, plumule and seedling) respectively. However, growth inhibition percentage of radical and plumule were increased from 0.00% to 58.11% and 0.00% to 49.44% with increasing concentration from 0% to 20% of *Aloe vera* leaf gel's aqueous extract. Pervious study have reported the effects of phenolic substances on seed germination and seedling growth, highlighting the role in promoting plant growth and influencing hormonal activity (Subatin *et al*., 2002). The results are in agreement with (Yilmaz *et al*., 2013).

Table 2. Effect of concentration levels *Aloe vera* leaf gel's aqueous extract on germination speed and seedling growth characteristics of wheat (*Triticum aestivum* L.).

Concentration	Germination	Radicle	Plumule	Seedling	Radicle	Plumule	Seedling	Radicle	Plumule

levels %	speed (seed/day)	length (cm)	length (cm)	length (cm)	dry weight (mg)	dry weight (mg)	dry weight (mg)	growth inhibition %	growth inhibition%
0 %	18.56 a	16.44 a	12.14 a	28.58 a	113.57 a	152.17 a	265.73 a	0.00 e	0.00 d
5 %	17.28 ab	14.45 ab	10.55 ab	25.00 b	69.03 b	91.20 b	160.23 b	39.21 d	40.07 c
10 %	16.16 bc	13.51 bc	10.15 b	23.66 bc	65.73 c	82.70 c	148.43 c	42.12 c	45.65 b
15 %	15.09 c	11.29 cd	9.79 b	21.08 cd	51.43 d	82.43 c	133.87 d	54.71 b	45.83 b
20 %	14.41 c	9.93 d	8.75 b	18.68 d	47.57 e	76.93 d	124.50 e	58.11 a	49.44 a

* The different letters means significance differences between means.

The results from Figure 1 indicates that all concentration levels significantly affected the germination percentage, inhibition percentage and seedling growth inhibition. According to the findings with increasing concentrations from 0% to 20% decreased germination from 100% to 81.67% respectively. Whereas, inhibition of germination and seedling growth percentage increased from 0.00% to 18.33% and from 0.00% to 53.15% respectively. The effects were concentration dependent on all studied parameters (Ali, 2016). In addition, these significant effects could be due to the presences of bioactive compound in the leaf gel of *Aloe vera* Table 1, recent studies showed that allelochemical compound affected on germination percentage and other germination parameters (Joseph, 2010).

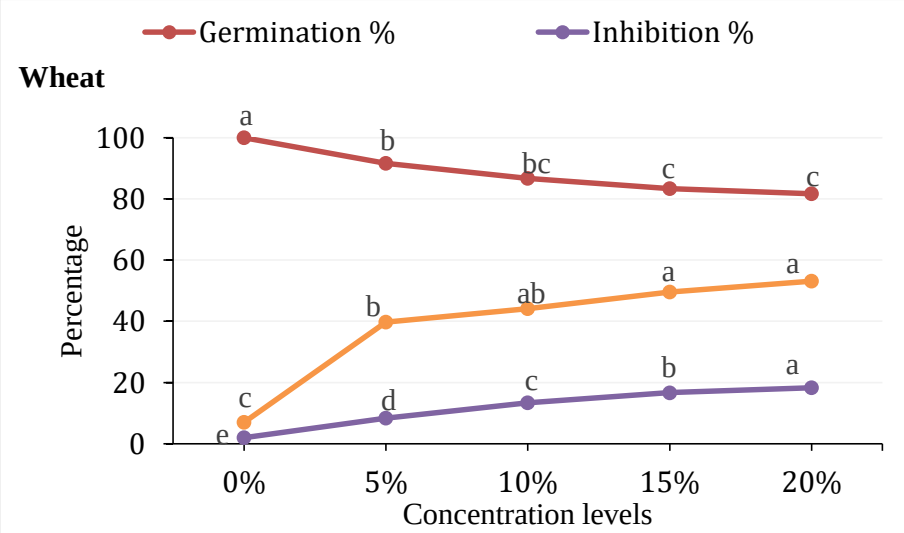

Figure 1 Effect of concentration levels *Aloe vera* leaf gel's aqueous extract on germination, inhibition percentage and seedling growth percentage of wheat (*Triticum aestivum* L.).

3.2: Influence of *Aloe vera* leaf gel's aqueous extract on germination and seedling growth characteristics of wild barely (*Hordeum* spontaneum).

As shown in Table 3 all studied characteristics were significantly ($P \leq 0.01$) affected by aqueous extracts of *Aloe vera* leaf gel. The results indicate that in wild barley germination speed, radicle, plumule, seedling length, radicle, plumule and seedling dry weight recorded lowest values 5.21 seed/ day, 3.21 cm, 8.71 cm, 11.92 cm, 5.73 mg 11.57 mg and 17.30 mg at 20% concentration respectively. While, the radical and plumule growth inhibition traits the recorded highest data are 80.05% and 88.46% respectively at the same concentration. The reduction in radicle and plumule length, radicle and plumule dry weight of the tested species can be because of the impact allelochemicals, for instance phenolic acids that appear in *Aloe vera* shoot extracts (Sarah, 2018). The results are in line with (Hamad *et al*., 2023).

Table 3. Effect of concentration levels *Aloe vera* leaf gel's aqueous extract on germination speed and seedling growth characteristics of barley (*Hordeum spontaneum*).

Concentration	Germination	Radicle	Plumule	Seedling	Radicle	Plumule	Seedling	Radicle	Plumule

levels %	speed (seed/day)	length (cm)	length (cm)	length (cm)	dry weight (mg)	dry weight (mg)	dry weight (mg)	growth inhibition %	growth inhibition%
0 %	18.56 a	9.69 a	14.41 a	24.10 a	28.77 a	100.17 a	128.93 a	0 e	0 e
5 %	10.83 b	7.33 b	11.14 b	18.47 b	24.30 b	49.43 b	73.73 b	15.54 d	50.65 d
10 %	9.61 b	5.85 bc	10.40 bc	16.25 bc	18.80 c	44.50 c	63.30 c	34.68 c	55.57 c
15 %	7.36 c	4.79 cd	9.55 bc	14.35 cd	9.60 d	36.23 d	45.83 d	66.62 b	63.83 b
20 %	5.21 d	3.21 d	8.71 c	11.92 d	5.73 e	11.57 e	17.30 e	80.05 a	88.46 a

* The different letters means significance differences between means.

The results from Figure 2 indicated that germination percentage, inhibition percentage and seedling growth inhibition of wild barley were significantly affected. Where, germination percentage reduced but inhibition percentage and seedling growth inhibition were increase with increasing concentration levels from 0% to 20%, that the highest record were 100%, 70% and 86.59% at control, and 20% concentration respectively. Allelochemicals found *Aloe vera* leaf gel extract might be the source of the decrease in seed germination and increasing germination inhibition. These compounds may have detrimental impact on physiological processes and cell division. In addition, secondary compound may be the cause of unstable in the cell membranes permeability studied crop and weed species (Kruse *et al*., 2000). These findings are in agreement with those of El-Rokiek, *et al*., (2024), who found that the effect of extracts on seed germination was concentration dependent.

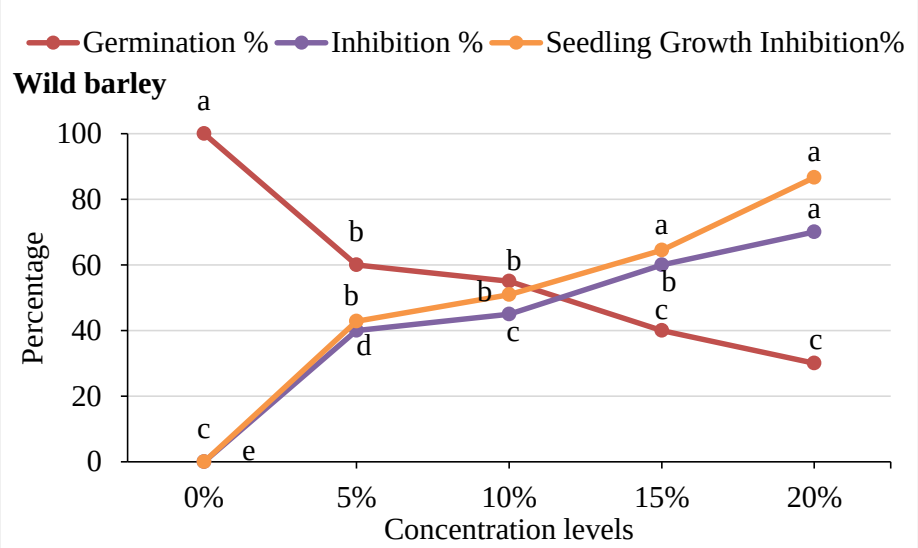

Figure 2 Effect of concentration levels *Aloe vera* leaf gel's aqueous extract on germination, inhibition percentage and seedling growth percentage of wild barley (*Hordeum spontaneum*).

3.3: Influence of *Aloe vera* leaf gel's aqueous extract on germination and seedling growth characteristics of black mustard (*Brassica nigra*).

As Shown the results in Table 4 aqueous extracts of *Aloe vera* leaf gel at all concentration levels significantly affected germination speed, radicle, plumule, seedling length, radicle, plumule and seedling dry weight of black mustard, lowest records 2.96 seed/ day, 0.63 cm, 1.20 cm, 1.83 cm, 2.93 mg 2.20 mg and 5.13 mg were showed at 20% concentration respectively. While, the radical and plumule growth inhibition recorded highest data were 57.50% and 84.93% respectively at the same concentration. According Singh *et al*., (2019) the extract of *Aloe vera* leaf significantly reduced the dry weight, shoot and root length of black nightshade and redroot pigweed seeds.

Table 4. Effect of concentration levels *Aloe vera* leaf gel's aqueous extract on germination speed and seedling growth characteristics of black mustard (*Brassica nigra*).

Concentration	Germination	Radicle	Plumule	Seedling	Radicle	Plumule	Seedling	Radicle	Plumule

levels %	speed (seed/day)	length (cm)	length (cm)	length (cm)	dry weight (mg)	dry weight (mg)	dry weight (mg)	growth inhibition %	growth inhibition %
0 %	18.21 a	3.37 a	4.53 a	7.90 a	6.90 a	14.60 a	21.50 a	0.00 e	0.00 e
5 %	8.59 b	3.10 a	4.23 a	7.33 a	6.07 b	10.20 b	16.27 b	12.09 d	30.14 d
10 %	7.37 b	2.87 a	4.13 a	7.00 a	5.50 c	7.30 c	12.80 c	20.29 c	50.00 c
15 %	5.12 c	1.50 b	2.37 b	3.87 b	4.10 d	3.50 d	7.60 d	40.59 b	76.03 b
20 %	2.96 d	0.63 c	1.20 b	1.83 c	2.93 e	2.20 e	5.13 e	57.50 a	84.93 a

* The different letters means significance differences between means.

Figure 3 shows that germination percentage, inhibition percentage and seedling growth inhibition of black mustard were significantly affected. Where, germination percentage reduced but inhibition percentage and seedling growth inhibition were increase with increasing concentration levels from 5% to 20%, that the highest value were 100%, 75% and 76.13% at control and 20% concentration respectively. The presences of allelopathic compounds in *Aloe vera* could be suffer the cell membrane permeability of tested plants during germination test. Respiration process, reduction RNA and ATP concentration and functions of secondary messengers that are essential for seed germination and seedling growth may alter with phytochemicals. (Abrahim *et al*., 2000).

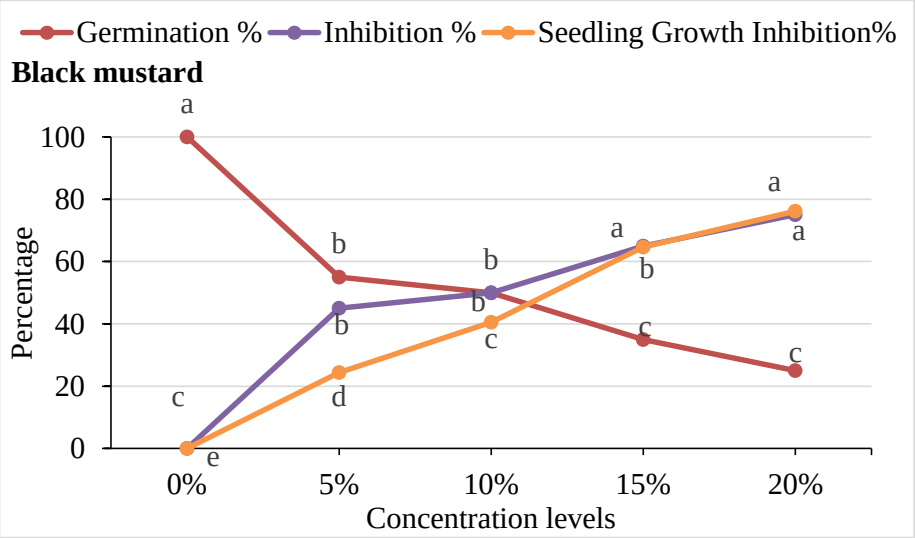

Figure 3 Effect of concentration levels *Aloe vera* leaf gel's aqueous extract on germination, inhibition percentage and seedling growth percentage of black mustard (*Brassica nigra*).

4. Conclusions:

Findings of this study illustrated that application *Aloe vera* leaf gel's aqueous extracts inhibited seed germination and some seedling growth characteristics of wheat, wild barley and black mustard especially at higher concentration levels 20%. Consequently, these findings could permit the development of bioherbicides to eliminating weeds as a natural tool for sustainable weed management, while ensuring this extract is not applied to wheat crops.

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