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Natural Control of Weed Invasions in Hyper-Arid Arable Farms: Allelopathic Potential Effect of *Conocarpus erectus* against Common Weeds and Vegetables

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Abstract: Utilization of plant allelopathic potential to control weed infestations provides an effective, cost-efficient, labor-free, and environmentally acceptable alternative to traditional chemical and mechanical methods. *Conocarpus erectus*, known as buttonwood, belongs to the Combretaceae family with high contents of phytochemicals and antioxidant activity. There have been no studies on the allelopathic potential of *C. erectus*. The present study (1) examined the allelopathic potential of *C. erectus* against selected weeds (*Chenopodium murale* and *Amaranthus viridis*) and crops (*Solanum lycopersicum* and *Cucumis sativus*) via investigating the growth inhibition ability of its aqueous extract, and (2) identified the potential allelochemicals found in this plant. Aqueous extracts were prepared from leaves, roots, and seeds of *C. erectus* by immersing the dried powder of the examined plant parts in sterile distilled water for 24 h on a shaker set to 180 rpm. The resulting filtrate was considered as 100% solution, and then dilutions were made to various concentrations (75%, 50%, and 25%). *C. erectus* leaves and seeds showed the highest rate of inhibition at all concentrations against *Chenopodium murale* and *Amaranthus viridis* grown in either Petri dishes or pots. Conversely, all the studied extracts did not show any toxic effects against tomato and cucumber plants grown in pots. In Petri dishes, a slight reduction in growth was observed. HPLC analysis of total phenolic contents in *C. erectus* methanolic extracts showed that leaves have the highest contents of gallic acid, caffeic acid, and ferulic acid (153.963, 69.135, and 39.801 ppm, respectively). The finding of the current study demonstrated that the part of the plant and the concentration of extraction have a significant effect on phytotoxicity. The positive results of this study might be used to develop environmentally-friendly herbicides for agricultural purposes.

Keywords: allelopathy; invasive plants; weeds; *Concarpus*; phenolics

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1. Introduction

Non-indigenous plants (including weeds) seriously threaten their neighboring plants. In most cases, invasive plants possess several phytotoxic compounds that hinder the germination and seedling growth of surrounding plant species at both ecosystem and species levels. Crop plants face many obstacles during their growth period, especially in the fields of hyper-arid desert areas such as Saudi Arabia, where many weeds are aggressively invading these fields due to the availability of niches, moisture, water, nutrients, and shading in these new habitats. Weeds are thus one of the most significant problems that plants encounter during their growing phase. Weeds compete for their resources with crops, as they emerge rapidly and cause a significant decrease in the crop yield, with losses incurring up to 34% each year and thereby affecting global crop production [1]. Hence, weed management has always been a major challenge in agriculture fields. Polyculture and crop

rotation are two traditional weed management techniques that are desirable [2]. However, due to the growing demand for food supply, numerous methods have been developed, adopted, and implemented. Some of them, such as mechanical weeding and herbicides, have shown excellent results in the past few years [3–5]. Although hand and mechanical weeding gives good results and is safe, this process is expensive and labor-intensive. Synthetic herbicides on the other hand have shown excellent results and have been used all over the world extensively to meet the demand for crop production [6–8]. The overuse of synthetic chemical herbicides has shown a negative impact on human health and the environment [9–14]. Recently, world pesticide production and consumption in world markets have increased remarkably. Recent statistics show that 45% of the expenditures has been made for herbicides, followed by 14% for insecticides, and 10% for fungicides. Herbicides had the largest portion of global consumption in the world market, which has reached 24,727 million and is constantly increasing [15]. Hence, there is a need to adopt safer yet cheaper and more effective ways to control negative effects and utilize the positive effects of allelopathy such as searching for alternative weed management strategies.

A new approach to mitigate the adverse impacts of synthetic herbicides on crop production is by using natural herbicides [16,17]. The most dominant and invisible challenge on competition between crops and weeds in ecosystems occurs by allelopathy [18]. Allelopathy is a biological and natural phenomenon that constitutes an important subdiscipline of chemical ecology. The eco-physiological interactions between higher plants are mediated via secretion of certain chemical compounds known as "allelochemicals". Those chemical compounds could be found naturally in many parts in plants, e.g., roots, seeds, leaves, and stems, with different portions [9,10,13,19]. Most natural allelochemicals derived from plants are not toxic to humans, do not pollute the environment (soil and water), and are easily biodegradable [20–22]. They can serve as an excellent, safe, and environmentally-friendly weed management strategy [23]. The major application of plant allelopathy is the identification of allelochemical activity of phenolic compounds in plant extracts and using them as herbicides or for crop protection [9,10,13,19,24]. Plants or weeds with phytotoxic natural products have great potential to be exploited for weed management [25,26]. *Chenopodium album*, *Amaranthus retroflexus*, and *Cynodon dactylon* were shown to produce allelopathic compounds, which caused reduction in crops, with *C. dactylon* having the most adverse effects compared with *A. retroflexus* and *C. album* [27]. Nevertheless, *C. album* showed allelopathic effects that damaged different plant parts. Allelochemicals affect growth, development, reproduction, survival, and distribution of other plants and microorganisms in agricultural systems or natural communities [28,29]. Previous literature has shown that some of these compounds may stimulate crop production and/or inhibit weed growth [30–33]. In most cases, the allelopathic compounds regulate the growth and development of plants, e.g., photosynthesis, respiration, transpiration, mineral uptake, inhibition or stimulation of specific enzyme activity, protein synthesis, and DNA or RNA synthesis [17,34].

Utilization of plant allelopathic potential to control weed infestations provides an effective, cost-efficient, labor-free, and environmentally-acceptable alternative to the traditional chemical and mechanical methods [35]. Furthermore, plants having allelopathic effects against weeds may have increased agricultural output and play important roles in maintaining ecological stability [36,37].

Conocarpus erectus, commonly known as buttonwood, is a member of the Combretaceae family that grows as a shrub but may develop to be a 20-m-tall tree. This species originates from Florida, Mexico, and the West Indies and was introduced to Saudi as urban greening in roads and now spreads as an exotic plant in all regions of Saudi Arabia and other Arab countries. *C. erectus* has high contents of phytochemicals and antioxidant activity [17,38]. No reports have examined the potential allelopathic activity of *C. erectus*. A preliminary study indicated the antifungal and herbicidal potential of extracts of *C. pennisetiformis* [39]. Methanolic extracts of all the parts of *C. pennisetiformis* reduced the fungal biomass in a variable manner, suggesting an alternative control strategy of fusarium wilt in tomato caused

by *Fusarium oxysporum* f. sp. *Lycopersici* [40]. Moreover, leaf extracts of *C. lancifolius* (Engl.) inhibited the seed germination of *Zea mays* and *Vigna sinensis* with excellent antifungal activity against *Sclerotinia sclerotiorum*, *Rhizoctonia solani*, and *F. oxysporum* f. sp. *Lycopersici*.

Plant allelopathy involves the interaction between the donor and the target plants, which may exert either a positive (e.g., crop protection, weed control or crop re-establishment) or negative effect (e.g., autotoxicity, biological invasion, soil degradation through allelochemicals) [29]. The most reliable and common method for the evaluation of allelopathic effects is by examining the inhibitory effects of different plant parts' extracts against growth of weeds and cultivated crops either in vitro or in pots. Therefore, the current study (1) examined the allelopathic potential of *C. erectus* against selected weeds (*Chenopodium murale* and *Amaranthus viridis*) and crops (*Solanum lycopersicum* and *Cucumis sativus*) via investigating the growth inhibition ability of its aqueous extract, and (2) identified the potential allelochemicals found in this plant.

2. Materials and Methods

2.1. Collection of Plant Materials

Leaves, roots, and seeds of the donor plant (*Conocarpus erectus*) and seeds of target weeds (*Chenopodium murale*, *Amaranthus viridis*) were collected locally from different regions in Riyadh, Saudi Arabia, during 2019–2020. Seeds of target crops were obtained from a commercial seed company (*Solanum lycopersicum*, AC 55 VF, Pomodoro) and (*Cucumis sativus*, beta Alpha, Agrimaxspin, Dallas seeds). Seeds of target weeds and crops were sterilized using ethanol solution (70%) for 2 min. Then, 2.0% sodium hypochlorite (NaOCl) was added for 5 min. Sterile distilled water was used to rinse seeds five times.

2.2. Preparation of Aqueous Extracts

Leaves, roots, and seeds of the donor plant (*Conocarpus erectus*) were collected from three different plants and extracted separately. The collected plant parts were washed thoroughly under running tap water and then with sterile distilled water and then dried in shade for 2–3 weeks at room temperature. The dried plant parts were grinded. Different aqueous extracts were prepared by immersing the dried powder (1 g) of the examined plant parts in sterile distilled water (100 mL) for 24 h on a shaker set to 180 rpm. The extracts were then filtered to remove debris, initially with cheese cloth followed by no. 1 Whatman filter paper [17]. The resulting filtrate was considered as 100% solution, and then dilutions were made to various concentrations (75%, 50%, and 25%). These reconstituted extracts were used for bioassays and growth experiments.

2.3. Petri-Dish Bioassay

Five seeds from target plants with three replicates were placed in Petri dishes with a double layer of sterile filter paper. Then, 5 mL of each concentration of donor plant extracts (leaves, roots, or seeds) was added in Petri dishes. The control from each target plant was treated with distilled water only. The Petri dishes were placed under cool fluorescent light (350 µmol m^{-2} s^{-1}) at 25 °C with a 12/12 h (light/dark) photoperiod. Seedling and radical growth of recipients was observed after treatment for 7–14 days using a ruler. Each treatment was replicated five times. The experiment was laid out in completely randomized design (CRD).

2.4. Growth Inhibition by Aqueous Extracts

The extracts of each donor plant with different concentrations (100%, 75%, 50%, 25% v/v) were mixed separately in plastic pots (30 cm in diameter) with sterilized potting soil (pH 5.0–6.0, Bass Van Buuren, The Netherlands). Seeds of target plants were planted in these pots. Each treatment was replicated five times. Each replicate consisted of three pots, each containing five seeds of target plants. Pots were watered using sterilized distilled water every two days for 7–14 days. The lengths of roots and shoots were measured. The experiment was laid out in randomized complete block design (RCBD).

2.5. Phenolic Acids Analysis via HPLC

The phenolic acids were quantified by HPLC with UV detection (Alliance 2695 Separations Module, Waters Instruments, Inc., Milford, MA, USA). The analyses were carried out on a reverse-phase C18 column (Pinnacle C18 column, 250 × 4.6 mm, 5 µm, Shimadzu, Kyoto, Japan). The mobile phase was composed of (A) 2% acetic acid in ultra-pure water (acidified water), and (B) acetonitrile and methanol (65:35, v/v) using a flow rate of 1 mL/min. The optimized gradient program was as follows: 0–10 min (10–45% B), 10–20 min (45–90% B), 20–23 min (90–10% B), and 23–25 min (10% B). Samples were injected into the system as 10 µL, and the analysis was performed at a single wavelength of 280 nm.

2.6. Statistical Analysis

All the collected data were analyzed using Statistical Package for the Social Sciences SPSS® Statistics 28 (IBM, Armonk, NY, USA). Two-way analysis of variance (ANOVA) was applied with the part of the donor plant and the solution concentration as the two independent factors. Means were compared using Duncan's Multiple Range Test (DMRT) with significance level of 0.05.

3. Results

3.1. Bioassay and Growth Experiments

The phytotoxic potential of *C. erectus* (leaves, roots, and seeds) aqueous extracts on selected weeds and crops was examined based on changes in shoot and root lengths of seedlings. Average lengths of target seedlings treated with different extracts compared to controls were calculated to confirm the phytotoxic effects of donor plant extracts in each concentration. Shoot and root lengths of *C. murale* seeds in Petri dishes were significantly inhibited by all concentrations of *C. erectus*. Average length of shoots gradually decreased by roughly 90–100% after treatment with leaf extract in comparison to control untreated seedlings (Table 1). Moreover, the average length of target seed shoots decreased compared to the control by approximately 73% after treating with seed extract and 53% with root extracts of donor plants. Root lengths of *C. murale* decreased by 94% and 96% with leaf and seed extracts at 100% concentration, respectively, and 58% with root extract at the same concentration. *Conocarpus* leaves and seeds showed the highest rate of inhibition at all concentrations, which was above 50%. In contrast, the lowest inhibition rate was by *C. erectus* root extract on *C. murale* seeds (Figure 1).

Table 1. Effect of leaf, root, and seed extracts of *Conocarpus erectus* with different concentrations (100, 75, 50, and 25%) on *Chenopodium murale* seed growth in vitro or in pots.

Part	Concentration (%)	Petri Dishes		Pots	
		Shoot Length (cm)	Root Length (cm)	Shoot Length (cm)	Root Length (cm)
Leaves	0	4.83 ± 0.55 a	2.97 ± 0.61 a	4.96 ± 0.56 a	2.67 ± 0.84 a
	25	1.62 ± 0.38 e	0.72 ± 0.12 d	3.51 ± 0.26 d	1.44 ± 0.12 b
	50	1.10 ± 0.82 ef	0.44 ± 0.17 e	3.96 ± 0.49 b	1.24 ± 0.17 c
	75	0.92 ± 0.57 f	0.23 ± 0.14 g	4.07 ± 0.49 b	1.05 ± 0.14 d
	100	0.57 ± 0.28 g	0.22 ± 0.13 g	3.84 ± 0.48 b	0.56 ± 0.13 h
Roots	0	4.83 ± 0.55 a	2.97 ± 0.61 a	4.96 ± 0.56 a	2.67 ± 0.84 a
	25	3.31 ± 0.38 b	2.90 ± 0.43 a	3.45 ± 0.62 d	0.97 ± 0.43 e
	50	3.10 ± 0.40 b	2.04 ± 0.52 b	3.29 ± 0.90 e	0.90 ± 0.52 f
	75	2.60 ± 1.03 c	1.51 ± 0.20 c	3.64 ± 0.66 c	0.73 ± 0.20 g
	100	2.29 ± 0.86 de	1.29 ± 0.29 c	3.35 ± 0.73 e	0.69 ± 0.29 g

Table 1. *Cont.*

Part	Concentration (%)	Petri Dishes		Pots	
		Shoot Length (cm)	Root Length (cm)	Shoot Length (cm)	Root Length (cm)
Seeds	0	4.83 ± 0.55 a	2.97 ± 0.61 a	4.96 ± 0.56 a	2.67 ± 0.84 a
	25	2.22 ± 0.20 de	0.54 ± 0.09 e	3.29 ± 0.48 e	1.50 ± 0.09 b
	50	2.07 ± 0.18 de	0.48 ± 0.08 e	2.97 ± 0.71 e	1.22 ± 0.08 c
	75	1.55 ± 0.56 e	0.34 ± 0.06 f	2.67 ± 0.73 f	0.95 ± 0.06 e
	100	1.44 ± 0.62 e	0.27 ± 0.02 g	2.55 ± 1.93 f	0.54 ± 0.02 h
F-values	Part	95.73	89.60	13.95	1.55
	Concentration	210.58	101.12	27.27	45.92
	Part × Concentration	6.39	8.42	2.08	0.66
p-values	Part	0.00	0.00	0.00	0.35
	Concentration	0.00	0.00	0.00	0.00
	Part × Concentration	0.00	0.00	0.01	0.94

Means in the same column followed by the same letter are not significantly different ($p \leq 0.05$).

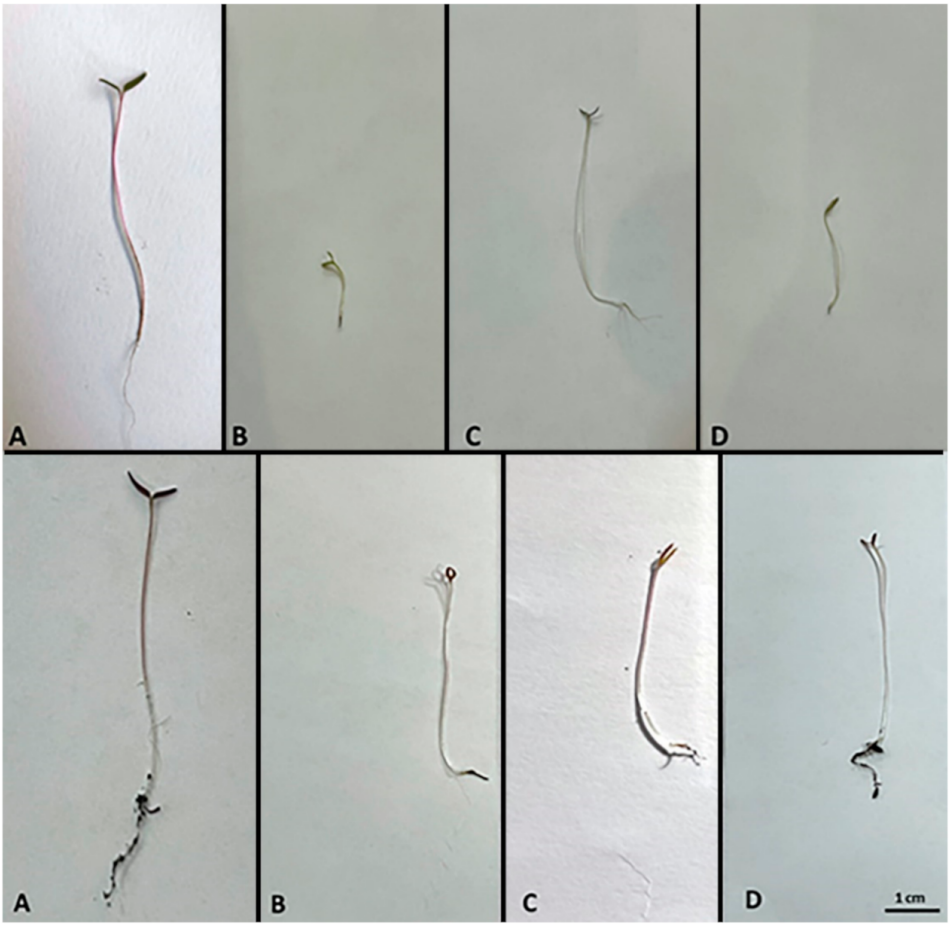

Figure 1. Effect of different *Conocarpus erectus* extracts on the growth of *Chenopodium murale* seedlings grown in either Petri dishes (top) or pots (bottom): (**A**) control, (**B**) 100% leaf extract, (**C**) 100% root extract, (**D**) 100% seed extract.

In pots, root lengths of *C. murale* plants were significantly inhibited by *Conocarpus* extracts at all concentrations. Similarly, average lengths of target roots showed significant inhibition after exposure to leaf, seed, and root extracts. Inhibition percentage of leaf extracts was about 83%. However, seed and root extracts inhibited donor plant growth by 77% and 68%, respectively (Table 1). Nevertheless, the inhibition rates on *C. murale* shoot lengths were lower than 50% by all the studied extracts. Generally, *C. erectus* extract

significantly inhibited root growth of *C. murale* plants grown in pots at 100%, 75%, and 50% concentrations, but did not have an inhibition effect on shoot lengths. In general, shoots and roots of target plants were inhibited after exposure to extracts of all parts of the donor plant and at each concentration in Petri dishes. The target plants grown in pots, on the other hand, exhibited a substantial decrease in root length at all concentrations. Moreover, in both Petri dishes and pots, *C. erectus* leaf extracts showed the highest inhibition rates against *C. murale* growth, followed by seed extracts (Figure 1). The least adverse impact to seeds germination was imposed by the root extracts.

Shoots and roots of *A. viridis* seedling lengths were significantly inhibited by leaf extracts at all concentrations in Petri dishes (Table 2). Average shoot length decreased approximately over 80% by 100% and 75%, and over 50% by 50% and 25% of *C. erectus* leaf extracts. Root lengths of *Amaranthus* seedlings were significantly inhibited by 82% and 93% after exposure to 50% and 100% leaf extracts, respectively. Uniquely, at 25% of *C. erectus* leaf extract, *Amaranthus* root lengths were inhibited by more than 50%. Moreover, *C. erectus* seed extracts significantly inhibited 70% of *Amaranthus* root lengths at concentrations of 50%, 75%, and 100%, and over 60% at all concentrations. Nevertheless, shoots of the target seeds were inhibited only by 100% *C. erectus* seed extracts (Figure 2). Similarly, root extract inhibited root lengths at 100% concentration. Root extract had the lowest inhibition effect on shoot and root lengths at less than 50%.

Table 2. Effect of leaf, root, and seed extracts of *Conocarpus erectus* with different concentrations (100, 75, 50, and 25%) on *Amaranthus viridis* seed growth in vitro or in pots.

Part	Concentration (%)	Petri Dishes		Pots	
		Shoot Length (cm)	Root Length (cm)	Shoot Length (cm)	Root Length (cm)
Leaves	0	2.13 ± 0.11 a	1.43 ± 0.13 a	3.75 ± 0.43 a	1.81 ± 0.17 a
	25	0.95 ± 0.02 c	0.68 ± 0.09 c	3.37 ± 0.56 b	0.77 ± 0.08 c
	50	0.72 ± 0.13 c	0.35 ± 0.01 e	3.43 ± 0.43 b	0.7 ± 0.09 cd
	75	0.36 ± 0.07 d	0.26 ± 0.05 e	2.96 ± 1.25 b	0.61 ± 0.13 d
	100	0.33 ± 0.03 d	0.20 ± 0.02 e	2.40 ± 1.66 c	0.55 ± 0.17 d
Roots	0	2.13 ± 0.11 a	1.43 ± 0.13 a	3.75 ± 0.43 a	1.81 ± 0.17 a
	25	2.12 ± 0.28 a	1.01 ± 0.15 b	2.50 ± 0.61 c	0.51 ± 0.06 de
	50	2.12 ± 0.43 a	1.11 ± 0.42 b	2.63 ± 1.24 c	0.5 ± 0.19 e
	75	2.10 ± 0.35 a	0.78 ± 0.13 c	2.89 ± 1.46 b	0.59 ± 0.2 d
	100	2.09 ± 0.65 a	0.74 ± 0.17 c	2.74 ± 1.33 bc	0.59 ± 0.19 d
Seeds	0	2.13 ± 0.11 a	1.43 ± 0.13 a	3.75 ± 0.43 a	1.81 ± 0.17 a
	25	1.37 ± 0.06 b	0.54 ± 0.07 d	3.81 ± 0.47 a	1.23 ± 0.06 b
	50	1.25 ± 0.05 b	0.47 ± 0.06 d	3.04 ± 1.28 b	0.87 ± 0.42 c
	75	1.17 ± 0.01 b	0.45 ± 0.10 d	2.32 ± 1.38 c	0.52 ± 0.32 de
	100	0.81 ± 0.29 c	0.41 ± 0.17 d	3.28 ± 0.48 b	0.35 ± 0.15 f
F-values	Part	243.21	56.47	1.81	3.88
	Concentration	65.58	99.81	5.29	105.40
	Part × Concentration	17.99	5.76	2.07	4.74
p-values	Part	0.00	0.00	0.23	0.01
	Concentration	0.00	0.00	0.01	0.00
	Part × Concentration	0.00	0.00	0.04	0.00

Means in the same column followed by the same letter are not significantly different ($p \leq 0.05$).

In pots, root lengths of *Amaranthus* plants were inhibited by more than 50% after treatment with *C. erectus* leaf, seed, and root extracts at 100%, 75%, and 50% concentrations. In addition, roots exposed to 100% and 75% of leaf extracts were shorter by over 70% as compared to control seedlings, while those treated with 50% and 25% of leaf extracts were 60% shorter than controls. Furthermore, 100% of *C. erectus* seed extract inhibited about 85% of *A. viridis* root lengths, while the other concentrations decreased root lengths by more than 50%. *C. erectus* root extract at all concentrations inhibited root length of *Amaranthus* by

more than 70%. Moreover, shoot length of *Amaranthus* seedlings was inhibited by less than 50% when exposed to all *C. erectus* extracts. The highest inhibition rate was found after exposure to seed extracts (Figure 2). *C. erectus* leaf and root extracts had similar effects at 100% and 75% concentrations.

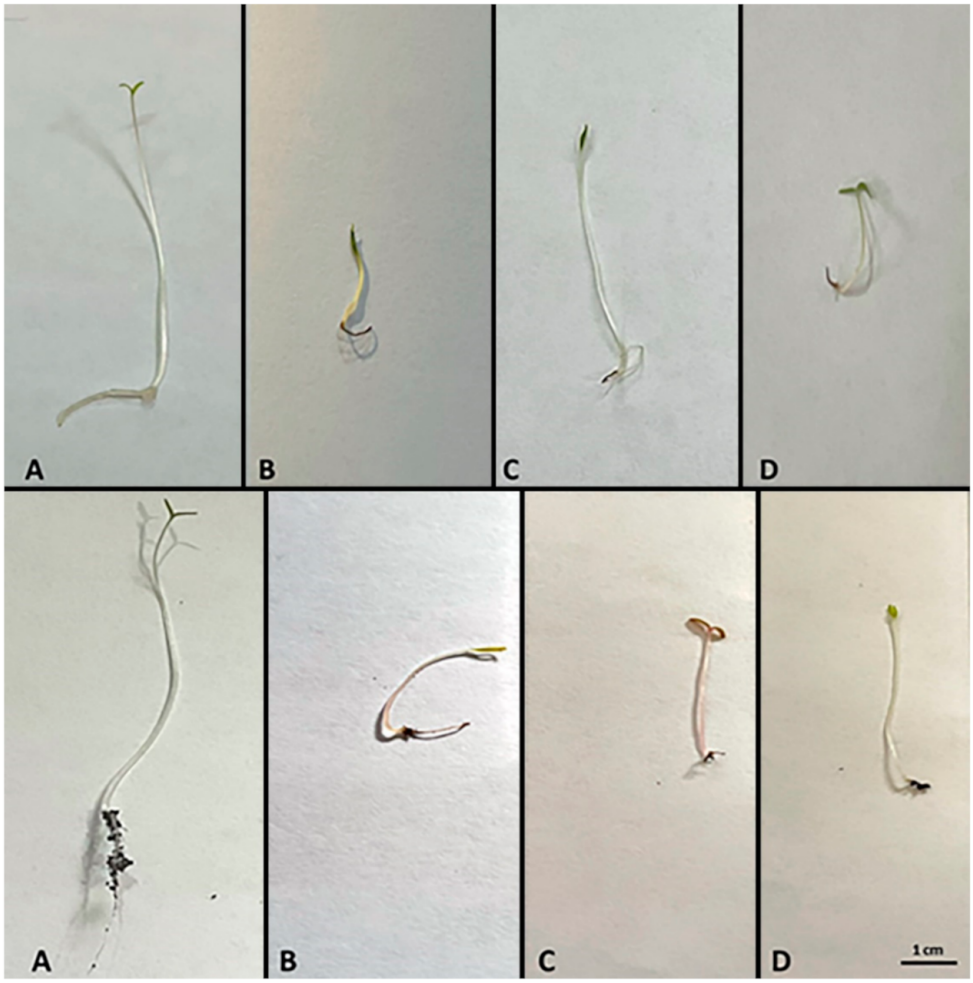

Figure 2. Effect of different *Conocarpus erectus* extracts on the growth of *Amaranthus viridis* seedlings grown in either Petri dishes (top) or pots (bottom): (**A**) control, (**B**) 100% leaf extract, (**C**) 100% root extract, (**D**) 100% seed extract.

Tomato seedling growth was observed to detect the effect of allelopathy of *C. erectus* extracts. In Petri dishes, the leaf extracts with concentrations higher than 50% showed adverse impacts on tomato growth (Table 3). Conversely, root extracts of *C. erectus* showed reduction of less than 50% on tomato shoot and root lengths. On the other hand, the inhibition percentage on tomato growth in pots showed that all donor plant parts (leaves, roots, seeds) and all concentrations did not inhibit tomato shoot and root growth since the inhibition rates of all donor parts at all concentration were less than 10% on shoot lengths and less than 33% on root lengths (Figure 3).

Table 3. Effect of leaf, root, and seed extracts of *Conocarpus erectus* with different concentrations (100, 75, 50, and 25%) on tomato (*Solanum lycopersicum*) seed growth in vitro or in pots.

Part	Concentration (%)	Petri Dishes		Pots	
		Shoot Length (cm)	Root Length (cm)	Shoot Length (cm)	Root Length (cm)
Leaves	0	7.73 ± 0.97 a	6.81 ± 1.05 a	9.62 ± 0.43 a	3.37 ± 0.46 a
	25	6.95 ± 0.79 a	2.70 ± 0.59 a	8.99 ± 0.95 a	3.56 ± 0.59 a
	50	3.14 ± 0.57 c	1.58 ± 0.37 c	9.18 ± 0.76 a	3.18 ± 0.67 a
	75	1.65 ± 0.48 d	1.10 ± 0.28 d	9.40 ± 1.37 a	3.38 ± 0.78 a
	100	1.54 ± 0.23 d	0.58 ± 0.03 d	9.03 ± 1.27 a	3.04 ± 0.12 a
Roots	0	7.73 ± 0.97 a	6.81 ± 1.05 a	9.62 ± 0.43 a	3.37 ± 0.46 a
	25	8.35 ± 1.85 a	6.66 ± 1.65 a	9.44 ± 0.32 a	3.06 ± 0.41 a
	50	8.39 ± 1.76 a	5.30 ± 1.56 a	9.33 ± 0.67 a	2.89 ± 0.38 a
	75	7.73 ± 1.20 a	5.10 ± 1.00 a	8.74 ± 1.19 a	2.78 ± 0.16 a
	100	7.58 ± 0.92 a	3.96 ± 0.72 a	9.31 ± 0.95 a	3.01 ± 0.51 a
Seeds	0	7.73 ± 0.97 a	6.81 ± 1.05 a	9.62 ± 0.43 a	3.37 ± 0.46 a
	25	7.42 ± 1.11 ab	3.21 ± 0.91 ab	10.38 ± 0.92 a	2.5 ± 0.18 a
	50	5.83 ± 0.92 b	2.55 ± 0.72 b	9.90 ± 0.95 a	2.82 ± 0.82 a
	75	5.83 ± 0.49 b	1.84 ± 0.29 b	10.02 ± 0.9 a	2.41 ± 0.34 a
	100	4.89 ± 0.58 b	1.52 ± 0.38 b	8.67 ± 1.33 a	2.3 ± 0.34 a
F-values	Part	233.83	143.86	3.00	9.36
	Concentration	79.39	120.17	1.70	3.06
	Part × Concentration	28.11	9.56	1.83	1.23
p-values	Part	0.00	0.00	0.18	0.10
	Concentration	0.00	0.00	0.34	0.54
	Part × Concentration	0.00	0.02	0.25	0.96

Means in the same column followed by the same letter are not significantly different ($p \leq 0.05$).

Figure 3. Effect of different *Conocarpus erectus* extracts on the growth of tomato seedlings grown in either Petri dishes (top) or pots (bottom): (**A**) control, (**B**) 100% leaf extract, (**C**) 100% root extract, (**D**) 100% seed extract.

Table 4 shows the effects of *C. erectus* extracts on cucumber growth. Average shoot lengths of cucumber plants grown in Petri dishes were inhibited by root extracts at all concentrations, and only at 100% of leaf and seed extracts. On the contrary, cucumber growth in pots was not inhibited by any donor plant part or any concentration. Shoot and root length rate of inhibition were less than 50%. In addition, root extract had the highest rate of inhibition, which was 13% at 100% concentration. Leaf and seed extracts, on the other hand, exhibited no inhibition at any concentration (Figure 4).

Table 4. Effects of leaf, root, and seed extracts of *Conocarpus erectus* with different concentrations (100, 75, 50, and 25%) on cucumber (*Cucumis sativus*) seed growth in vitro or in pots.

Part	Concentration (%)	Petri Dishes		Pots	
		Shoot Length (cm)	Root Length (cm)	Shoot Length (cm)	Root Length (cm)
Leaves	0	7.46 ± 0.76 b	10.62 ± 3.05 b	12.94 ± 0.93 c	11.61 ± 1.31 a
	25	10.05 ± 1.14 a	17.1 ± 1.43 a	15.31 ± 0.75 b	10.67 ± 0.08 a
	50	6.58 ± 2.13 c	6.43 ± 1.56 c	16.51 ± 1 c	9.95 ± 1.43 a
	75	5.46 ± 1.66 c	6.21 ± 0.99 c	14.24 ± 1.69 b	10.74 ± 0.77 a
	100	2.47 ± 1.72 ef	1.05 ± 0.75 g	14.81 ± 1.58 b	9.84 ± 1.70 a
Roots	0	7.46 ± 0.76 b	10.62 ± 0.75 b	12.94 ± 0.93 c	11.61 ± 1.31 a
	25	3.28 ± 0.96 ef	4.26 ± 1.15 e	13.28 ± 1.85 c	10.80 ± 1.43 a
	50	4.96 ± 0.85 d	5.32 ± 1.44 d	11.29 ± 1.15 d	8.28 ± 0.80 a
	75	2.89 ± 0.65 ef	2.06 ± 1.73 f	12.06 ± 1.59 c	8.58 ± 1.51 a
	100	2.14 ± 0.38 f	1.73 ± 0.89 f	11.38 ± 1.26 d	9.25 ± 1.28 a
Seeds	0	7.46 ± 0.76 b	10.62 ± 0.75 b	12.94 ± 0.93 c	11.61 ± 1.31 a
	25	8.97 ± 2.51 a	11.77 ± 0.97 b	14.26 ± 1.37 b	10.00 ± 0.85 a
	50	7.69 ± 0.93 b	6.45 ± 1.74 c	15.11 ± 0.83 b	10.00 ± 1.29 a
	75	5.15 ± 1.85 d	3.63 ± 1.65 e	14.26 ± 1.33 b	10.25 ± 1.33 a
	100	3.47 ± 1.54 de	3.32 ± 1.54 e	14.44 ± 1.46 b	10.00 ± 1.46 a
F-values	Part	21.29	25.23	36.10	4.27
	Concentration	29.96	76.01	3.97	6.78
	Part × Concentration	5.22	12.94	4.27	3.06
p-values	Part	0.00	0.00	0.00	0.22
	Concentration	0.00	0.00	0.00	0.13
	Part × Concentration	0.00	0.00	0.00	0.95

Means in the same column followed by the same letter are not significantly different ($p \leq 0.05$).

3.2. Total Phenolic Content

Total phenolic contents of *C. erectus* methanolic extracts were quantified by HPLC. Table 5 shows the contents of phenolic compounds in leaf, root, and seed extracts of *C. erectus*. The calculated phenolic contents of methanol extracts of leaf parts with reference to gallic acid, caffeic acid, and ferulic acid were 153.963, 69.135, and 39.801 ppm, respectively. Gallic acid clearly showed the highest concentration in leaves followed by caffeic acid, while ferulic acid had the lowest concentration (Figure 5). Root extracts showed 15.912 ppm gallic acid equivalence, 8.394 ppm caffeic acid equivalence, and 43.313 ppm ferulic acid equivalence. The highest concentration of phenolic compounds in roots was that of ferulic acid, while the methanolic extracts of seeds had 19.668, 43.219, and 16.784 ppm of gallic acid, caffeic acid, and ferulic acid, respectively. The highest concentration was caffeic acid, and reference gallic acid and ferulic acid had low concentrations compared to caffeic acids. Comparing the extract phenolic content of the three parts, the highest was gallic acid, which was in leaves, followed by caffeic acid in leaves followed by seed extracts. Ferulic acid showed the highest concentration in root extracts.

Figure 4. Effect of different *Conocarpus erectus* extracts on the growth of cucumber seedlings grown in either Petri dishes (top) or pots (bottom): (**A**) control, (**B**) 100% leaf extract, (**C**) 100% root extract, (**D**) 100% seed extract.

Table 5. Phenolic contents in leaf, root, and seed aqueous extracts of *Conocarpus erectus* as revealed by HPLC analysis.

Phenolic Acid	Leaves	Roots	Seeds	F-Value	*p*-Value
Gallic acid (ppm)	153.963 ± 10.18 a	15.912 ± 1.23 b	19.668 ± 2.11 b	33.89	0.001
Caffeic acid (ppm)	69.135 ± 5.34 a	8.394 ± 0.99 c	43.219 ± 3.13 b	21.13	0.000
Ferulic acid (ppm)	39.801 ± 2.09 a	43.313 ± 2.12 a	16.784 ± 0.15 b	30.11	0.000

Means in the same row followed by the same letter are not significantly different ($p \leq 0.05$).

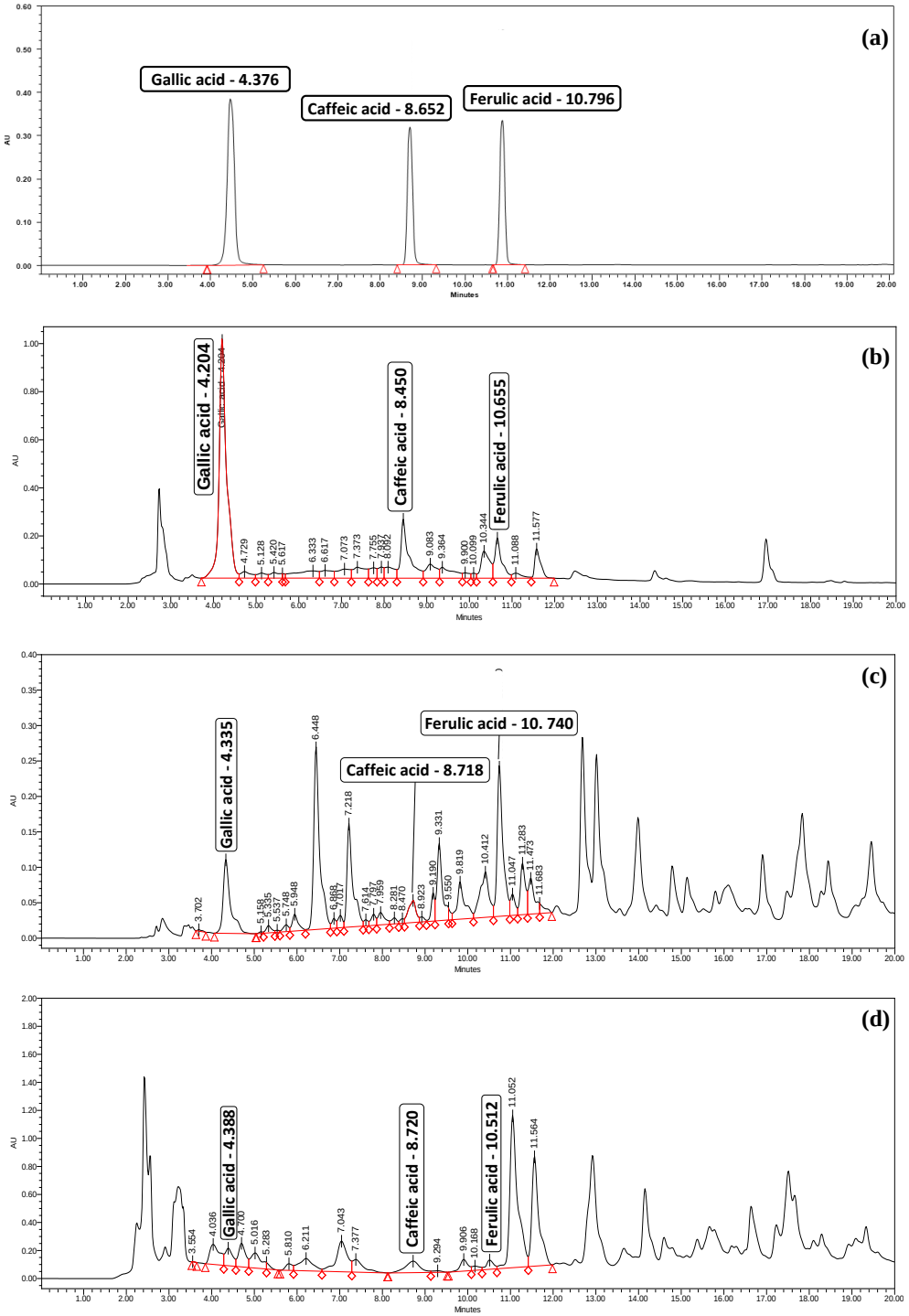

Figure 5. (**a**) Chromatograms of standard gallic acid, caffeic acid, and ferulic acid. (**b**–**d**) Chromatograms showing the concentrations of these acids in aqueous extracts of leaves (**b**), roots (**c**), and seeds (**d**) of *Conocarpus erectus*. The x-axis shows retention time in minutes, and the y-axis shows the absorbance units (a signal corresponds to the response created by the detector) at 280 nm.

4. Discussion

Allelopathy in agroecosystems can have a positive or detrimental impact on target plants, microbes, soil, and environment. Based on allelochemicals found in the donor plants, agricultural productivity might be improved by suppressing weed development and safeguarding the crop from disease. On the other hand, allelochemicals could lead to autotoxicity and soil sickness as negative effects [29,34]. Allelochemical activity of phenolic compounds

in plant extracts could be used to control weeds and protect crops [9,10,13,19,24]. A plant with phenolic compositions would have phytotoxic activity toward the environment or other organisms [13]. The results of the current study showed that leaf extracts of *C. erectus* plants are rich in phenolic compounds, which could indicate their allelopathy potential against weeds.

The obtained results indicate that the toxicity of phenolic compounds in *C. erectus* extracts varied with plant parts and concentrations. These results agree with previous literature [9,23,41]. Moreover, the phenolic compounds in *C. erectus* show variation in allelopathic activity toward weeds (*C. murale*, *A. viridius*) and crops (*S. lycopersicum*, *C. sativus*). The phenolic compounds found in the extracts of different parts in *C. erectus*, e.g., gallic acid, ferulic acid, and caffeic acid, are considered safe and natural for the environment [23,38,41]. Research provides evidence that *C. erectus* parts extracts (leaves, stems, fruits, and flowers) growing in Saudi Arabia have antioxidant, anticancer, and antimicrobial properties because of the high phenolic contents [38].

C. murale shoot and root lengths grown in Petri dishes were significantly inhibited by the extracts of *C. erectus* parts (leaves, seeds, and roots) at all concentrations. Likewise, in pots, *Conocarpus erectus* parts extracts at all concentrations inhibited *Chenopodium murale* root lengths significantly (Table 1). Because of their high metabolic rate and the fact that some allelochemicals dissolve in water, roots are thought to be vulnerable to allelochemical activity in soil [42,43]. In agreement with our study, previous studies noted that plant extracts in laboratory conditions caused more inhibition compared to pot experiments [13,44]. However, there was an inhibition by allelopathic activity of phenolic compounds to *C. murale* growth, and it increased with an increase in extract concentration in both Petri dishes and pots. Phenolic allelochemicals inhibit photosynthesis in target plants, reduce chlorophyll content, decrease energy metabolism via affecting cell root permeability, and inhibit cell division and root branching [24,45,46].

In addition, leaves and seeds of *C. erectus* had the highest inhibition effect on *C. murale* growth. This result may be attributed to the higher total phenolic compounds in leaves and seeds compared to roots based on HPLC analysis. Allelochemicals in high concentrations inhibit protein and carbohydrate synthesis, which lead to reduction in plant growth [24].

A. viridius shoot and root lengths were inhibited significantly by *C. erectus* leaf extracts at all concentrations in Petri dishes, and root lengths in pots were inhibited at all concentrations (Table 2). Root length was inhibited in pots significantly compared to shoot length, because roots are more sensitive to allelochemicals [36,47]. In another study, *A. viridius* significantly inhibited the growth of several aromatic plants by allelochemicals [48]. In addition, *A. viridius* showed an inhibition on plant growth either via shoot or root extracts by herbicidal activities of seven allelochemicals, and the inhibition rate varied based on the extract concentrations and phenolic contents in extracts [49]. *A. viridius* inhibited the growth of medicinal and aromatic plants and thus was suggested to be used for controlling weeds [50]. Growth inhibition of seedlings was attributed to changes in enzyme activity and osmotic pressure. Moreover, *A. retroflexus* seeds inhibited seedling growth by phenolic compounds, which affect enzyme activities, photosynthesis, mitosis division, DNA replication, decreasing cell growth, and metabolic energy for respiration [46,47,51].

The inhibition rate of *A. viridius* was also influenced by the part and concentrations of *C. erectus* extracts. The growth of sorghum seedlings was inhibited by the extracts of *A. retroflexus*, and the inhibition was dependent on concentration, part, and growth stage [47]. The results of the current study showed that *C. erectus* leaf extracts had the highest rates of inhibition against *A. viridius* growth, which indicated that leaf extracts have allelopathic activity because of the high content of phenolic compounds (gallic acid, caffeic acid, and ferulic acid). Conversely, root had the lowest effect on *A. viridius* growth, which may refer to the low phenolic content in roots, especially gallic acid and caffeic acid. Furthermore, the phenolic compound concentration to inhibit seed germination should be higher than the concentration to inhibit growth of seedlings [52].

Generally, allelopathic activity from *C. erectus* extracts showed high rates of inhibition toward weed (*C. murale*, *A. viridius*) seedling growth in Petri dishes and pots. The highest extract effect to both was leaves of *C. erectus*. A previous study found that phenolic compounds (gallic acid, caffeic acid, and ferulic acid) inhibit growth of weeds via several physiological effects that reduced growth, such as water stress, suppression of photosynthetic rate, and the hindering of the function of many enzymes [46,47,51–53]. Indeed, the results in bioassays had higher inhibition rates than in the soil. This could be attributed to some of the phenolic compounds being water soluble and that they leached from the root of target plants to soil, which may reduce the inhibition effect [54].

In contrast to weeds, crop plants, i.e., tomato and cucumber, grown in either Petri dishes or pots showed resistance to *C. erectus* extracts. Tomato plants showed low rates of inhibition to no inhibition by all extracts except leaves extracts in Petri dishes at high concentrations. Similarly, cucumber growth was only affected by extracts in Petri dishes at high concentrations. Indeed, tomato and cucumber showed high resistance to phenolic compounds. Weed seeds were more vulnerable to allelopathic compounds than were crops. This vulnerability could be due to the smaller size of seeds in weeds compared to crops. Weed with small seeds are more sensitive to allelochemicals because they have less carbohydrate storage [47,55]. Other research has exposed phenolic compounds to *C. sativus* and *A. palmeri* in bioassays, and their results indicated that small seeds of weeds have the potential to be controlled by allelopathic activity more than big seeds of crops [56]. Moreover, the sensitivity of weeds under examination to phenolic acids might be higher than the sensitivity of studied crops, i.e., tomato and cucumber. The experiments demonstrated that the concentration of extracts and their source had a substantial impact on phytotoxicity. The positive results of this research study may be used to develop eco-friendly herbicides for agricultural purposes.

5. Conclusions

The results obtained in the current study indicated that different extracts of *C. erectus* significantly inhibited the growth of weeds with little or neglectable effects on the growth of cultivated crops, e.g., tomato and cucumber. The highest inhibition of weeds (*C. murale* and *A. viridis*) growth was found following exposure to varied doses of extracts of leaves and seeds. The results of the current study lay the foundation for future studies examining the potential application of *Chenopodium murale* extracts in the biological control of weeds via allelopathic effects. Further research into the large-scale use of these extracts and their impacts on crop development and production is recommended.

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